

Measurement and Characterization of Bubble Nucleation in Beer

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ABSTRACT: A new method has been developed for characterizing the rate of bubble formation in beer. The method consists of forming bubbles in beer in a standardized glass using a controlled source of nucleation, then photographing the glass. The extent of bubbling is compared with that displayed in a set of reference standards to give a measure of the rate of nucleation. Comparisons were made among commercial beers to determine which physicochemical factors influence nucleation, and to what extent. Surface tension, density, and carbon dioxide content all influence the appearance of bubbles in the glass.

Keywords: nucleation, bubble, beer, carbon dioxide, method

Introduction

THE APPEARANCE OF BEER HAS BEEN SHOWN to influence the consumer's perception of overall quality, with the head of foam having arguably the largest impact of all visual cues. Bamforth (2000) showed that simple visual assessment of a beer's foaming characteristics influenced a consumer's perception of how it would taste, influencing such factors as perceived level of carbonation, temperature, and other flavor aspects. Ono and others (1983) presented tasting data showing that beers with different heads of foam did indeed perform differently in sensory evaluation tests. It appears therefore that foam can have both a direct and an indirect impact on beer flavor.

While much is known about the factors that stabilize bubble foam once formed (Bamforth, 1999), less attention has been paid to the formation of foam in the first place. Carbon dioxide release through nucleation adds new bubbles to a foam and the bubbling itself (known as "beading") may play a role in sensory perception. A beer showing little bubbling, or nucleation activity, in the glass may be perceived as flat, even if its CO₂ content is normal. In addition, active bubbling replenishes the head. The amount of nucleation activity displayed is impacted greatly by the physical container itself, and purveyors have even introduced scratches onto their glasses to promote a lively display of bubbling and good head retention (Parish, 1997).

Under the conditions found in beer, bubble nucleation occurs only at a nucleation site, where an existing bubble gives rise to a new one. Ward and others (1983) describe the geometry and growth of a bubble in a conical pit, a convenient geometry that has been used by many subsequent research-

ers. Figure 1 and 2 depict the common understanding of the behavior of conical pit nucleation sites in a variety of situations. Bisperink and Prins (1994), and later Prins and van Marle (1999) contributed additional findings based on geometry and mass transport, and their organization of the information for beer-based systems serves as a starting point for many calculations used in the present work.

Several methods have been developed to characterize the frequency of bubble formation in supersaturated liquids, but none is completely satisfactory. The simple technique of counting the number of bubbles seen by eye as they are formed over a given period of time was used by Casey (1988) in sparkling wine and by Shafer and Zare (1991) in beer. However, this method is limited by the human ability to count accurately at high frequency. Lubetkin and Blackwell (1988) constructed a microphone that recorded the sound of bubbles rupturing at the surface of the liquid. A computer was then configured to find the frequency of bubble formation using the sound data. The method works well for supersaturated water, but problems are encountered in beer systems because of its foam-forming nature. Liger-Belair and others (1999) developed a method for sparkling wine using a stroboscope. The frequency of the flashing is adjusted until the bubbles coming from a single nucleation site appear to stand still. That frequency corresponds to the frequency of bubble formation at that site. However, the method cannot be used to study numerous sites at once, since each one displays a different frequency of bubble formation.

The starting point for the present work, therefore, was to develop a simple yet reliable technique for assessing nucleation and

then to use that method to investigate the principal parameters that influence beading in beer.

Materials and Methods

Sample source and climate conditions

A range of commercial beers was purchased from local stores in the form of 12 ounce (355 mL) bottles, stored at room temperature, and used within 1 to 2 wk of purchase. All experiments were performed at 24 ± 2 °C and at ambient pressure unless otherwise indicated.

Measurement of nucleation rate

A 16 oz. (473 mL) (CHECK MATH) beer glass, with straight, angled sides (the "standard" US pint glass) was used. The glass was clean and scratch-free. Since beer will not spontaneously form bubbles in this situation, a source of nucleation sites was introduced in the form of a spherical, sintered glass bead, 5 mm in dia. Beer was introduced into the glass with minimum bubbling by tilting the glass and gently pouring the beer down the length of the side.

A set of reference standards was developed to occupy an arbitrary scale that described the number of bubbles present in a glass on a scale of 0 to 10. Using a Sony Mavica MVC-FD91 Digital Still Camera, pictures were taken of beer in the standardized glass displaying different levels of nucleation activity and a scale (a_n) was developed based on the numbers of bubbles in the photo. A value of 0 was assigned to a glass showing no bubbles, with a value of 10 representing the maximum number of bubbles that could be produced. Care was taken to ensure linearity in the scale, i.e. a beer glass

rated as 8 would contain 80% of the number of bubbles in a glass rated as 10. The rating of 10 was actually eclipsed in later experiments, a difficulty that was addressed by extending the scale to include ratings of 11 and 12. An example of a reference standard is shown in Figure 3.

The use of pressurized containers

With the aim of eliminating incidental nucleation sites, some studies were performed in pressure vessels, namely 20-liter steel containers rated to 100 psig. The containers were connected to a CO₂ cylinder and the pressure held constant using a regulator. We are confident that any container of a size capable of holding sufficient beer glasses and of maintaining such a high internal pressure would be suitable.

In a typical experiment, a series of glasses, each filled with 12 ounces (355 mL) of beer, were placed into the pressure vessel. They were each covered with a piece of paper to diminish airborne contamination while still allowing the free flow of gas. Using pure CO₂, the vessel was then purged three times (through pressurization and release) to ensure a headspace nearly free of air. After a final pressuriza-

tion, the glasses were left to equilibrate for at least three days. Upon removal, the glasses were typically free of nucleation activity until the controlled nucleation site was introduced.

Measurement of mass loss

Another proposed method for quantifying the degree of nucleation activity involved assessment of gas loss using a digital balance to record the change in mass of a glass of beer over time. The standardized glass was placed on a digital balance, capacity > 800 g, reading to 0.01 g. The balance was then tared, and the weight was recorded in 1-min intervals for the duration of the experiment (typically 2 h).

Measurement of CO₂ content

The content of CO₂ in bottled beer was assessed according to the method of the American Society of Brewing Chemists (1992). The piercing apparatus was a Zahm New Style Air Tester (Zahm & Nagel Co., Inc., Buffalo, N.Y., U.S.A.).

Measurement of interfacial surface tension

Static interfacial surface tension of beer against air (g) was measured via the Wil-

helmy Plate method, using a Krüss K 10 ST Digital Tensiometer (Krüss USA, Nazareth, Pa., U.S.A.) according to the manufacturer's instructions. Prior to testing, each sample of beer (355 mL) was degassed by being poured gently into a 500 mL beaker and left for 24 h in contact with a Teflon magnetic stir bar (without agitation). The stir bar provided a sufficient nucleation site for the beer to become degassed within 24 h, but without generating too much foam. A fermentation lock allowed CO₂ to escape while protecting against airborne microbial contamination.

To prepare the samples for testing, the beakers were placed on a magnetic stirrer and agitated to ensure a homogenous mixture. A 60 mL plastic syringe was used to draw a sample from the middle of the beaker. Prior to being carefully injected into the sampling vessels, some samples were filtered through a Fisherbrand G8 glass fiber filter circle and a Millipore type HA 0.45 mm filter.

Readings typically started to fall soon after the start of measurement, possibly due to time-dependent adsorption of surfactants onto the interface at the plate. Since the surface tension measurement is supposed to quantify the amount of buoyancy experienced by the bubble, and since bubbles *in situ* form quickly with little time for adsorption, the highest initial number was taken as the most meaningful reading.

Measurement of density

Density was measured using a Mettler/Parr digital densitometer, model DMA 46

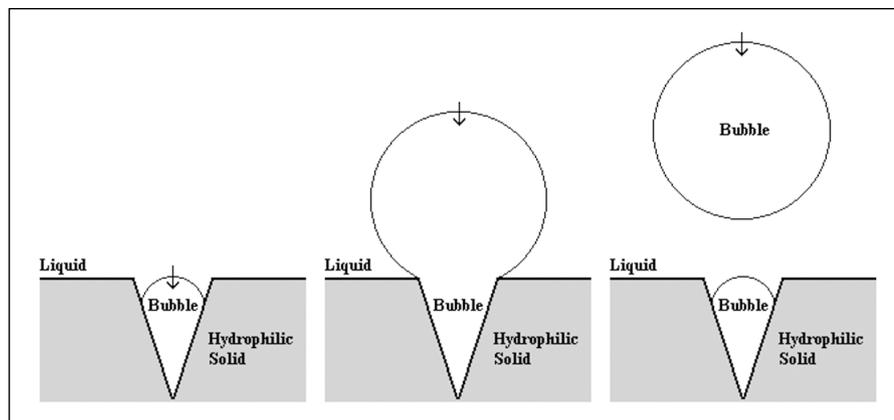


Figure 1—Bubble growth and detachment in a supersaturated liquid. The solid is wet by the liquid. Arrows indicate the direction of gas diffusion due to pressure gradient. From Prins and van Marle (1999).

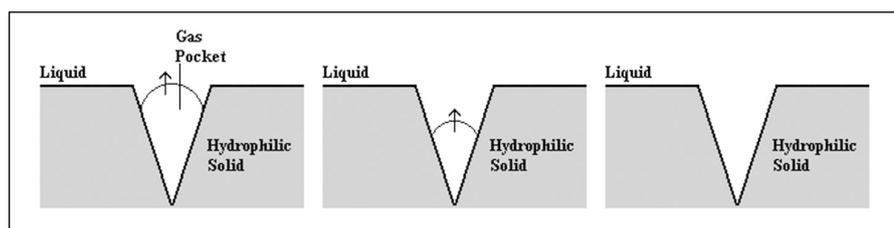


Figure 2—Nucleation site in a system at equilibrium, such as a sealed bottle or pressure container. Arrows indicate the direction of gas diffusion due to pressure gradient. When the site is on a hydrophilic solid, the higher Laplace pressure inside the gas pocket drives the gas into the liquid. Eventually the gas pocket disappears. From Prins and van Marle (1999).

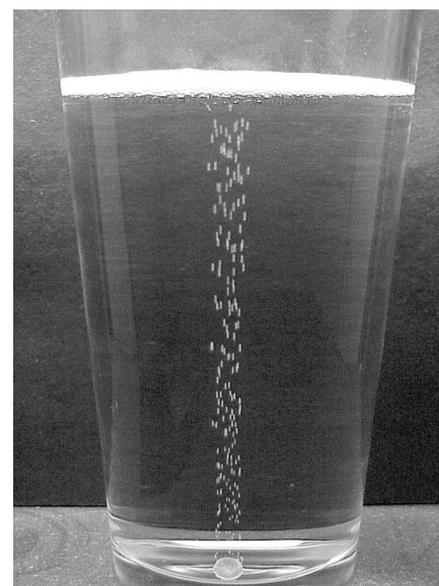


Figure 3—Example of reference standard. This picture represents a nucleation activity of 7 on the 12-point scale.

(Paar USA, Warminster, Pa., U.S.A.). Prior to analysis, samples were degassed and filtered through a Fisherbrand G8 glass fiber filter circle and a Millipore type HA 0.45 mm filter.

Measurement of pH

This was with an Orion Research Expandable ionAnalyzer, model EA 920, fitted with an Orion pH probe, model 915500. The device was calibrated using pH 4.00 and 7.00 buffers. Samples were not degassed prior to measurement.

Adjustment of beer composition

Dilution of beer was achieved by the addition of deionized water. Increases in alcohol content were achieved by the addition of absolute, non-denatured ethanol. Where indicated, sucrose was added in the form of granulated table sugar.

Statistical manipulation of the data

The data were analyzed using JMP® 4.0.3 software (copyright © 2000, SAS Institute Inc.). All models were constructed using the three variables of beer density, surface tension, and CO₂ content. The models were constructed using multiple linear regression, without any polynomial and interaction terms, to produce equations of the form $a_n = k_1g + k_2Dr + k_3C + b$, where the weighting factors k_{1-3} and the intercept b are found empirically. The same software, using a one-way ANOVA test, determined statistical significance, p . A p -value was calculated for each of the weighting factors.

Results and Discussion

Visual assessment of nucleation

To be completely accurate, the quantitative description of nucleation rate would be in the form of number of bubbles formed per unit time. However, as described above, obtaining this quantity presents some difficulty, and a more practical approach is desired. The method devised in this work consisted of placing beer in a standardized glass, taking a photograph of the beer and comparing the extent of bubbling in that photograph with that in a series of reference standards. The reference picture that most closely matched the beer sample was then used to afford a semi-quantitative measure of the frequency of bubble formation, since the number of bubbles seen in the glass at any one instant is proportional to the rate of bubbles formed per unit time.

This method is relatively simple and inexpensive and displayed reasonable reproducibility. Figure 4 shows the averaged results (6 repetitions) when the method was used on a single brand of beer. One source of error is likely to be the way the beer is poured into the glass: it is invariably a challenge to achieve total consistency in this with a manual dispense, and may justify a robotic approach in the future.

The method was able to differentiate beer brands into categories. For example, it could differentiate between a weakly-nucleating beer brand (average initial nucleation activity 7.3) and a well-nucleating brand (average initial nucleation activity 9.8) with $p < 0.01$.

Nucleation rates of unmodified beers

Though the physical container is of enormous importance in respect of affording nucleation sites in the form of scratches on the surface, the particular microcavities of all beer glasses in the trade vary widely and would be impossible to standardize. In addition, the serving conditions of a beer can seldom be controlled by the brewer, who instead can only manipulate the physicochemical properties of the product. For these reasons, our work only focused on the physicochemical properties of the beer and attempts to control for physical factors.

Eleven brands of beer were analyzed using the standardized glass (6 replicates), and their mass loss over time was also observed. In addition, other physicochemical properties were measured (3 replicates). The results for each brand's attributes were averaged, and are summarized in Table 1.

Every beer showed the type of behavior shown in Figure 5. The cumulative mass lost rose curvilinearly for the first 30 to 60 min, then linearly for the remaining duration of the experiment. The 2 regimes likely correspond to two different driving forces for mass lost. The bubbles observed in the glass within the linear region are usually quite small and sparse, and sometimes nonexistent, due to the depletion of most of the dissolved CO₂. Therefore, it is assumed that the bulk of the mass being lost from the glass in this region is due to CO₂ diffusion at the surface of the beer, and possibly some evaporation, as opposed to mass being carried away in bubbles, which is the primary means of mass loss in the beginning of the experiment.

It can be seen that mass loss over time and nucleation activity do not have any kind of linear or easily predictable correlation. This is primarily due to the fact that as the CO₂ in solution drops, the bubbles grow less on their way to the surface, and therefore carry less mass than the same number of bubbles from early in the experiment. Therefore, although using a balance is a more objective method, it does not describe the frequency of bubble formation as directly as the nucleation activity scale.

The physicochemical factors predicted to influence nucleation

According to Prins and van Marle (1999), a bubble at a nucleation site breaks away from that site when its buoyancy force overcomes the capillary force holding the bubble to the mouth of the site. When the site is a conical pit in a horizontal hydrophilic surface, this occurs when the following equation is satisfied:

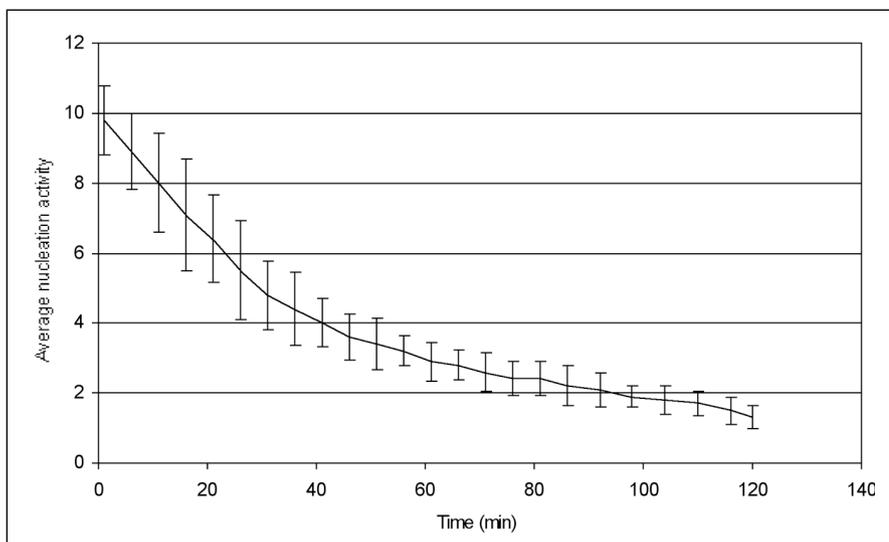


Figure 4—Nucleation activity over time of a single brand of beer, without the pressure containers. Average of 6 repetitions. The error bars show ± 1 standard deviation at each time.

Table 1—Summary of physical attributes for beers used in the study.

Brand:	A	B	C	D	E	F	G	H	I	J	K
Nucleation activity	9.5	10.1	8.7	7.2	7.0	7.0	9.0	7.3	7.0	7.8	8.1
CO ₂ (mL CO ₂ /mL)	2.73	2.63	2.62	2.38	2.44	2.14	2.63	2.86	2.39	2.64	2.57
Surface tension (mN/m)	46.4	47.9	44.5	44.4	44.0	45.6	43.6	46.9	42.8	43.3	44.5
Density (g/mL)	1.0066	1.0005	1.0055	1.0073	1.0094	1.0081	1.0087	1.0153	1.0100	1.0068	1.0065
pH	3.96	3.95	3.58	4.00	4.40	3.57	3.58	4.23	3.81	3.90	4.09

$$R_d = \left(\frac{3R_m\gamma}{2\Delta\rho g} \right)^{1/3} \quad (1)$$

where R_d is the radius of the bubble, R_m is the radius of the site mouth, γ is the surface tension of beer, $\Delta\rho$ is the density difference between beer and the gas phase, and g is the acceleration due to gravity. This equation indicates that each bubble released from a single site will be the same size. If it is known how long it will take the bubble to grow to that size, the rate of bubble release can be predicted. Prins and van Marle advance the following equation to predict the time needed for the bubble to grow to that size:

$$t_f = \frac{L^2}{\pi D} \quad (2)$$

where t_f is the time to form a single bubble, D is the diffusion coefficient of CO₂, and L is the distance over which the gas must diffuse. L may be calculated by finding the radius R of a sphere of liquid around the bubble that contains the same amount of gas as is present in the bubble. The difference between R and R_d is L . For beer, this equation can be solved as

$$L = R_d \left(\left(\frac{C+1}{C} \right)^{1/3} - 1 \right) \quad (3)$$

where C is the dissolved CO₂ content of the beer, expressed in volumes.

From equations 1 to 3, it can be seen that the rate of nucleation should be affected by four physicochemical properties of the beer: density, surface tension, the diffusion coefficient of the carbon dioxide, and the CO₂ content.

Since the number of bubbles seen in the glass at any one instant is proportional to the rate of bubbles formed per unit time, nucleation activity is inversely proportional to t_f . According to the presented theory, nucleation activity should depend linearly on

$$\gamma^{-2/3}, \Delta\rho^{2/3}, \text{ and } \left(\left(\frac{C+1}{C} \right)^{1/3} - 1 \right)^{-2}$$

Experimental observations and models

A model ('Model A') for nucleation activity based on the physicochemical properties given in Table 1 has been developed (Figure 6).

The formula for Model A is:
 $a_n^o = 3.11(C) + 0.0962(\gamma) - 218(\rho) + 216 \quad (4)$

Initial nucleation activity can be predicted with statistical significance using measure-

ments of C , g , and r . Incorporating pH, which is not thought to influence initial nucleation activity (a_n^o), into the model improves R^2 , but decreases p .

The equations for the models produced by the computer method used are not of the same form as the equations from theory. Therefore, a direct comparison of the empirical results and the predictions from theory is not possible. Multiple linear regression is, however, a useful tool for predicting how much the nucleation activity of beers should vary over the relatively small range of conditions found in commercial beers.

Carbon dioxide content is clearly crucial. Within commercial beers, carbon dioxide content is the physicochemical variable that has the strongest effect on nucleation activity. As little as 0.5 added volumes of dissolved CO₂ can mean the difference between a weakly and a strongly nucleating beer.

The p -value associated with the ρ coefficient is 0.03, indicating a statistically significant effect upon nucleation activity. However the direction seen here is opposite to predictions from Prins and van Marle's theory, which says that higher density should

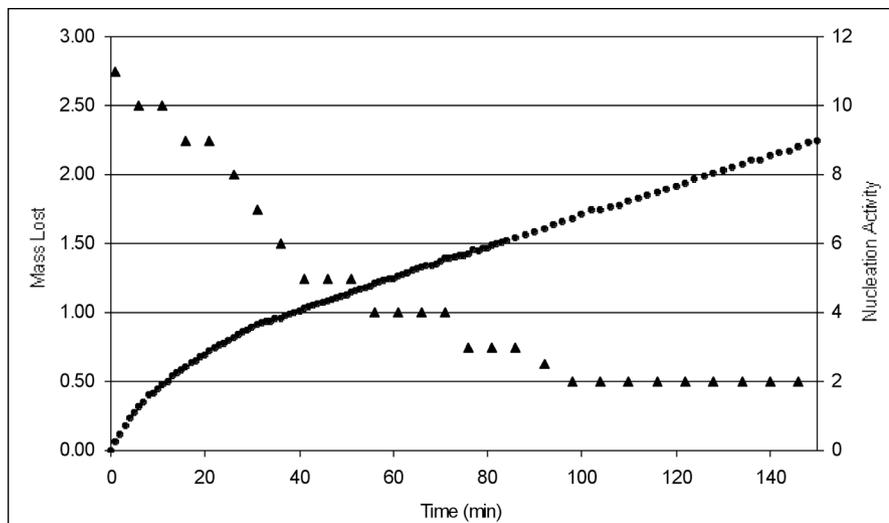


Figure 5—Typical behavior over time of a beer in the standardized glass, in this case Brand A. l = Mass lost. s = Nucleation activity.

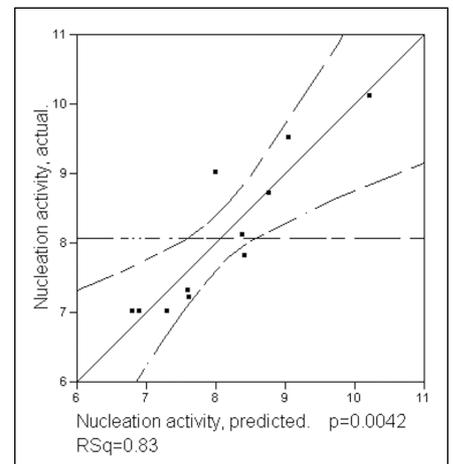


Figure 6—Predicted compared with observed plot for Model A, which predicts nucleation activity based on CO₂ content, surface tension, and density, using data from the standardized glass experiments. Each point represents the averaged values for a single brand.

lead to more buoyant bubbles that break off sooner (with a higher frequency) from the nucleation site. The density data in this case seems to be highly influenced by a single point, which corresponds to the non-alcoholic brand used in the study. It is possible that differences in CO₂ solubility or diffusivity in the non-alcoholic matrix introduced some error. It was calculated that without the influence of this point, there is no statistically significant trend present.

Conversely, the p-value associated with the γ coefficient is 0.44, indicating no statistically significant on a_n° within the range tested here. However, it would be unwise to suggest that γ has no effect whatsoever on nucleation activity. It is likely that the range for this variable in off-the-shelf beer is so small that any trend is difficult to perceive.

Nucleation rates of adjusted beers

The use of pressure containers eliminated the need for a scratch-free glass and meticulous pour due to the eradication of incidental nucleation sites (see Figure 2), and allowed for any beer to be standardized to a chosen CO₂ content, thereby controlling for this factor. In addition, samples could be manipulated to occupy a wider range for both surface tension and density, without adversely affecting CO₂ content. Use of the pressure containers also allowed all data for a sample to be collected from that sample itself. This is in contrast to the unrefined method, in which the value for CO₂ content must be assumed to be constant for separate bottles of each beer, as the method used for assessing CO₂ is destructive, and beer tested in this way cannot be used again to test other attributes.

A total of 33 samples were analyzed using the pressure containers. Some of the samples were manipulated to carry one or more of their attributes outside the normal range found in commercial beer. The aim of these experiments was the same as with the unmodified beers: to explore the correlations of CO₂ content, surface tension, and density with nucleation activity. The modification was carried out not to examine the effects of one particular change on that beer's nucleation rate, but rather to produce a data set that contained beers occupying a larger range of values for surface tension and density. The CO₂ content was standardized to ensure it was in a normal range after modification, and to minimize the overpowering effect of CO₂ content on nucleation activity.

All samples were prepared using the method described earlier and left to equilibrate under 35 psig CO₂ for 3 to 5 d. The manipulation of the beers was using deionized water, 200 proof ethanol, or sugar (sucrose).

The addition of water changes the surface tension of beer without greatly affecting its absolute density. The addition of sugar changes the density of beer without greatly changing its surface tension, and the addition of ethanol lowers both the surface tension and the density of beer. Therefore, a set of beers occupying a larger range of physicochemical values could be obtained without letting the changes vary co-linearly. The results are summarized in Table 2. In this case the nucleation activity was measured as actual number of bubbles present rather using the empirical 0 to 10 scale, in order to increase the objectivity of the data.

The set of beers modified with sugar presents an interesting case. After removal from the pressure vessel, each of the samples showed the same behavior when the sintered glass bead was introduced. Initially, a fair amount of nucleation activity was present, though the bubbles did appear noticeably smaller than normal. However, the activity fell away to virtually zero within 1 to 2 min, even though carbonation was still present in the beer (data not shown). This extremely fast decay in activity prevented the accurate measurement of a_n° . The cause of this phenomenon is unknown, but one possibility is that the high sugar concentration led to a change in the wetting properties of the liquid, which could have caused a disappearance of nucleation sites. Another possibility is that D was lowered significantly by the increased viscosity due to the addition of sugar.

The predicted vs. observed plot for the model fit to the modified beer data (designated 'Model B') is shown in Figure 7.

The formula for Model B is:

$$a_n^\circ = -10.7(\gamma) + 3060(\rho) - 2406 \quad (5)$$

Surface tension and density are clearly important in the situation where CO₂ content is not the overwhelming controlling factor of activity. The p-value for the γ coefficient is less than 0.0001, and the p-value for the ρ coefficient is 0.011, in each case indicating statistically significant trends. Furthermore the trends for both variables operate in the expected direction (both opposite to Model A). A rise in density produces a rise in nucleation activity, just as a rise in surface tension produces a fall in nucleation activity.

The results of statistical modeling can be summarized as follows: Even in beers standardized to the same CO₂ content, differences can be observed in nucleation activity, and these differences can be predicted in a statistically significant way using the physicochemical properties of interfacial surface tension and density of the beer.

In the overall model, the data points corresponding to pure water may appear to have undue influence, especially since they do not in actuality represent beer systems. However, when these data points are removed from the model, the qualitative result does not change (data not shown).

It is important to note that while C, γ , D, and ρ may be the sole physicochemical factors influencing nucleation, these quantities are in turn affected by other characteristics of the beer. For example, protein content can change γ , just as sugar and alcohol content can affect ρ .

Future work should focus on refining the arbitrary scale to account for differences in speed of bubbles as they ascend, which would be predicted to be influenced *inter alia* by bubble size and beer viscosity. The scope of the work might also be expanded to include the effects of nitrogen gas on the nucleation system. However the levels of N₂ that are introduced into beer to promote foam stability (Carroll, 1979) are three orders of magnitude lower than those of CO₂, which is consistent with the experiential observation that nitrogenated beers with low carbonation are "reluctant" to foam, hence the development of "widgets" as nucleation devices. Bubble size is a significant issue with regard to the stability of foams once formed, smaller bubbles affording longer retention of the head (Carroll, 1979).

Conclusions

AN ARBITRARY SCALE WAS DEVELOPED TO give a quick and convenient way to measure nucleation activity. In addition, a standardized glass was developed in order

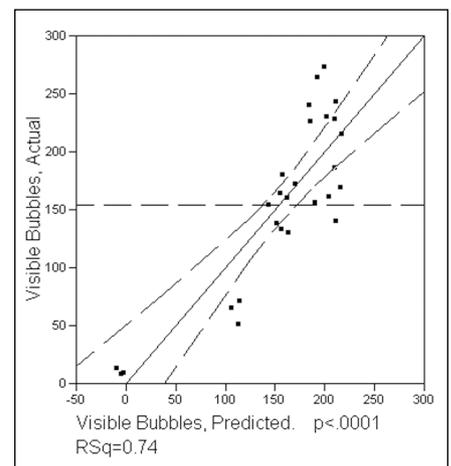


Figure 7—Predicted vs. observed plot for Model B, which predicts nucleation activity based on surface tension and density, using data from the pressure vessel experiments. Each point represents the data from one sample.

Table 2—Summary of results from pressure container experiments.

Brand	Preparation	Visible bubbles	ρ (g/mL)	γ (mN/m)
A	1:3 (v/v) Beer:Water	50	1.0016	51.1
A	1:3 (v/v) Beer:Water	64	1.0015	51.7
A	1:3 (v/v) Beer:Water	70	1.0015	51.0
A	1:1 (v/v) Beer:Water	137	1.0035	48.1
A	1:1 (v/v) Beer:Water	129	1.0035	47.0
A	1:1 (v/v) Beer:Water	153	1.0036	48.8
A	3:1 (v/v) Beer:Water	159	1.0058	47.7
A	3:1 (v/v) Beer:Water	168	1.0056	42.7
A	3:1 (v/v) Beer:Water	171	1.0056	46.9
B	80 mL ethanol added to 355 mL beer, reduced to 355 mL	132	0.9831	41.8
B	80 mL ethanol added to 355 mL beer, reduced to 355 mL	179	0.983	41.7
B	80 mL ethanol added to 355 mL beer, reduced to 355 mL	163	0.9838	42.1
(water)	Deionized water	12	0.9953	60.7
(water)	Deionized water	7	0.995	60.2
(water)	Deionized water	8	0.995	60.0
L	Unmodified	242	1.0068	43.5
L	Unmodified	227	1.0068	43.6
L	Unmodified	214	1.0068	42.9
L	Unmodified	263	1.00595	45.0
L	Unmodified	185	1.006	43.3
L	Unmodified	272	1.0059	44.3
M	Unmodified	139	1.0034	42.5
M	Unmodified	229	1.0034	43.4
M	Unmodified	160	1.0034	43.1
M	Unmodified	225	1.0032	44.8
M	Unmodified	155	1.0033	44.4
M	Unmodified	239	1.0032	44.9

to control for physical factors that could influence bubbling frequency, and the use of pressure containers further refined this method.

Laboratory data showed that the variables expected to be influential from theoretical considerations did have a measurable influence on the appearance of beer in a typical glass. CO₂ content was found to be the most influential variable, while the typical ranges for surface tension and density usually were not large enough to have a pronounced effect on what a consumer would see.

A brewery wishing to manipulate its product with regard to nucleation should consider that a raised CO₂ content will likely lead to more frequent nucleation, more head replenishment, and bigger average bubbles. A lowered surface tension would be expected to lead to more bubbling, with the average bubble size being reduced. A raised density would also probably lead to more frequent and smaller bubbles. It is important to note that the most influential factor influencing the appearance of the bubbles in the glass remains the physical

container itself. The particular number of scratches and potential nucleation sites present in the glass will largely dictate whether a beer will display lively or subdued bubbling.

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