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The microbiological quality of draught no- and low-alcohol beers

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Abstract

Why was the work done: To determine the microbiological quality of draught no- and low-alcohol beers in the on-trade and to provide insight into the factors that may influence susceptibility to spoilage.
How was the work done: 53 samples comprising of 17 brands, seven different beer styles (lager, India pale ale, pale ale, wheat, radler, pilsner and amber ale), were sampled at least twice in 12 different public houses in Nottingham, UK. Spoilage potential was determined using a forcing test with beers incubated statically at 30°C for 96 hours. Quality was assessed - using four categories - based on the relative increase in absorbance at 660 nm, reflecting the growth of beer spoilage microorganisms present in the beer at dispense. The physiochemical properties of each beer (ethanol, present gravity, pH) were evaluated pre- and post-forcing.
What are the main findings: 54.7% of the draught beers sampled were of ‘unacceptable’ quality due to microbial growth. Additionally, the concentration of ethanol was elevated, with some samples exceeding the limit for no- and low-alcohol beers in the UK. Principal Component Analysis showed that samples ranked as ‘unacceptable’ were positively correlated with a high present gravity and high pH. However, the susceptibility of no- and low-alcohol beers to spoilage is also determined by interacting intrinsic factors (lack of ethanol and carbohydrate profile) and extrinsic influences (hygiene of dispense equipment together with the rate of beer throughput).
Why is the work important: The reduction or elimination of ethanol (an antimicrobial hurdle) in the production of no- and low-alcohol beers results in a product that is more susceptible to spoilage by microorganisms. To ensure product quality and food safety, it is recommended that no- and low-alcohol beers should have a pH < 4.0, contain minimal residual sugar content, and undergo appropriate stabilisation procedures prior to packaging. The production of ethanol by dispense microorganisms presents an additional risk to the no- and low-alcohol category. This should be considered by brewers to ensure that this does not become a compliance issue. Further, particular care should be taken in the hygienic handling of no and low products, with implementation of robust and regular cleaning regimes for conventional long line dispense systems.

Keywords

alcohol-free, low-alcohol, NoLo, NABLAB, draught beer, dispense, microbial spoilage, forcing test.

Introduction

In recent years, the no- and low-alcohol (NoLo) drinks category has seen a rapid expansion, driven by demand from consumers seeking to moderate alcohol consumption (Roselli et al. 2024). No- and low-alcohol beers (see *footnote* below) have been at the forefront of this transition, enabling manufacturers the opportunity to both satisfy market demand and expand the consumer base through product diversification (Bellut and Arendt 2019).

Commensurate with the growth in sales volume, the introduction of draught no- and low-alcohol beers to the on-trade is growing in popularity worldwide. In the UK, draught beer is mostly dispensed using 'long draw' or 'long line' systems. Kegged beer passes through stainless steel coils housed in a remote cooler containing chilled water or glycol. Since UK cellars are typically maintained at 12°C, the purpose of the cooler is to chill the beer to the appropriate serving temperature rather than to deliver microbial stability. From there, the beer is pumped to the tap through dispense tubing (lines) bundled within an insulated 'python', which also contains two larger bore lines circulating chilled water or glycol. Long draw dispense systems are inherently complex and offer multiple sites for microbial contamination and biofilm development, including the keg coupler, fob detector, dispense line, and nozzle. Additional fittings within the system can also create dead spots that are difficult to clean. Some of these issues are mitigated by 'short draw' dispense systems that position kegs in refrigerated cabinets close to the tap and use disposable dispense lines, thereby avoiding some of the microbiological issues associated with long draw systems (Quain 2025). However, such systems are not always practical or incur additional cost, such that they are currently not as prevalent in the UK. Irrespective of the precise mode of delivery, the effects of microbial spoilage in draught beer can range from minor sensorial defects which may not be noticed by the consumer, to perceptible changes in flavour, aroma and appearance. Ineffective hygienic (cleaning) regimes and low throughput further exacerbate variability in draught beer quality, negatively impacting the consumer experience (Quain 2021). Spoilage outcomes reflect both the product composition and the microorganisms able to grow in the beer (Quain and Jevons 2023: Roselli et al. 2024: Britton and Hill 2025). The principal microorganisms that spoil beer include aerotolerant gram-positive bacteria (*Lactobacillus*, *Pediococcus*), aerobic or microaerophilic gram-negative bacteria (*Acetobacter*, *Gluconobacter*), facultatively aerobic yeasts (*Saccharomyces*) and aerobic yeasts (*Brettanomyces*, *Candida*, *Pichia*, *Rhodotorula*) (Rainbow 1981).

Previous studies on the microbial composition of draught beer in the UK on-trade have focused on standard beer types and the detection of known spoilage organisms (Wiles 1950: Hemmons 1954: Hough et al. 1976: Harper et al. 1980: Harper 1981: Casson 1985: Jevons and Quain 2022). While these studies are insightful, the selective microbiological methods implemented seek to identify the presence of key organisms and do not always capture the beer microbiome in its entirety, or the overall effect on beer quality. Furthermore, standard beer types present a microbiologically stable environment: the raw materials used provide antimicrobial compounds (specifically isomerised hop bitter acids) (Sakamoto and Konings 2003), while the fermentation process creates a series of antimicrobial 'hurdles' including low pH, limited nutrient availability, low oxygen, high levels of carbon dioxide, and ethanol (Menz et al. 2011: Vriesekoop et al. 2012). The removal or reduction of ethanol (in the case of NoLo products) significantly alters the environment for microbes, allowing for the survival and proliferation of spoilage microorganisms, including those not typically associated with standard beer (Kurniawan et al. 2021). Consequently, the overall susceptibility of no- and low-alcohol products to microbial spoilage is enhanced (Quain 2021: Roselli et al. 2024: Britton and Hill 2025). To demonstrate the extent to which this occurs, Quain (2021) sought to identify the susceptibility of alcohol-free and low-alcohol beers to microbial spoilage via a series of challenge tests. Microflora isolated from standard draught beers were inoculated into no- and low-alcohol samples, and spoilage was evaluated from the increase in turbidity using a forcing test (Mallett et al. 2018). Results showed that microbial growth was two to five times greater in no- and low- lager type beer compared to standard beers at 4.5% alcohol by volume (ABV) (Quain 2021). Further, the quality of draught beers at the point of sale is

variable ranging from 'excellent' to 'unacceptable' (Mallet and Quain 2019) with the microbial quality of no- and low-alcohol beers unexplored in the on-trade.

Quality management in the on-trade is typically out side the control of the brewer and is delivered by retail staff in the account or third-party cleaning companies. The efficacy of handling and hygienic procedures determine the microbial quality of beer at dispense. With enhanced susceptibility to spoilage, the introduction of no- and low-alcohol beers to long line dispense systems carries a greater threat to product quality. In this study, a broad pool of UK-wide NoLo beers were analysed using a forcing method designed to evaluate the microbial quality of draught no- and low-alcohol beers served in the on-trade. Physiochemical parameters (pH, present gravity, ABV) of beer samples were determined at dispense and post-forcing to identify key intrinsic factors with a causative link to spoilage. This insight will lead to a greater understanding of the risk of microbial spoilage of no- and low-alcohol beverages and the subsequent impact on the quality of products delivered to the consumer via draught dispense systems.

The definition of the no- or low-alcohol (NoLo) beer categories is not universal, and the values for alcohol content vary between Europe, USA and elsewhere (Quain 2021: Okaru and Lachenmeier 2022). In this work, the terminology used refers to the current UK guidance, where no-alcohol (No-) beers are those containing ≤ 0.05% ABV and low-alcohol (Lo-) beers comprise ≤ 1.2% ABV (Gov.UK 2018).

Materials and methods

Beer samples and accounts

The beer brands and on-trade licensed public houses (accounts) used in this study are anonymised. No- and low-alcohol beers (n=20) were purchased between January and April 2025 in Nottingham, UK. In all, 53 samples were purchased, comprising of seven different beer styles, 17 brands and 12 different accounts. All beers are UK-wide keg brands, which are also sold in can or bottle (small pack). The beers sampled were either non-alcoholic or low-alcohol beers with a declared ethanol content ranging from 0.0-0.5% ABV. Beer styles included 'lager' (L), 'India pale ale' (IPA), 'pale ale' (PA), 'pilsner' (P), 'amber ale' (A), 'radler' (R) and 'wheat' (W). Beers within the stout category were not subject to investigation during this study.

Sampling of draught beer

To minimise the effect of uncontrollable variables known to impact draught beer quality (such as regularity/time of line cleaning, and beer throughput), repeat sampling was conducted (Mallett and Quain 2019). Where a single beer was available across different accounts, this enabled the acquisition of 'replicates' through sampling at different locations (Sample Group 1). Some accounts offered a permanent no- or low-alcohol product on draught that did not change over time, allowing for sampling in triplicate (Sample Group 2). Where accounts served a rotating 'guest' draught no- and low-alcohol beer, this occasionally limited sample replicates to only two per beer style (duplicates), depending on turnover rates. However, in such instances, the quality of a single keg could be tracked over time by collecting samples from the same account within a one week window (Sample Group 3). Finally, data from each of these subsets were combined to provide a broad and comprehensive summary of the microbiological quality of different draught no- and low-alcohol products across different accounts over a period of time.

In all instances, sampling was performed between 13:00 and 17:00 hours. Draught beers were purchased from each account, and 200 mL aliquots sampled directly into sterile Duran bottles (250 mL). Samples were kept cold in transit and refrigerated overnight at 4°C prior to analysis. All analyses post sampling were performed aseptically.

Forcing test

The microbiological loading of draught beer samples was evaluated using the forcing test of Mallett et al (2018). Triplicate 25 mL samples were incubated statically in 30 mL plastic Universal bottles (with loosened cap to enable gas exchange) at 30°C for 96 hours. Prior to forcing, each sample was thoroughly mixed by inversion, and the turbidity assessed by measurement of absorbance at 660 nm (Jenway 7315 spectrophotometer). At the end of incubation ('forcing'), the increase in absorbance (ΔA_{660}) was determined. Hazy, or

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heavily spoiled beers were diluted (1:1 or 1:3 as required) with sterile water prior to measurement.

Quality assessment and ranking

The increase in absorbance over time (ΔA_{660}) reflects the degree of microbiological spoilage of the beer samples (Mallett et al. 2018). Beers were categorised as 'excellent/A' (ΔA_{660} 0-0.3), 'acceptable/B' (ΔA_{660} 0.31- 0.6), 'poor/C' (ΔA_{660} 0.61-0.9) and 'unacceptable/D' (ΔA_{660} > 0.91), according to established quality bands (Mallett et al. 2018).

For groups of samples (e.g. sampling the same and/or different beers from the same account) a cumulative 'quality index' was calculated using the sum of individual quality bands (where A = 4, B = 3, C = 2, D= 1) in relation to the number of samples (n), as previously described (Mallett et al. 2018: Mallett and Quain 2019). For example, where all samples were ranked as 'excellent', the quality index would be 100%, whereas in a scenario where all samples were ranked as 'acceptable', the index would be 75%.

$$Quality\ index = \frac{\Sigma\ Individual\ quality\ bands}{n \times 4} \times 100$$

Beer analyses

Draught and forced beer samples were analysed for alcohol content and present gravity via density analysis (DMA 4500 M, Anton Paar), and for pH (Mettler Toledo SevenEasy pH meter). Prior to analysis, samples were degassed by centrifugation at 3,220 x g for 20 mins at 4°C. Samples were then allowed to equilibrate to room temperature.

For comparative purposes, the total carbohydrate and fermentable sugar content of the beers was obtained from labelling, the manufacturer's website, or via direct enquiry with the brewing company. Similarly, where stated, information regarding the production technique was obtained through communication with each brewery. Most of the beers analysed (16 out of 20) were labelled as low alcohol (0.5% ABV) and within the criteria recommended for assessing ABV in low alcohol products using NIR-based methods (Toivola et al. 2005). However, some values were below 0.37% ABV which requires the use of gas chromatography with flame ionisation detection for the accurate analysis of ethanol. Consequently, below 0.37% ABV, the Anton Paar/NIR system lacks the required accuracy, repeatability and reproducibility and should be treated with caution (Toivola et al. 2005).

Statistics

Data visualisation and statistical analysis was performed in XLSTAT (Version 2024.3.0) and GraphPad Prism (Version 10.6.1). A Wilcoxon signed-rank test was performed to determine whether increases in absorbance (ΔA_{660}) pre and post forcing were statistically significant. To examine relationships between the physiochemical characteristics of no- and low-alcohol beer samples and their spoilage potential, Principal Component Analysis (PCA) was performed in XLSTAT. Differences in physiochemical properties (ABV, pH and present gravity) were evaluated using separate mixed-effects models for each variable, comparing average values of pre- and post-forcing groups. Where significant effects ($p < 0.05$) were detected, multiple comparisons were performed using Bonferroni's correction.

Results and discussion

Microbial quality of draught no- and low-alcohol beers

To understand the microbial spoilage of draught no- and low-alcohol beer, 20 different NoLo brands were evaluated using a forcing test. Analysis of data from the entire sample set indicated that a statistically significant increase in absorbance (ΔA_{660}) pre- and post-forcing was observed ($p < 0.0001$), indicating that microbial stability was compromised to some extent. Individual product data was then analysed and used to assign each beer a quality category band based on the change in absorbance at A_{660}. Based on this analysis, the predominant quality category band for draught NoLo beer samples was 'unacceptable' (54.7%) followed by 'acceptable' (22.6%), 'poor' (17.0%) and, only rarely, 'excellent' (5.7%) (Figure 1A). Furthermore, visual indicators of microbial spoilage including sedimentation, haze, and pellicle formation were frequently observed, most obviously in 'unacceptable' but also in 'poor' beer samples. Based on the data presented here, no- and low-alcohol draught beers were especially vulnerable to spoilage. This was evident in light of a previous study investigating the microbial quality

of standard draught beers. Using the same methodology and classification, Mallett and Quain (2019) reported that standard draught beers (ABV ≤ 4.2%) were predominantly either 'acceptable or excellent' (cumulatively 77.9 for ale and 72.7% or lager), compared to 28.3% in the present work.

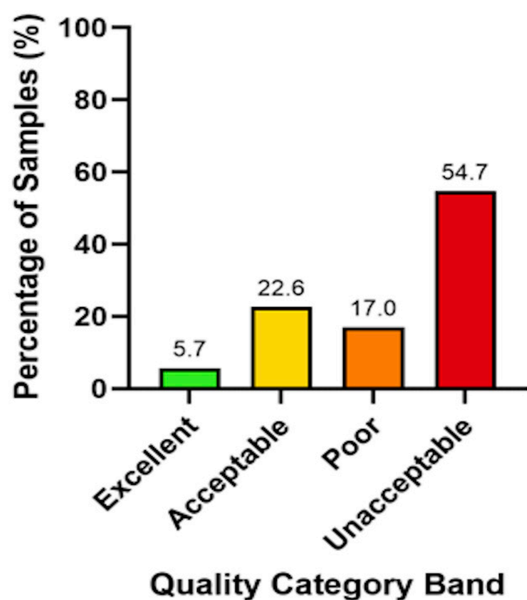

Figure 1A. Quality of draught no- and low-alcohol beers. Distribution of quality bands for 53 beers based on changes in absorbance (ΔA_{660}) after forcing.

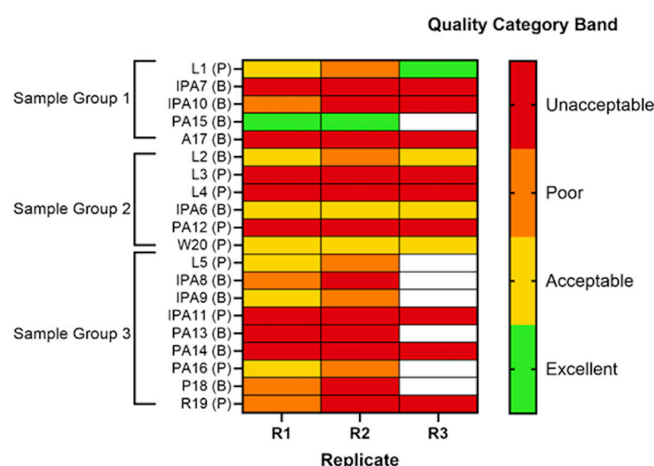

Figure 1B. Quality of draught no- and low-alcohol beers. Heat map showing quality bands of Sample Groups. Columns R1-R3 show replicate samples collected at consecutive timepoints. Where R3 boxes are white, there are only two replicates. The method by which each NoLo beer sample was produced is indicated in the y-axis using the suffix 'B' (biological) or 'P' (physical), Brackets on the y-axis indicate sample groups: 'Group 1' denotes brands sampled from different accounts, 'Group 2' includes brands from the same account at different time points and 'Group 3' refers to samples taken from the same keg over time.

Analysis of brands available in multiple accounts (Sample Group 1) showed that the quality of most samples remained broadly consistent (Figure 1B). For example, brand PA15 was 'excellent' at both accounts, while IPA7 and A17 were consistently 'unacceptable'. With brand L1, quality was variable ranging from 'poor' to 'excellent'. Where single accounts offered a permanent no- and low-alcohol brand (Sample Group 2), the quality of products was typically consistent between samples and ranged from acceptable (L2, IPA6 and W20) to unacceptable (L3, L4 and PA12). Finally, in those instances where it was possible to sample the same keg over a week (Sample Group 3), a decline in quality was typically observed over time (L5, IPA8, IPA9, PA16, P18 and R19), with later samples (indicated by R2 and R3) typically falling into lower quality bands compared to the initial sample (R1). For samples IPA11, PA13 and PA14, the quality was unacceptable at the initial sampling (R1), and this remained consistent across subsequent replicates (Figure 1B).

To further understand the microbiological quality of draught no- and low-alcohol beers from individual accounts, the cumulative 'quality index' was calculated on a scale of 0-100%, with higher values equating to improved microbial stability. Based on this, the quality index for beers sampled from different accounts was observed to vary widely (Figure 2). Analysis indicated that seven accounts dispensed beer with a quality index (QI) of 25-50%, corresponding to mostly unacceptable or poor beers. The remaining five accounts offered draught beer with a QI ranging from 50-75% (ranging from poor to acceptable), and no accounts in the survey had a QI greater than 75% (predominately acceptable or excellent beers) (Figure 2).

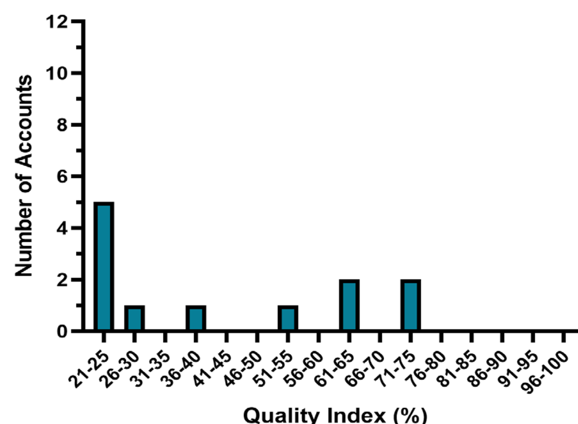

Figure 2. 'Quality index (%) of on-trade accounts. 20 different no- and low-alcohol draught beers from 12 different accounts. The number of samples from individual accounts ranged from two to 14.

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It should be noted that the intent of this work was not to specifically highlight the performance of individual accounts or the microbiological quality of individual brands *per se*, but to understand the challenges that the sector faces with respect to draught NoLo beer. The data presented here clearly demonstrates that the microbiological instability of no- and low-alcohol beers on draught is a risk to quality, irrespective of beer style, and represents a significant concern for the industry.

Table 1. Draught no- and low-alcohol beers used in this work.

Beer	Account	Beer style	ABV (%)	Production method	Notes[†]	Carbohydrates (sugars) (g/L)
L1	5, 5, 6		0.5	Physical	Unfiltered	35 (< 1)
L2	3, 3, 3		0.5	Biological[†]	Pasteurised	26 (20)
L3	12, 12, 12	*Lager*	0	Physical	NA	47 (17)
L4	11, 11, 11		0	Physical	NA	48 (13)
L5	4, 4		0.5	Physical	Unpasteurised	43 (5)
IPA6	3, 3, 3		0.5	Biological[†]	Pasteurised	30 (18)
IPA7	2, 2, 10		0.5	Biological	Pasteurised	39 (18)
IPA8	7, 7	*India Pale Ale*	0.5	Biological	Unpasteurised, Preservatives used	43 (1)
IPA9	4, 4		0.5	Biological	Unpasteurised	8 (1)
IPA10	4, 4, 6		0.5	Biological	Unpasteurised, Preservatives used	NA
IPA11	7, 7, 7	*West Coast IPA*	0.5	Physical	Unpasteurised	35 (<5)
PA12	9, 9, 9		0.5	Physical	Pasteurised, Preservatives used	27 (1)
PA13	1, 1		0.5	Biological[†]	NA	NA
PA14	1, 1, 1	*Pale Ale*	0.5	Biological[†]	Pasteurised	NA
PA15	4, 1		0.3	Biological	Unpasteurised	49(<5)[†]
PA16	4, 4		0.5	Physical[†]	Unpasteurised, Preservatives used	38 (2)
A17	7, 7, 10	*Amber Ale*	0.5	Biological[†]	Unpasteurised	NA
P18	4, 4	*West Coast Pilsner*	0.5	Biological[†]	Unpasteurised	NA
R19	4, 4, 4	*Radler*	0.5	Physical	Unpasteurised	43 (5)
W20	8, 8, 8	*Wheat*	0	Physical	NA	34 (0)

Production method, ABV, and concentration of carbohydrates/sugars was obtained from product labels, brand website or [†]from direct enquiry.
Carbohydrates = maltotriose/dextrins DP>3;; sugars = fermentable sugars (glucose, maltose, fructose, sucrose).
N/A = not available.

Production method and the microbial quality of no- and low-alcohol beers

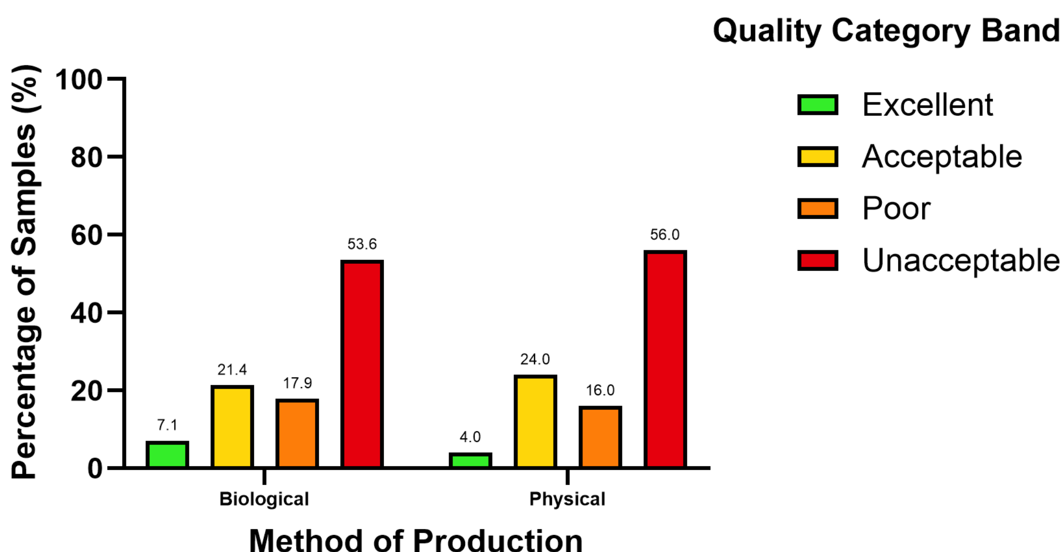
No- and low-alcohol beers are produced using biological and/or physical methods and then packaged into kegs that may or may not be pasteurised, filtered or treated with preservatives prior to sale. Biological production processes rely on limiting ethanol formation during fermentation, whereas physical techniques remove ethanol from standard beer using vacuum distillation, dialysis or reverse osmosis (Brányik et al. 2012). Previous work has suggested that production methods yielding beers with higher residual extract (arrested/limited fermentation), may increase the susceptibility of the product to microbial spoilage (Quain 2021). To investigate this, the data presented above was further analysed with respect to individual production methods. More than half of the beers were produced using biological methods (n=28; Table 1) and, after forcing, the distribution of the quality bands was 'unacceptable' (53.6%), 'acceptable' (21.4%), 'poor' (17.9%) and 'excellent' (7.1%). For those beers produced using physical methods (n=25; Table 1), the distribution of results was similar with 56% 'unacceptable', 24% 'acceptable', 16% 'poor' and 4% 'excellent' (Figure 3). Consequently, it can be seen that the method of production (biological or physical dealcoholisation) did not impact the microbial stability of the beer.

The idea that NoLo beers produced by arrested fermentation result in elevated concentrations of residual sugars compared to those using physical dealcoholisation methods is not necessarily the case. Several of the NoLo beers produced using biological methods (L2, IPA6 and IPA9; considered to be of 'acceptable' quality) had a present gravity that was lower than those beers produced using physical methods. Furthermore, supplementation of NoLo products with glucose or fructose syrups is used as a strategy to improve flavour and body (Brányik et al. 2012), a practice that may inadvertently lead to an increased risk of spoilage in draught dispense.

The microbial stabilisation of no- and low-alcohol beers, involves methods such as sterile filtration, pasteurisation (flash or tunnel), and, occasionally, the use of preservatives or antimicrobial agents (Roselli et al. 2024: Britton and Hill 2025). Although, physical treatments of beer for packaged beer would not provide any 'protection' to the spoilage of draught beer, anti-microbial preservatives may protect against groups of microorganisms.

To evaluate the relative spoilage potential of no- and low-alcohol beers and microbial preservation, the quality bands of products within each of these categories were compared. Pasteurised products did not exhibit greater microbial stability than unpasteurised samples. Three pasteurised products (Table 1; IPA7, PA12 and PA14) were universally 'unacceptable' (Figure 1B), while two (Table 1; L2 and IPA6) were placed in the 'acceptable' category

Figure 3. Production method and quality category bands of draught beers.



Similarly, the addition of anti-microbial preservatives such as sodium benzoate, sodium metabisulphite and potassium sorbate (Table 1: IPA8, IPA10, PA12 and PA16) did not offer enhanced spoilage protection (Figure 1B). To emphasise this point, products without any form of microbiological stabilisation (Table 1) included the beer with the best microbial stability (PA15), together with two that were classified as 'unacceptable' (IPA11 and A17).

Although the focus of the work reported here was draught beer quality, a subset of no- and low-alcohol beers in small-pack were assessed for spoilage using the forcing technique. In all cases, (Figure 4) beer quality was 'excellent', demonstrating the importance of hygiene management in the draught dispense of no- and low-alcohol beers.

Impact of physiochemical properties on the spoilage potential of no- and low-alcohol beers

The microbial quality of draught NoLo beers is principally determined by hygienic management at the point of sale. However, there may be additional interacting factors, including the intrinsic characteristics of beer and final product composition. To gain insight into the relationship between beer properties and spoilage potential, the physiochemical properties of no- and low-alcohol beer samples were recorded at the point of dispense (Table 2) and used to identify causative trends related to spoilage through multivariate statistical analysis. Principal Component Analysis (PCA) was conducted to determine the relationship between residual sugar content (present gravity), pH, ABV and microbial growth (ΔA_{660}).

Table 2. Physiochemical characteristics of the draught beers.

Sample	pH (pre-forcing)	pH (forced)	ABV (%) (pre-forcing)	ABV (%) (forced)	PG (g/L) (pre-forcing)	PG (g/L) (forced)
L1	4.3	4.2	0.45	0.47	1.0155	1.0149
L2	3.93	3.78	0.16	0	1.0097	1.0099
L3	4.02	3.37	0.02	0.99	1.0176	1.0079
L4	4.34	3.36	0.02	0.4	1.0185	1.0139
L5	4.23	3.88	0.35	0.27	1.011	1.0104
IPA6	3.89	3.43	0.33	0.01	1.0084	1.0091
IPA7	4.23	3.6	0.36	1.74	1.0148	1.0028
IPA8	4.37	3.41	0.69	0.22	1.0188	1.018
IPA9	4.32	3.98	0.57	0.59	1.0088	1.0082
IPA10	4.28	3.65	0.35	1.17	1.0137	1.0061
IPA11	4.33	3.8	0.4	0.35	1.0181	1.0166
PA12	4.14	3.73	0.35	0	1.0116	1.0116
PA13	4.31	3.07	0.71	0.86	1.0219	1.0168
PA14	4.25	3.58	0.31	1.16	1.0135	1.0063
PA15	3.95	3.95	0.3	0.28	1.0189	1.019
PA16	4.22	3.83	0.28	0.24	1.0141	1.0132
A17	4.3	3.44	0.78	0.34	1.0121	1.0119
P18	4.43	3.75	0.93	0.47	1.0048	1.0053
R19	3.67	3.57	0.36	0.07	1.0264	1.0257
W20	4.4	4.34	0.01	0.07	1.0139	1.0133

Data is the mean value for each sample group (as detailed in Table 1).

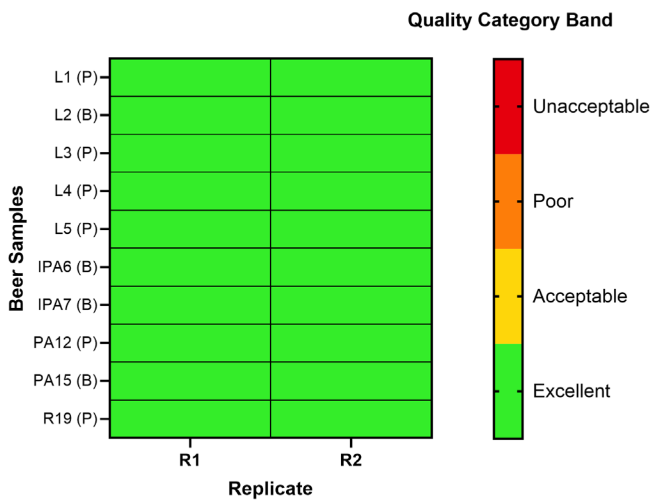
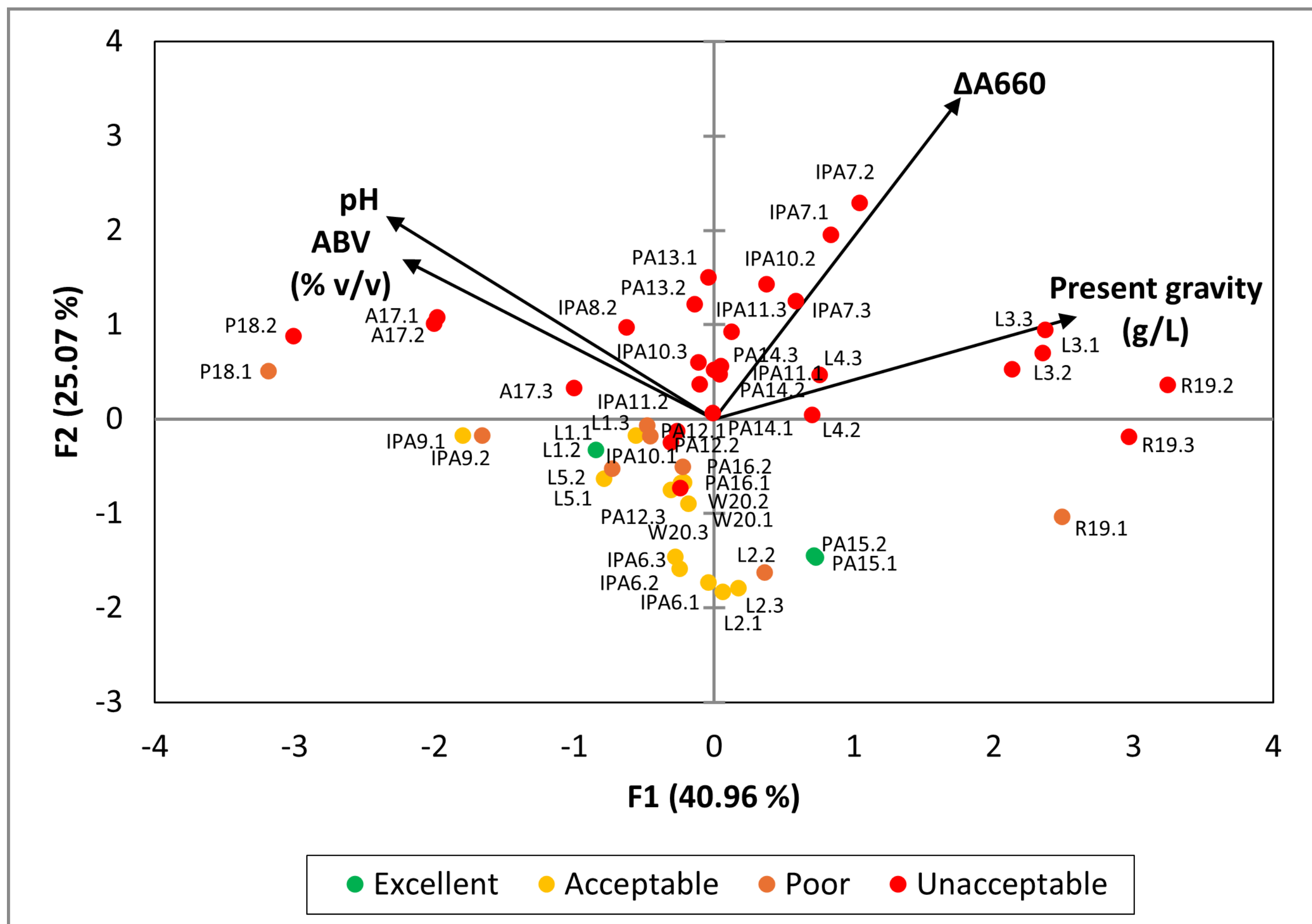


Figure 4. Heat map of quality category bands of small pack forced beers. Columns R1-R2 denote replicate samples collected from separate cans. The method by which each NoLo beer sample was produced (biological or physical) is denoted in the y axis using the suffix 'B' or 'P', respectively.

Figure 5 shows that the principal component 1 (F1) had a strong positive correlation with present gravity (0.737) and was negatively correlated with pH (-0.666) and ABV (-0.633). Principal component 2 (F2) yielded a strong positive correlation with ΔA_{660} (0.758), allowing samples of 'unacceptable' quality to be clearly grouped together. In contrast, all samples that exhibited negative F2 values included those categorised as being of 'acceptable' or 'excellent' quality. In essence, these data indicate that conditions incorporating increased gravity, pH and ABV are all conducive to enhanced spoilage. This interpretation supports previous data with respect to gravity and pH; it is accepted that residual sugars encourage cellular proliferation through provision of assimilable carbohydrate, while low pH can negatively impact microbial growth via cytoplasmic acidification and the collapse of proton gradients (Lund et al. 2020).

Figure 5: Principal Component Analysis (PCA) of the spoilage potential (ΔA_{660}) of draught no- and low-alcohol beer in relation to ABV (%), pH and present gravity (g/L). Individual PC scores represent samples, coloured to reflect their assigned quality band. Arrows indicate variable loadings showing the contribution of each property towards the principal components. F1 and F2 explain 66.03% of the total variance (40.96% and 25.07%, respectively).



The observation that an increase in ABV appeared to encourage spoilage was perhaps surprising, but it should be noted that 'high' ABV values were less than 1% (i.e. unlikely to be toxic). Furthermore, as discussed below, it is likely that much of the ethanol present was produced through microbial growth and is therefore an artefact, rather than a cause, of spoilage potential. This suggestion is supported by analysis of samples A17 and P18, which were predominately categorised as being of 'unacceptable' quality. In these instances, elevated levels of alcohol (> 0.5% ABV) were detected at the point of dispense, indicating that fermentation, because of spoilage, had likely occurred in-keg or in-line, and before forcing of the samples.

The relationship between factors is inevitably complex. For example, samples with a low sugar content (e.g. P18) were not always resistant to microbial growth, while those with a high sugar content (e.g. PA15) did not exhibit enhanced spoilage potential. In the case of PA15, the high sugar content may have been offset by the low pH (positive F1, average pH=3.95), suggesting that for this product, pH may have acted as a factor to suppress spoilage; an hypothesis supported by the absence of changes to ethanol concentration or present gravity, suggesting no secondary fermentation had occurred (Table 2).

Physiochemical changes occurring because of microbial spoilage

Given that microorganisms grew in the majority of beers analysed, changes in fundamental beer properties - ABV, present gravity and pH - over time (pre- and post-forcing) were determined. Of particular note, were the physiochemical changes occurring during forcing with a statistically significant increase in ethanol concentration in five of the samples analysed (L3, L4, IPA7, IPA10 and PA14; Figure 6A), accompanied by a corresponding decrease in present gravity (Figure 6B). These data demonstrate fermentation can occur in draught NoLo beers, resulting in final ABV values that, in this study, ranged from 0.40-1.74% (Table 2).

Importantly, an analysis of the ethanol concentrations in beers pre-forcing, was often higher than the reported ABV for the product. For example, IPA8, IPA9, PA13, A17 and P18, had initial

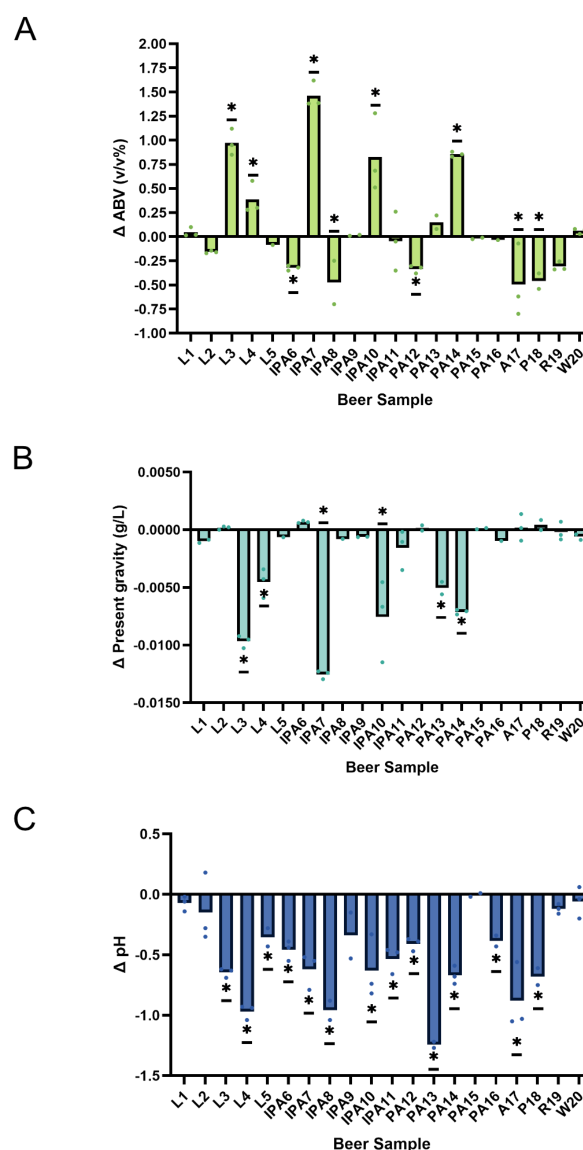


Figure 6. Physiochemical characteristics of draught no-and low-alcohol beers at the point of dispense and post forcing. (A) changes to alcohol by volume (% ABV); (B) changes in pH; and (C) changes in present gravity (g/L). For each parameter, differences between the average values of pre- and post-forcing groups were evaluated using separate mixed-effects models for each variable. Where significant effects (* $p < 0.05$) were detected, multiple comparisons were performed using Bonferroni's correction.

pre-forcing) ABV values of 0.69, 0.57, 0.71, 0.78 and 0.93%, some 0.07-0.43% higher than the declared ABV (Table 2). It is noteworthy, that except for PA13 (whose status was unknown), these beers were all unpasteurised, suggesting the importance of stabilisation methods within the brewery. This data also further highlights the significance of spoilage in keg, in-line or at the point of dispense, potentially causing alcohol levels to be elevated beyond the current limits specified

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for no- and low-alcohol beers in the UK. Although. beyond the scope of this study, additional factors influencing the extent of ethanol formation are likely to include the freshness and/or age of the beer, storage temperature and handling procedures prior to and including dispense, and the throughput and rate at which the beer was sold once the keg has been broached.

An increase in ethanol was not observed in all samples. Some samples (IPA6, IPA8, PA12, A17 and P18) showed a significant decrease in ethanol content (Figure 6A). A possible explanation for this is the presence of ethanol-tolerant microorganisms including yeasts and bacteria that are able to metabolise ethanol as a carbon source (Ingledew 1979). Notably, acetic acid bacteria including *Gluconobacter* and *Acetobacter* which are ubiquitous in draught beers (Jevons and Quain 2022). This is supported by the observation that samples exhibited a decrease in pH after forcing, albeit to varying degrees (Figure 6C). It is likely that the growth of lactic acid and acetic acid bacteria (resulting in the synthesis of lactic and/or acetic acids respectively) contribute to the acidification of the spoilt beer (Menz et al. 2010: De Roos and De Vuyst 2019).

Conclusions

The work reported here considers the microbial stability and product quality of draught NoLo beers, whilst exploring the impact of additional factors on spoilage potential. This involved 'forcing' to provide a retrospective analysis of microbial loading of spoilage microorganisms in draught no- and low-alcohol beer at the point of dispense. The data clearly indicates that no- and low-alcohol beers were highly susceptible to spoilage, with most products being compromised to varying extents. Attempts to identify extrinsic aspects of NoLo beer (i.e. those related to production) that could be linked to spoilage potential were unsuccessful, with no clear links to production strategy (biological production or physical dealcoholisation), the application of preservatives, or in-pack preservation (pasteurisation/filtration). This suggests that the microbial quality of draught NoLo beer is dictated by the nutritional and physiochemical properties of the beer, together with the efficacy and frequency of hygienic practices used across draught dispense.

Analysis of the relationship between intrinsic factors using PCA indicated that those products with a lower pH and minimal residual sugars were more resistant to spoilage. Although this was not entirely unexpected, the data provides further support to suggest that these attributes should be key considerations for ensuring the microbial stability of beer. Furthermore, preservatives were found to be ineffective, which is likely to be related to beer pH, since more 'alkaline' conditions (pH > 4.0) reduce the efficacy of weak organic acids used as preservatives. These conclusions support recent data suggesting that low pH (in conjunction with elevated carbonation and low oxygen) can restrict the growth of a range of microorganisms in NoLo beers, including common food borne pathogens such as *E. coli* O157 and *Salmonella* (Rachon et al. 2024). However, our analyses also highlight the multifactorial nature of microbial instability in no- and low-alcohol beers, with potential microbial spoilage arising from complex interactions between intrinsic factors. Although elevated concentrations of fermentable sugars logically contribute to increased spoilage risk, other parameters including the availability of nutrients (nitrogen, sulphur, mineral ions and vitamins), pH, levels of carbonation, and, possibly, antimicrobial compounds (including exogenous preservatives) are likely to collectively shape the spoilage potential of the final product.

It is acknowledged that in this work, product 'quality' is based on the development of biological haze over time, rather than full characterisation of beer properties. Importantly, analysis of alcohol concentration showed that ethanol levels were frequently found to be elevated, exceeding that currently imposed in the UK for no- and low-alcohol beers. This emphasises that the impact of microbial spoilage extends beyond sensory concerns, with broader implications for the sector in general. Beyond the immediate compliance and legislative consequences, the presence of elevated levels of ethanol in non-alcoholic beverages may pose a risk or concern to consumers who may be avoiding alcohol for physiological, medical or religious reasons.

The data reported here shows that hygienic handling of draught beer is of primary importance in ensuring the microbial stability of NoLo products. Rapid throughput, efficient cooling, and robust

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cleaning of lines, tap nozzles and keg couplers are effective preventative strategies that should be routinely implemented in the on trade. However, continual developments in alternative strategies around hygienically designed stand-alone (countertop) devices or compact short-draw dispense systems may prove increasingly effective in reducing complexity and lowering the risk of microbial spoilage in no- and low-alcohol beverages (Quain 2021). However, one of the key challenges is to ensure that the link between hygienic best practice and product quality is recognised by all those involved in the dispense of draught beer. Effective and regular education in understanding and encouraging good dispense hygiene has beneficial consequences irrespective of product type, enhancing product and brand reputation, and ensuring a positive consumer experience.

Author contributions

Giulia Roselli: Conceptualisation, investigation, methodology, writing (original draft).
Matthew Crow: Project administration, resources, supervision.
Katherine Smart: Resources, supervision, writing (review and editing).
Chris Powell: Conceptualisation, supervision, project administration, funding acquisition, writing (review and editing).

Conflict of interest

The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

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