AN ABSTRACT OF THE THESIS OF

<u>Christina Veronique Edwards van Muijen</u> for the degree of <u>Master of</u> <u>Science in Food Science and Technology</u>, presented on December 19, 2001.

Title: Development of a Lexicon for Staling Aromas in North American Lager and Analysis of Consumer Preference and Difference

Abstract approved: ______ Mina R. McDaniel

Staling in beer is inevitable. Oxidation and increased temperatures are the main causes of stale flavors in bottled beers. Since 1934, research has been done to identify compounds and pathways involved in creating stale flavors. Although many research papers report stale flavors, there is not a lexicon per se of stale aromas for North American lagers. This study has been divided into two parts, 1) the process of developing a lexicon for stale aromas for North American lagers and its application, and 2) the use of a consumer panel to determine whether the North American lager consumer has a preference for aged versus fresh beer and whether the consumer could perceive a difference between the two.

In the first study, three brands of North American lagers were aged in 27°C storage for three months and 38°C storage for two weeks. A lexicon for staling aroma for North American lagers was created, using a trained descriptive panel. The lexicon had two tiers; the first tier had five descriptors that were category headings for the second tier of descriptors. Panelists were instructed to rate the first tier descriptors, but using the descriptors in the second tier was optional.

To validate the lexicon, descriptive analysis was performed on the stored samples. The data were analyzed using principal components analysis (PCA) for the first tier descriptors and generalized procrustes analysis (GPA) for the second tier descriptors. The lexicon was used successfully, characterizing the control beers as *sulfury* and *fruity* and the aged beers as *sweet brown*. Within the *sulfury* category, panelists described control beers with the attributes *perm solution* and *skunky*. Within the *sweet brown* category, panelists described the aged beers with the descriptors *baked pineapple* and *honey*. The beers stored at different storage temperatures behaved differently across time. The maps constructed with PCA and GPA show a tendency for control samples to start out *sulfury* and, through time, age with *sweet brown* characteristics.

In the second study, a consumer panel was implemented to 1) determine if the average North American lager consumer had a preference for fresh versus stored beer, and 2) to determine if perceivable differences existed between the fresh versus stored samples.

A consumer test was designed using the three North American lagers that were tested in the trained panel. The target number of consumers for each brand was 100. The aged beer was stored at 38°C for 1 and 2 weeks, and the control was stored at 1°C for that time period. A preference test, followed by a triangle test, was performed on control versus 1 week at 38°C and control versus 2 week at 38°C for each brand (2 preference and 2 triangle tests for each brand; control versus 1 week and control versus 2 weeks). The results showed no significant preference for any brand/time point. Brand A had the only significant difference $(p \le 0.05)$ between samples stored at 38°C for 2 weeks and the control (The results showed no preference). Additional research must be done for more conclusive information, but this research shows that a small group of North American lager consumers do not have a preference between aged beer and fresh beer and, for the most part, cannot tell a difference between aged beer and fresh beer.

Development of a Lexicon for Staling Aromas in North American Lager

and Analysis of Consumer Preference and Difference

by

Christina Veronique Edwards van Muijen

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APPROVED:

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I understand that my thesis will become part of the permanent collection of Oregon State University libraries. My signature below authorizes release of my thesis to any reader upon request.

Christina Veronique Edwards van Muijen

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This work is dedicated to Tante Marie.

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DEVELOPMENT OF A LEXICON FOR STALING AROMAS IN NORTH AMERICAN LAGER AND ANALYSIS OF CONSUMER PREFERENCE AND DIFFERENCE

I. INTRODUCTION

The first study of the thesis focuses on creating and applying a practical lexicon for staling aromas in three brands of North American lagers. This study presents a compilation of resources and a more sharply defined lexicon for staling aromas that did not exist in the literature and that could be a useful tool for industry when working with sensory panels.

DeClerk (1934) was the first to discover that the main cause of flavor deterioration was due to oxygen ingress in bottled beer. Since then, flavor stability in beer has been a major priority in beer research. Meilgaard (1972), the inventor of the beer flavor wheel, describes the changes in beer flavor over time as, first a drop in intensity, then an increase of stale flavors. At approximately four weeks, a cardboard flavor develops, reaches a maximum at 8-12 weeks and then subsides. At 4-8 weeks, sweetish, woody and leathery notes first appear and increase in intensity at about 6-9 months of natural aging. Researchers examining staling flavors in beer have reported many descriptors to describe the aromas and flavors of stale beer, but they have not necessarily used formal sensory methods to validate their descriptors. Researchers have also proposed several storage regimes, but this study follows industry standards for accelerated and long-term storage, 27°C for 3 months, and 38°C for 2 weeks.

The second study of the thesis evaluates consumer response to aged beer. Do consumers prefer fresh or aged beer? Can consumers tell a difference between fresh and aged beer? Prior to 1996, freshness dating in the beer industry was used primarily for distributors to pull beer off shelves when it had reached the end of its shelf life. In September of 1996, Anhueser Busch launched a \$40 million dollar advertising campaign to introduce the "born on" date to make consumers aware of the freshness of the beer (Khermouch 1996). Other major brands followed suit, making the "born on" or "best before date" easier to read on packaging, which has become an industry standard (Hunter B 1996). This thesis study found consumers did not have a preference and overall, could not tell the difference between aged versus fresh beer.

II. LITERATURE REVIEW

This study develops a comprehensive lexicon to describe the staling of North American lager beer. The issues involved in developing this lexicon concern the ingredients, the processes used to brew, the optimal characteristics of, and the degradation processes associated with North American Lager beers. This degradation process would include Streker degradation, oxidation of higher alcohols, degradation of isohumulones, and oxidation of fatty acids.

North American Lager Beer Characteristics

Ingredients

North American lager beer is made primarily of malted barley, adjuncts, water, hops and yeast. Malted barley is used in beer as the source of extractable carbohydrates and also contributes important enzymes, which convert starch to sugar. Water is an essential ingredient because its mineral make up can affect the outcome of finished beer. Hops add bitterness and distinctive aroma and also act as an antioxidant. Yeast is added to ferment the beer, to produce alcohol and carbon dioxide.

Malted Barley

Malting of barley weakens the structural integrity of the kernel, and activates enzymes in the barley to convert starch to fermentable sugar, an integral ingredient in fermentation. Malt contributes to color, mouthfeel, sweetness, astringency, and alcohol strength (Papazian 1993). The four major steps in malting are 1) kernel selection, 2) steeping, 3) germination and 4) kilning (Lewis and Young 1996). In kernel selection, it is important to evaluate the kernel size for the best yield, and discoloration and aroma to ensure there is no mold growth (Noonan 1996).

Steeping is a process in which the barley moisture content is raised from approximately 10% to approximately 42%. This process starts the synthesis and migration of enzymes into the endosperm which ultimately lead to breakdown of the starches (Bamforth 1998). After barley has steeped for 24-48 hours, the barley goes through germination. This process lasts 3-5 days at 15-20°C, depending on barley variety and malting objective (Lewis and Young 1996). The 3-5 days of germination allow enough time for cell walls and protein to degrade and to make enzymes such as glucanases, proteases and amylases which convert starch to fermentable sugar.

Kilning stops the germination process so that the embryo does not grow too large. The temperature during kilning is usually between 50-85°C for approximately 24 hours depending on desired color and flavor. Flavors developed during kilning are toasted, grainy characters and dimethyl sulfide (canned cream corn aroma) (Papazian 1993). The moisture content of the malt is reduced to about 4%, which helps for storage and friability in milling.

Adjuncts

Adjuncts are cereals, syrups, and sugars that are used to supplement malt starch. Major brewers use adjuncts to replace malt and/or modify the beer flavor. The addition usually results in a lighter flavored beer. (Lewis and Young 1996). North American lager beer usually contains adjuncts if it is from a large brewery.

Water

Water, since it comprises 85-90% of the beer, is an important ingredient. Its mineral make up can effect the flavor, appearance and brewing process. The most important step to having a good water source is to find out from where the water came and of what it is comprised. Rainwater and seasonal streams and ponds are usually bad water sources since they may be highly polluted and variable in composition. Tap water from municipal water supplies are usually good sources. Water can be filtered, aerated or deionized to get the target composition for the desired beer. pH is also a concern, because it effects factors such as enzyme activity, acidity, hop extraction, protein precipitation in the kettle, yeast performance and clarification during fermentation. pH is hard to optimize since it needs to vary at different stages during the brewing process. The target pH is 5.2-5.5 for the saccharification rest in all mashes. This range is known to be efficient for all factors that affect the finished beer (Noonan 1996). Water hardness in the form of calcium and magnesium is also a factor that needs to be analyzed in water, since it can contribute to mash acidity and rob yeast of vital elements.

Hops

Hops contribute bitterness, flavor, and aroma. Aroma character can vary from metallic to citrusy to floral (Papazian 1993). Siebert (1994) notes that hops have a number of flavor descriptors. They include floral, grapefruit, citrus, spicy, fruity, resinous, piney, herbal, and cheesy. Yang and others (1993) found that humulene epoxides in hops contribute cedar, lime and spicy aroma notes to the beer. Kaltner and others (2001) report that by being selective in hop choice, a brewer can differentiate one's own brand from that of the competitor.

Hops are green leafy flower cones that grow on trellises that may be from 16-26 feet tall. Only female cones are utilized in brewing because they contain a resinous material called lupulin. These are glands in which the bittering compounds, the humulones, are found. Hops are harvested in mid-august and then dried in kilns at 55-65°C to reduce moisture to about 9% (Bamforth 1998). Hops are pressed into 150 kg bales and can be further processed into hop extract and pellets. The hops and hop products must be stored in dark, cold storage. There are many hop varieties, and only a small amount of sensory research characterizing their aromas has been done (Peacock and others 1981; Peacock and others 1980; Sanchez and others 1992; Stucky 1996, Stucky and McDaniel 1997). The varieties differ in the ratio of oil to resin, which translates into the finished product as aroma to bitterness. Researchers have determined that the main source of aroma in hops comes from the oil portion (DeMets and Verzele 1968; Sharpe and Laws 1981; Siebert and others 1989; Tressl and others 1978). Hop varieties high in oil such as Saaz also cost more. Brewers add *aroma* varieties at the end of the boiling period to produce the particular character of the hop desired. If pure bittering is desired, a less costly variety of hop is used and the aromas are driven off.

Yeast

In brewing, yeast produces ethanol, carbon dioxide and metabolic products that contribute flavor (Lewis and Young 1996). Yeast used in brewing is of the genus *Saccharomyces* and species *cerevisiae*. Lager beers in which the yeast ferments at the bottom of the tank are classified as *Saccharomycees uvarum* and ferment at low temperatures, approximately 6-15°C. Ale yeast (*Saccharomyces cerevisiae*) which ferments on top is carried out at higher temperatures (15-20°C) and ferment for a shorter period of time. Yeast strains are unique to many breweries, giving the beer a distinctive character, and therefore are not available to competitors (Lewis and Young 1996). It is important that the breweries keep the

same yeast strain to maintain a product that is consistent in aroma and flavor. The aromas that yeast can produce range from fruity to solvent-like (esters), clove-like (phenolic) if wild yeasts enters the culture, lard-like (fatty acids) if yeasts burst, rancid butter (diacetyl), and sulfur compounds when the yeast react with proteins (Papazian 1993).

Process

The brewing process involves many steps, each of which can influence the finished beer. Especially in lighter beers, ingredients and process can affect the outcome of the finished product due to the nature of the lighter flavor and color. The basic steps in brewing are as follows: 1) milling the malt into grist, 2) hydrating the grist to form mash, 3) separating the mash from the wort, 4) boiling wort with hops, 5) clarifying the wort, 6) cooling and aerating the wort, 7) fermenting, 8) conditioning the beer or lagering, and then packaging (Lewis and Young 1996; Noonan 1996).

Malt is milled using roller mills which cracks up the malt into a particular particle size determined by the brewer and type of malt used. When using malt husks for a filter bed, the particle size of the starchy endosperm must be fine enough so that it can be solublized by water. Malt that is well modified (has turned most of the starch into sugar) will not need as much milling (Bamforth 1998). Hydrating the milled malt or grist is called mashing. The grist is hydrated with liquor (hot water) and mixed thoroughly. This is the stage in which the enzymatic reactions take place. Some additions may be made during this stage such as lactic acid that helps to keep the pH at an optimum of 5.2-5.6 to produce more efficient reactions (Bamforth 1998; Lewis and Young 1996).

Separating the wort (liquid extracted from the mash) from the spent grain (milled malt) is a challenging process. The wort must be bright (clear) and contain no insoluble particles that may cause complications further down the process. To carry out this separation, a lauter tun (cylindrical vessel with a false bottom) is used. The false bottom of the lauter tun acts as a sieve so that larger particles of grain do not flow through. The grain on top of the false bottom also acts as a filtering aid. In a process called vorlauf, wort is cycled through the grain for clarification. After 10-20 minutes of vorlauf, the wort is transferred to the boiling kettle (Noonan 1996).

In the boiling kettle, wort is boiled from 1-2 hours. The boil serves many functions. It inactivates enzymes that have made it through the lautering process. Boiling sterilizes the wort from any undesirable microflora that might compete with the yeast during fermentation. The high heat causes cross-linkage (hot break) between the proteins and polyphenols that make the trube (polyphenol and protein precipitate) and wort easier to separate. The boil also isomerizes the alpha acids in the hops that cause them to become bitter. Lastly, the wort becomes more concentrated due to evaporation (Noonan 1996; Bamforth 1998).

After the boil, wort is separated by filtration from the polyphenol-protein mush (trube) and is cooled and oxygenated before entering into fermentation tanks.

In the fermentation tanks, yeast is added and then a time-temperature regimen is implemented until the beer has reached the desired specific gravity. This means that yeast has produced ethanol, which is less dense (has a lower specific gravity) than the wort. During the fermentation yeast also produce CO_2 , esters, higher alcohols, and secrete organic acids that cause a drop in pH (Bamforth 1998).

After fermentation, the beer is aged and conditioned. It is first separated from the yeast, which has settled to the bottom of the fermentation tank, and put into another tank. In this conditioning tank, beer is chilled to 0°-1°C so yeast and protein precipitate, which may cause haze in beer, will fall out of solution.

Following conditioning the beer is filtered and then packaged into kegs or bottles. To ensure that no microorganisms or yeast survive, beer may be sterile filtered or pasteurized (Lewis and Young 1996).

Lager Characteristics

Lagers are characterized by their low fermentation temperature (6-14°C) and longer duration of fermentation of up to two weeks. Lagers also use a different yeast species, *Saccharomyces uvarum*, which ferments at low temperatures and drops to the bottom of the yeast tank (Bamforth 1998). Lagers are mostly pale beers that are low to moderately bitter. They are made without a strongly expressed flavor (Kunze 1996). Typical North American Lagers only have three flavor components, isohumulone, ethanol and carbon dioxide (Meilgaard 1991). In a pale beer such as an American Lager, the fermentation esters are first to be perceived. Examples of these esters are the banana esters, which are the acetataes of the fusel alcohols, and apple esters, which are the ethyl esters of butyric, caproic, caprylic and capric acids (Meilgaard 1991). U.S. Lagers are high in fruity/estery and alcoholic flavors and low in bitterness and caramel flavors (Meilgaard 1982). The alcohol content of American lagers is approximately 4.5% depending on the brand. The top three beer producers in America making lagers and their light beer counterparts have 78% of the market share.

Pathways Involved in Creating Stale Flavors in Beer

The pathways involved in staling create flavors and aromas, which are comprised of carbonyl compounds, aldehydes, ketones, esters. The sensory characteristics resulting from the following proposed pathways are all similar since the pathways catalyze the reactions of the long chain aldehydes associated with staling aromas and flavors. Many pathways have been proposed and studied, which include: Strecker Degradation (Palamand and others 1970; Lermusieau and others 1999; Devreux and others 1982, Tressl and others 1978; Hashimoto 1972), Melanoidin mediated oxidation of higher alcohols (Hashimoto 1972), oxidative degradation of isohumulones (Hashimoto and Eshima 1977; 1978; Williams and Wagner 1979), oxidation of fatty acids (Drost and others 1971; Meilgaard and others 1971; Grigsby and others 1972; Stenroos 1973; Drost and others 1974; Whitear and others 1979; Drost and others 1990; Uchida and Ono 1996). These are all pathways in which researchers believe compounds are synthesized *de novo*, but Barker and others (1983), suggests that staling aldehydes are already present in beer and are detected when the sulfur components are driven off.

Strecker Degradation

Strecker degradation of amino acids is a Maillard reaction between dicarbonyls and alpha amino acids which degrades amino acids into aldehydes and ketones (Meilgaard 1972). Flavors and aromas developed are not among the main staling components. Bamforth (1999) notes, that these compounds may only be developed at levels substantially below their flavor thresholds. Cumulatively these and many other carbonyls may contribute to perceived staling. The conditions in which Strecker degradation is promoted are high heat and/or long term storage, pasteurization, malt kilning, mash and wort boiling. Some flavors/aromas that occur are high-threshold aldehydic notes and bread flavor when pasteurized.

Kamimura and Kaneda (1993) proposed that trans-2-nonenal is produced through the aldol condensation reaction between acetaldehyde and heptanal. Various other carbonyls may be produced through aldol condensations with proline as a necessary catalyst (Fix 1989). Acetaldehyde and n-butanal form 2butenal, 2-ethyl-2-butenal, trans-2-hexanal and 2-ethyl-2-hexenal with the presence of alanine. Hashimoto and Eshima (1977) concluded that amino acids may serve as a basic catalyst in the aldol condensation reaction of the aldehydes through the formation of an imine intermediate.

Melanoidin Mediated Oxidation of Higher Alcohols

Hashimoto (1972) found that volatile aldehydes such as trans-2-nonenal (paper/cardboard aroma) were formed from their corresponding alcohols when melanoidins accept hydrogen atoms from alcohols. This work was done in a model system. Devreux and others (1982) argue that this pathway is of little significance since the reaction occurs very slowly in the darkness and is also inhibited with low amounts of polyphenols. Irwin and others (1991) also argues that this pathway does not contribute to perceivable trans-2-nonenal (the paper aroma) because there is not enough 2-nonen-1-ol (nine carbon alcohol) in the beer. Conditions that may promote this reaction are increased temperatures, high levels of oxygen and low pH (Hashimoto 1972).

Oxidative Degradation of Isohumulones

It is well known that sun-struck flavors develop from photooxidation of isohumulones, but isohumulones also contribute to volatile aldehydes associated with stale aromas. High levels of isohumulones can undergo oxidadive degradation to yield volatile aldehydes associated with stale aromas in bottled beer during storage (Hashimoto and Eshima 1978). Isovaleraldehyde and isobutyraldehyde are the most common aldehydes formed and have a cheesy note (Hashimoto 1972). The isovaleryl and 2-methylbutyryl side chains of isohumulones can also form ethyl isovalerate and 2-methyl butyrate which contributes to the vinous character in aged beer (Kamimura and Kaneda 1993). These reactions usually take place during storage of bottled beer.

Oxidation of Unsaturated Fatty Acids

As with all of the pathways discussed, the oxidation of unsaturated fatty acids is also controversial. This pathway has attracted more attention than any other pathway for carbonyl formation (Bamforth 1999). Even though there are numerous papers on lipid oxidation in the context of staling, there is no evidence that this pathway has a greater contribution than any other beer deterioration pathway (Bamforth 1999). There are two routes of lipid oxidation; enzymatic (lipoxygenase-catalysed) and non-enzymatic.

Enzymatic Oxidation of Unsaturated Fatty Acids

Lypoxygenase from barley oxidizes linoleic and linolenic fatty acids to form carbonyl compounds (Bamforth 1999). Kamimura and Kaneda (1993), suggest that it is unlikely that the lipoxygenase enzymes survive in finished beer and directly contribute to the staling flavor. It is possible that some compounds formed by the enzymes during wort may survive in the finished beer and can contribute to stale flavor. This pathway is promoted by oxygen, and is inhibited by heat and polyphenols.

Non-enzymatic Oxidation of Unsaturated Fatty Acids

Superoxide and hydroxyl are very reactive radical forms of oxygen that perform non-enzymatic oxidation, which have breakdown products similar to those of lypoxygenase. In beer, the extremely reactive hydroxyl radical will most likely react with a sugar or ethanol before it finds a fatty acid. Superoxide is less reactive and tends to migrate and find unsaturated fatty acid molecules. Therefore, superoxide poses a greater risk to brewers (Bamforth 1999).

Drost and others (1971) concluded after several experiments, that a hypothetical precursor of cardboard flavor is a polar c18 fatty acid with one or more double bonds and one or more hydroxyl groups. Dalgliesh (1977) remarks that the reaction products of unsaturated aldehydes, e.g., 2,4-decadienal, 2,4heptadienal, and 2,4,7-decatrienal, are associated with fishy or cod liver oil type flavors.

The radical reactions are promoted by light and certain enzyme systems; most important are the transition metal ions. Radical formation is promoted by metal ions when they are in their more reduced state (Bamforth 1999).

Descriptive Sensory Analysis Methods

Descriptive analysis is used to define the sensory attributes (taste, odor, texture, sight, sound) of anything from food products to napkins using specific word descriptors. In a descriptive panel, panelists are instructed not to express their liking or disliking toward the product; rather their task is to recognize, identify and quantify sensory attributes of a particular product. Panelists for this type of descriptive technique are usually experts or have been trained for some period of time before they test a product. Four commonly used descriptive tests are: The Flavor Profile Method, The Quantitative Descriptive Analysis (QDA) Method, Free Choice Profiling and The Spectrum Method.

Flavor Profile

The Flavor Profile Method was developed by Arthur D. Little, Inc., in the later part of 1940 (Caul 1957). This method was the first developed using a group consensus rather than a technical expert. It lead the way for research and development of all different aspects of sensory evaluation (Stone and Sidel 1993). This method can be applied in situations where many and varied samples must be judged by a few highly trained tasters. During training, panelists are introduced to a wide variety of samples representing the product range. The panelists and panel leader develop and define terminology or lexicon (list of descriptors for a particular product). For evaluation of a product, the panelists are seated around a table and individually evaluate one sample at a time and record the intensities of attributes using a 7-point scale. It is essential, in this method, that panelists are able to work together in a group since the panel must come to a consensus profile for each sample (Meilgaard and others 1991).

Quantitative Descriptive Analysis (QDA)

The QDA method was developed by Stone and others (1974) to satisfy the needs of an increasingly competitive market, develop new products and improve data analysis methods. In developing this method, it was imperative that the results were actionable. Therefore, it was important to take into account responsiveness to sensory properties of the product, limited number of screened panelists (10-12), testing in individual booths, unbiased language development, quantitative data with replications, and a practical data analysis system (Stone and Sidel 1993). Discriminatory ability of sensory attributes among samples is the major criteria for panelist selection in the QDA method. Training of QDA panelists is similar to other descriptive methods in their use of product and ingredient references. Evaluation of products is done in individual booths to reduce distraction and panelist interaction.

Free-choice Profiling

Free-choice profiling is an approach to sensory profiling of foods and beverages in which each panelists use a list of their own descriptors without having to describe the meaning. The method was developed by Williams and Langron (1984). This method can be used if time is a limiting factor and descriptive analysis must be performed. Panelists need a minimal amount of training because they create their own descriptive terms on their individual ballot, but they must use those terms consistently. Besides the time saving, an advantage of free choice profiling is that panelists can still be regarded as naïve consumers since they are trained only on how to use the scale. Data from this method are analyzed by Generalised Procrustes analysis which allows for terms to be combined if they seem to measure the same attribute, and also adjusts for different scale usage among panelists. The result of the analysis is a consensus configuration revealing the interrelationships between the samples for the panel as a whole (Williams and Langron 1984).

Spectrum

The Spectrum Method was created by Gail Vance Civille and utilizes a customized approach to panel development, selection, training and maintenance. Civille and Lawless (1986) recognized the importance of language in describing perceptions and to train a panel to use the terms for different products. In training for the spectrum method panelists are exposed to the products and they create a list of descriptors based on the product. Panelist descriptor lists are compiled and then organized into a list of descriptors (lexicon) that all panelists will use. Reference standards are used in training to define descriptors so that all panelists describe the attribute in the same way. The Spectrum method does not require a specific scale, but the universal category scale (16-point) is most often used with at least two references anchored along the scale (Meilgaard and others 1991). Samples are evaluated individually in separate booths.

Sensory Studies Involving Beer Staling

Meilgaard has done extensive work in the flavor of beer by naming, identifying and defining over 239 compounds and their thresholds in beer (Meilgaard and others 1971; Meilgaard 1975a; 1975b), and compiling the data to create the beer flavor terminology system to use in descriptive analysis (Meilgaard and others 1979; Meilgaard 1982; Meilgaard and others 1982; Meilgaard and Muller 1987, Meilgaard 1991). Although the beer flavor wheel has been very useful in descriptive analysis, it lacks stale descriptors (Meilgaard 1991). Various researchers have used descriptive analysis methods to quantify the staling and/or quality of beer. In the following research presented it is evident there is a lack of terminology to define the stale/oxidized aroma. Mercredy and others (1974) used a modified QDA method to profile beer using the term "oxidized" as the only staling descriptor. Guinard and others (1999) used the Spectrum method to rate
the quality of beer using the staling descriptor "oxidized/cardboard". Furusho and others (1999) used sensory evaluation to reveal that Japanese pilsner beer has flavor changes during storage. The term "stale" was defined as "oxidized, molasses-like, and whiskey-like". Ogane and others (2000) correlated the original freshness scale (a mathematical equation expressed as a function of time and storage temperature) with sensory analysis. Schmitt and Hoff (1979) used graphic linear scales to measure rates of staling beers. The 6-inch scale was anchored with the words not oxidized on the left end and extremely oxidized on the right end. A technique that can be effective when combined with descriptive analysis is gas chromatography olfactometry (GCO). In GCO, a sniff port connected to a gas chromotograph (GC), is utilized to smell effluents as they are separated by the GC. Evans and others (1998) used a gas chromatography olfactometry technique to identify flavor components of fresh, naturally aged and forced aged lagers (accelerated). Their results illustrated that the fresh sample was associated with a bitter character, the naturally aged sample had a papery character, and the accelerated storage sample had a sherry type character. They also determined that it is more likely that aldehydes associated with staling aromas are not synthesized de novo, but rather already exist, but are masked by other aroma components. Sakuma and others (2000) used GC olfactometry techniques to identify off-flavors in beer, and used descriptors such as disinfectant, sea urchin/egg-like and musty.

Storage Regimens

Storage regimens are a factor in research on stale flavor. Table 1.2 contains examples of storage regimens in beer literature.

Table 2.2 – Some storage regimens in staling beer literature

Length in storage	Temperature	Author
4 days/1 day shaking	40°C	Back 1999
6 months	20°C and 30°C	Furusho 1999
8-12 weeks	15°C	Clapperton 1976
30, 60, 90 days	22°C and 38°C	Strenroos 1973
16 weeks (agitated)	33°C	Meilgaard 1972

As shown in the table, time/temperature conditions vary with author, some using accelerated storage and some not. There are conflicting reports on the useful application of accelerated storage. Bamforth (1999) notes that flavor changes occur 30 fold faster at 35°C than at 0°C. This kind of stressed heat on the beer would be detectable within a week, whereas the 0°C stored beer will not start to show signs of aging for 6 months. Back (1999) reports that beer stored at 40°C for four days corresponds in sensory characteristics and analytically to beer stored for 3-4 months at 20°C. Evans and others (1998) and Bright and others (1993) disagree. Evans and others (1998) reports the naturally aged beer imparts a papery note, while the accelerated storage beer imparts a sherry note. Bright and others (1993), performed chemical analysis on American lager beer and found that the stale compounds development during storage varied considerably between accelerated storage and natural aging.

III. DEVELOPMENT AND APPLICATION OF A LEXICON FOR STALING AROMAS IN NORTH AMERICAN LAGERS

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Abstract

A lexicon to more precisely define staling aroma for North American lagers was created using a trained descriptive panel. This lexicon had two tiers: the first tier had five descriptors which also served as category headings for the second tier of descriptors. Three brands of North American lagers, selected on the basis of their popularity, were aged for 4, 8 and 12 weeks in a storage room with a controlled environment set at 27°C. Samples from the same production batch were aged for 5, 8 and 14 days in a separate storage room with a controlled environment set at 38°C. The control (fresh beer) was stored at 1°C. To validate the lexicon, panelists were instructed to rate the stored beer, using the first tier descriptors, but rating the second tier descriptors was optional.

The panelists' responses were analyzed using principal components analysis (PCA) for the first tier attributes and Generalized Procrustes analysis (GPA) for the first and second tier attributes.

The lexicon was used successfully, characterizing the control beers as *sulfury* and *fruity* and the aged beers as *sweet brown*. Within the *sulfury* category, panelists described control beers with the attributes *perm solution* and *skunky*. Within the *sweet brown* category, panelists described the aged beers with the attributes *baked pineapple* and *honey*. The maps constructed with PCA and GPA show a tendency for control samples to start out *sulfury* and, through time, age with *sweet brown* characteristics.

Introduction

In the brewing industry a lexicon of beer flavor and aroma has been developed, however, the descriptors dealing with staling aromas needs to be elaborated. Terms describing staling aroma descriptors are scattered among journal articles, but research creating a lexicon of staling aromas for North American Lagers does not exist. Researchers examining staling flavors in beer have reported many descriptors, but not necessarily using formal sensory methods to validate their descriptors. Table 3.1 contains a list of descriptors compiled from several research articles describing the aromas and flavors of stale beer.

Author	Descriptors
Bamforth 1999	Declined fruity/estery, floral, ribes (black currant),
	Paper/cardboard, bready, sweet, toffee-like, honey, metallic, earthy, straw, woody, winey (sherry-like)
Furusho and	Stale (oxidized, molasses-like, whiskey-like), Papery
others 1999	(cardboard, bready), Leathery (caramel-like, toffee-like)
Meilgaard 1982	Oxidized-stale-musty (moldy, leathery, papery, catty, stale)
Back 1999	Sweet-malty-off-flavor
Kaneda 1995	Caramel
Hashimoto and others 1977	Rough, dull
Grigsby and others 1972	Sour grainy

Table 3.1—Compilation of descriptors from several research articles describing the aromas and flavors of stale beer.

Meilgaard and others (1979) created a beer flavor terminology system. The system was organized into a wheel, which has several classes of descriptors and is further defined into first tier and second tier terms. The system was developed to enable brewers to communicate effectively about flavor and aroma descriptors. Each flavor descriptor has its own name, definition, and is illustrated with a flavor standard. One area, in which the wheel is lacking, is the stale flavor and aroma (Meilgaard 1991). Consequently, this study's objective was to create a lexicon for a descriptive panel to use to describe staling aromas in North American Lager beer. To induce staling aromas, industry professionals as well as previous research were consulted to determine appropriate time/temperature conditions. The two storage regimens used were those practiced by industry -- 27°C (80°F) for two weeks and 38° (100°F) for three months. Another objective of this research was to monitor the changes in beer aroma during the two different storage regimens.

Researchers have previously concluded that beer held at 0-4°C fails to display signs of oxidation even after months of storage (Bamforth 1999). When stored at higher temperatures, chemical reactions occur causing stale flavors. There are several reviews of beer flavor perception through storage (Bamforth 1999; Dalgliesh 1977; Meilgaard 1972). Bamforth (1999), based upon the research of Meilgaard 1972, and Dalgliesh 1977 and others, described the following observations of flavor changes in lager beer. Between 1-3 months beer will decline in bitterness and fruity/estery notes, it will increase in ribes and papery cardboard notes. As the beer ages it is described as bready, sweet, toffee-like, honey, metallic, earthy and straw. Finally a beer will become woody and winey.

Going beyond the research on staling itself, an exhaustive body of literature exists concerning research on time/temperature storage conditions. The following research articles are an example of the many different time/temperature combinations that have been researched. Stenroos (1973) looked at the parameters of storage time, temperature, and air content to better understand the aging process of finished bottled beer. The beer was stored for 30, 60 and 90 days at 0°C, 22°C, and 38°C. Meilgaard (1972) assessed flavor stability by tasting the beers just after bottling and again after 4, 8, 12 and occasionally 16 weeks of agitated storage at 33°C. Lindsey (1974) discussed the range of temperature storage as follows: Suppressed staling rate 0°C-10°C, Usual (Natural) staling rate 21°C-30°C, Accelerated 35°C-45°C and higher. To measure rates of staling in beer using graphic linear scales, Schmitt and Hoff (1979) stored beer for their first study at 0° C and 24°C, which contained varying levels of air. In their second study, they stored their beer with varying levels of air at 0°C, 15.5°C, 24°C and 38°C. Clapperton (1974), in his study of ribes (black currant) flavor in beer found that at about 15°C with increased headspace air, the ribes odor developed within 4 weeks from time of bottling and then decreased at 8-12 weeks of storage. Furusho and others (1999) used Japanese pilsner beers to develop a descriptive sensory test in which beer was stored at 20°C or 30°C for 1,2, 3 and 6 months then transferred to 0°C until testing.

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Since the research cited above has numerous variations on storage methodology, the time/temp regimes used in this study are based on industry practices within major U. S. breweries. The objectives of this research are to (1) create a lexicon for stale aromas in North American lagers and (2) to use the developed lexicon to monitor the changes occurring during 27°C storage for 3 months and 38°C storage for 2 weeks.

Materials and Methods

Samples

Three commercial bottled North American Lagers, which will be referred to as Brand A, Brand B and Brand C, were selected for this study on the basis of market position and brand. All brands were produced within 10 days of each other and were purchased from distributors.

Sample Storage

The samples were aged at 27°C (80°F) for three months and 38°C (100°F) for two weeks. Temperature and time in storage are representative of industry practices. The samples were stored at 1°C (34°F) after purchase and were then put into controlled temperature storage rooms at their respective time point so that all

samples were taken out of storage at the same time. Samples were held at 27°C for 4, 8 and 12 weeks and were held at 38° C for 5, 8 and 14 days. The control samples were held at 1°C until testing to minimize degradation reactions.

Panelist Selection

Nine panelists were selected on the basis of their performance on a preceding panel that also involved staling aromas in North American lagers. Panelists were experienced in rating intensities and using the protocol since the previous study had 31 one-hour training sessions and 11 one-hour testing sessions. All but two panelists were used for the current study. The panel consisted of four males and five females ranging in age from 24-51.

Panelist Training

Lexicon Development

Aroma descriptors for stale and fresh beer were introduced to the panelists by using standards from the literature and previous work. During training with actual test samples, panelists were able to add and remove descriptors that were not useful. Panelists discussed descriptors and came to a consensus on 19 descriptors and their corresponding standards (Table 3.2 Lexicon, standards and

Attributes	Definitions	Standards
Overall Aroma	The overall aroma intensity	
Fruity	Overall fruity intensity	
Artificial Fruit*	Estery notes associated with artificial fruit	1 Peach-O,1 stick Juicy Fruit gum
Pineapple*	The aroma associated with canned pineapple	20 g Dole crushed pineapple canned
Apple/Pear*	Aromatic characteristic of pome fruits	6 cubes (¼"x¼") Gala apple, 6 cubes Bartlett pear
Sweet Brown*	Overall aromatic associated with the results of reactions of amino acids and reducing sugars and/or the caramelization of sugars	
Caramelized*	Sweet aromatic characteristic of browned sugars and other carbohydrates	1" Score bar w/o choc, 3 pieces butter rum Life Saver, 10 g caramel corn, ¼" cube gjetost
Baked Pineapple	The volatile aromatic compounds generated from Betty Crocker pineapple upside-down cake topping	20 g Betty Crocker pineapple upside-down cake topping
Honey*	The sweet, c aramelized floral and w oody a romatic associated with honey	20 g Barkman's clover honey
Prune/Raisin*	A browned sweet fruity aromatic reminiscent of dried prunes or raisins	1 Sunsweet prune, 15 g Albertson's raisins
Cooked Fruit	Aromatic associated with the process of heating/cooking fruit	20 g cooked Del Monte canned peaches, 20 g cooked Oregon Fruit canned plum
Sherry	The aromatic associated with cream sherry	2 g Sheffield cream sherry
Sulfury*	Aromatic associated with hydrogen sulfide	.
Skunky*	Aromatic associated with the sulfur compound 3-methyl-2- butene-1-thiol (MBT), which exhibits a skunk-like character	20 g Corona beer
Cream Corn (DMS)*	A cooked vegetable aroma caused by Dimethyl sulfide	20 g canned cream corn
Yeast	The aroma of yeast	¹ / ₄ tsp. yeast in 40 ml of malt
Perm Solution	The aromatic associated with permanent solution	Aroma stick dipped 1/4" into Oglvie permanent solution
Paper	An a romatic t hat is slightly musty and similar to brown craft paper	2"x2" brown paper towel moistened
Musty	Aromatic characteristic of damp/wet basements or turned soil	2"x2" clean cotton dish towel moistened

Table 3.2 Lexicon descriptors, definitions and standards used by descriptive analysis panel in training and testing

Bold = 1st tier terms, indented = 2nd tier terms *Definition from ASTM DS 66 (Civille 1996)

definitions). Although panelists came to a consensus on the descriptors, actual application of rating intensities was very difficult since the beers were so light and similar in aroma. To help panelists stay on task and minimize fatigue, the lexicon and ballot were constructed using a tier system (Table 3.2 Lexicon, standards and definitions). The first tier consisted of five descriptors which also served as category headings for the second tier of descriptors. For example, the first tier term "*fruity*" was the category heading for the second tier terms "*apple/pear*, *pineapple* and *artificial fruit*". Panelists were instructed to rate the first tier descriptors, but rating the second tier descriptors was optional.

Training and Practice

Training consisted of lexicon development, intensity scaling, and practice sessions using the stored samples. Panelists had 22 one-hour training sessions and 6 one-hour testing sessions. Of the 22 training sessions, 13 were used for lexicon development and learning use of the 16-point intensity scale, where 0 = None, 4 = Just Detectable, 8 = Slight, 12 = Moderate, and 15 = Moderate to Large. During lexicon development, panelists were exposed to all of the test samples to ensure that the lexicon was appropriate for all samples. The other 9 sessions were used for practice.

Sample Preparation

One ounce (30 ml) of sample was served in an eight-ounce teardrop wineglass covered with a plastic lid (Sweetheart USL3) at 10-13°C (50-55°F). Samples were gently poured into stainless steel one ounce measuring cups and then into wineglasses in a 6°C (42°F) cold room to minimize carbonation and temperature loss. Samples were held at room temperature until they reached 10°C (3-5 minutes) and then served immediately to panelists.

Testing Protocol

Before each testing session, panelists could re-visit the standards with which they trained. Panelists were served four sets of three samples during each session and recorded their ratings on a paper ballot. Panelists rested 5 minutes between the first and second set, 10 minutes between the second and third set, and 5 minutes between the third and forth set. The resting periods were implemented to minimize adaptation and fatigue.

For aroma assessment, panelists were instructed to hold the glass by the stem, swirl gently, take three short sniffs, recap the wineglass, and rate intensities.

Experimental Design

Each brand/temperature condition was tested as an independent experiment in order to maximize the differences found in the aged beers. The order of testing for each experiment was randomly assigned and applied to each panelist. Within each brand/temperature condition, a balanced completely randomized block design was employed. Panelists were treated as the blocking criteria and the effects of the four storage times were tested. Three replications were performed for each sample.

Data Analysis

Data were analyzed using multivariate analysis of variance (MANOVA) to obtain means and multivariate significant differences. The data from each brand/temperature condition were analyzed separately. In MANOVA, time in storage, panelist and replication were main effects and treated as fixed effects. ANOVA was performed after the MANOVA in order to indicate the influential factors that caused the differences found in the MANOVA. In ANOVA models, time in storage, panelist and replication were main effects. The time and the replication were treated as a fixed effect; and the panelist was treated as a random effect. Tukey's HSD was used to detect significant differences of descriptors indicated by ANOVA. Principal Components Analysis (PCA) was used to create a perceptual map of first tier descriptors, and Generalized Procrustes Analysis (GPA) was used to evaluate term usage and map the second tier descriptors. First tier descriptors were mapped along with the second tier to determine weather the second descriptors would fall into their categories. These methods allowed determination of the lexicon's effectiveness and which descriptors describe which samples. The analysis for MANOVA followed by ANOVA for each descriptor and PCA was done with SPSS v. 10.1 (2000). GPA was conducted using Senstools v. 2.3 (1997).

Results

Lexicon

Panelists agreed upon five first-tier descriptors and fourteen second-tier descriptors to be used in describing the aroma of the beer used in this study (Table 3.2). Through MANOVA significant differences overall across storage times within all brand/temperature treatments were obtained except for Brand C stored at 38°C. ANOVA and the Tukey's HSD multiple comparison procedure were performed detect and indicate which descriptors represented the differences among the samples within the brand /temperature treatments.

Analysis of Variance for First Tier Descriptors

Brand A

Through MANOVA significant differences between samples were found for Brand A stored at 27°C (Table 3.3). After running an ANOVA and looking at pairwise comparisons with Tukey's HSD, the descriptor *overall intensity* was significant (F= 3.776, p-value =0.024), but panelists were unable to separate between samples due to the small magnitude of difference (this is due to ANOVA having a higher sensitivity than Tukey). *Sweet brown* was significantly higher in the 4, 8 and 12week samples than the control (F = 10.516, p-value = 0.000). *Sulfury* was significantly higher in the control and the four week samples than the 8 and 12 week samples (F = 8.863, p-value = 0.000). The descriptors *fruity* and *papery* were not significant (Table 3.3, Figure 3.1).

MANOVA results show that there were significant differences between samples for Brand A stored at 38° C (Table 3.4). After running ANOVA and looking at pairwise comparisons with Tukey's HSD, the descriptor *overall intensity* was significantly higher in the 8 day sample than the control (F= 4.068, p-value = 0.018). The descriptor *sulfury* was significantly higher in the control and 8-day samples than the 14-day sample. The descriptor *sweet brown* was significantly higher in the 14-day sample than the 5-day and control samples.No significant differences were obtained for the descriptors *fruity* and *paper* (Table 3.4, Figure 3.2).

Storage			_		
time			Sweet		_
(weeks)	Overall	Fruity	Brown	Sulfury	Paper
0	11.0	6.0	3.7°	7.8ª	2.0
	(1.1)	(4.0)	(3.5)	(2.7)	(2.7)
4	10.9	4.4	6.8 ^b	6.4ª	2.0
	(1.2)	(4.1)	(2.9)	(3.1)	(2.5)
8	11.2	4.3	8.7 ^a	4.4 ^b	1.1
	(1.0)	(3.8)	(2.2)	(3.3)	(2.2)
12	11.5	4.7	8.6ª	3.7 ^b	2.2
	(1.4)	(4.1)	(3.2)	(3.9)	(2.9)

Table 3.3-- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand A samples stored at 27°C.

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)

Table 3.4 -- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand A samples stored at 38°C.

Storage time (days)	Overall	Fruity	Sweet Brown	Sulfury	Paper
0	10.3 ^b	5.5	5.1 ^b	7.4 ^a	0.7
	(1.2)	(3.8)	(3.8)	(1.8)	(1.9)
5	10.8 ^{ab}	5.3	5.4 ^b	6.5 ^{ab}	2.1
	(1.1)	(4.0)	(3.3)	(2.8)	(2.7)
8	11.2 ^a	4.9	5.8 ^{ab}	7.5 ^a	2.1
	(1.2)	(4.6)	(3.6)	(3.0)	(3.0)
14	11.1 ^{ab}	4.7	7.4 ^a	5.5 ^b	1.1
	(1.2)	(4.0)	(2.6)	(4.0)	(2.0)

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)



Figure 3.1 First tier descriptors for Brand A stored at 27°C for 0-12 weeks



Figure 3.2 First tier descriptors for Brand A stored at 38°C for 0-14 days

Brand B

MANOVA results show that there were significant differences between samples for Brand B stored at 27°C (Table 3.5). After running ANOVA and looking at pairwise comparisons with Tukey's HSD, the descriptor *fruity* was significantly higher in the control than the 8 and 12 week samples (F= 6.315, pvalue = 0.003). *Sweet brown* was significantly higher in the 4, 8 and 12-week samples than the control (F= 12.739, p-value = 0.000). *Sulfury* was significantly higher in the control and 4-week samples than the 8 and 12-week samples (F= 8.127, p-value = 0.001). Brand B stored at 27°C had no significant differences for the descriptors *overall intensity* and *paper*. (Table 3.5, Figure 3.3).

MANOVA results show that there were significant differences between samples for Brand B stored at 38°C (Table 3.6). After running ANOVA and looking at pairwise comparisons with Tukey's HSD, the descriptor fruity was significantly higher in the 8-day samples than the 14-day samples. The descriptor *sweet brown* was significantly higher in the 14-day sample than the 8-day, 5-day and control samples ($F_{\gamma 1, \gamma 2} = 5.298$, p-value = 0.006). *Sulfury* was significantly higher in the control and 5-day samples than the 14-day sample ($F_{\gamma 1, \gamma 2} = 4.048$, pvalue = 0.018). No significant differences were obtained for the descriptors *overall intensity* and *paper*. (Table 3.6, Figure 3.4).

Storage time			Sweet		
(weeks)	Overall	Fruity	Brown	Sulfury	Paper
0	10.8	6.7 ^a	3.5 [♭]	7.8ª	1.1
	(1.4)	(3.6)	(3.4)	(1.7)	(2.0)
4	10.9	5.3 ^{ab}	7.3 ^a	5.9 ^b	0.8
	(1.3)	(4.0)	(3.0)	(2.7)	(2.0)
8	11.1	3.8 ^b	8.9 ^a	3.4 ^c	2.1
	(1.1)	(4.2)	(1.9)	(2.9)	(2.9)
12	11.1	4.4 ^b	8.6 ^a	4.2 ^c	1.4
	(1.2)	(3.9)	(3.1)	(3.3)	(2.3)

Table 3.5-- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand B samples stored at 27°C.

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)

Table 3.6-- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand B samples stored at 38°C.

Storage time (days)	Overall	Fruity	Sweet Brown	Sulfury	Paper
0	10.5	5.9 ^{ab}	4.3 ^b	6.9ª	1.6
E	(1.3)	(3.3)	(3.9)	(2.8)	(2.7)
2	(1.4)	3.4 (3.6)	(3.2)	6.7 (2.6)	1.7 (2.4)
8	10.8 (1.1)	6.6ª (3.9)	6.0 ^b (3.6)	6.4 ^{ªb} (2.6)	1.5 (2.7)
14	11.0	4.5 ^b	8.2ª	4.8 ^b	1.6
	(1.0)	(4.6)	(2.7)	(3.3)	(2.7)

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)



Figure 3.3 First tier descriptors for Brand B stored at 27°C for 0-12 weeks



Figure 3.4 First tier descriptors for Brand B stored at 38°C for 0-14 days

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Brand C

MANOVA results show that there were significant differences between samples for Brand C stored at 27°C (Table 3.7). After running ANOVA and looking at pairwise comparisons with Tukey's HSD, Brand C stored at 27°C, the descriptor *sweet brown* was significantly higher in the 4, 8 and 12-week samples than the control. The descriptor *sulfury* was significantly higher in the control than the 8 and 12-week samples (F= 3.934, p-value = 0.020). No significant differences were obtained for the descriptors *overall intensity*, *fruity*, and *paper*. (Table 3.7, Figure 3.5).

MANOVA results show that there were no significant differences among samples for Brand C stored at 38°C (Table 3.8). After running ANOVA and looking at pairwise comparisons with Tukey's HSD, the descriptor *paper* was significant (F= 3.39, p-value =0.034), but unable to separate between samples due to the small magnitude of differences. No significant differences were obtained for the descriptors *overall intensity*, *fruity*, *sweet brown* and *sulfury* (Table 3.8, Figure 3.6).

Principal Component Analysis (PCA)

PCA was performed to summarize the correlated descriptors on the first tier of the lexicon. The location of samples in this spatial mapping technique was determined along with influential descriptors inherent in each sample. In the PCA

Storage time			Sweet		
(weeks)	Overall	Fruity	Brown	Sulfury	Paper
* <u></u>					
0	10. 9	4.8	4.8 ^b	7.4 ^a	2.0
	(1.0)	(3.6)	(3.9)	(2.2)	(2.7)
4	10.9	4.1	6.5ª	6.2 ^{ab}	1.6
	(1.3)	(3.7)	(3.6)	(2.7)	(3.0)
8	11.1	39	8 () ^a	4 6 ^b	15
0	(10)	(3.9)	(3, 2)	(3.6)	(2.4)
	(1.0)	(3.7)	(3.2)	(3.0)	(2.4)
12	10.6	3.0	7.0 ^a	5.1 ^b	1.7
	(1.0)	(3.9)	(3.2)	(3.5)	(2.7)

Table 3.7 -- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand C samples stored at 27°C.

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)

Table 3.8-- Mean response (using a 16-point scale) and standard deviations () of first tier aroma descriptors for Brand C samples stored at 38°C.

Storage time (days)	Overall	Fruity	Sweet Brown	Sulfury	Paper
0	10.7	4.3	6.0	7.3	1.3
	(1.1)	(3.7)	(3.8)	(2.1)	(2.3)
5	10.9	4.1	7.4	5.7	1.6
	(1.3)	(4.0)	(2.9)	(3.2)	(2.6)
8	11.0	4.2	6.1	6.9	2.1
	(1.1)	(4.2)	(3.2)	(3.4)	(2.8)
	. ,				
14	11.0	4.1	6.8	6.8	2.3
	(1.1)	(3.9)	(3.1)	(3.2)	(2.6)
	. /	. ,	. ,	. ,	. ,

^{abc}Means with common superscripts within columns are not significantly different ($p \le 0.05$)



Figure 3.5 First tier descriptors for Brand C stored at 27°C for 0-12 weeks



Figure 3.6 First tier descriptors for Brand C stored at 38°C for 0-14 days

map, the farther along the axis a sample lies, the more intense the characteristic descriptors of that axis' direction. Panelists found it difficult to use first tier descriptors precisely for samples that are close to the (0,0) coordinate. Principal component 1 had significant sample separation in most of the brand/temperature conditions, but principal component 2 had no significant sample separation.

Brand A

For Brand A stored at 27°C, principal component 1 (PC1) explained 43.7% of the variation while principal component 2 (PC2) explained 26.0 % of the variation (Figure 3.7). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, the samples separated into three groups. The first group located on the right side of the plot is the control explained by the descriptors *fruity* and *sulfury*. The second group, in the center of the plot, is the 4-week and 8-week samples. The third group contains the samples 8-week and 12-week, which are explained by the descriptor *sweet brown*.

For Brand A stored at 38°C, PC 1 explained 34.1% while PC 2 explained 30.3% (Figure 3.8). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, the samples did not separate into groups. However, there is a trend for the 14-day samples to be grouped and located toward the location of *sweet brown* on the PC map.



Figure 3.7 -- Principal component plot for Brand A stored at 27°C



Figure 3.8 -- Principal component plot for Brand A stored at 38°C

For Brand B stored at 27°C, PC 1 explained 38.7% of the variation while PC 2 explained 23.5% of the variation (Figure 3.9). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, the samples separated into three groups. The first group was the control sample located on the right side of the plot, which was explained by the descriptors *sulfury* and *fruity*. The second group was the four-week sample, which was located in the center of the plot. The third group was the 8-week sample and 12 week sample. These samples are located on the left side of the plot and are explained by the descriptor *sweet brown* on PC1 and *paper* on PC 2.

For Brand B stored at 38°C, PC 1 explained 44.0% of the variation while PC 2 explains 26.5% of the variation (Figure 3.10). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, the samples separated into two groups. The group on the right, control 5 and 8-week samples are characterized by the descriptors *sulfury* and *fruity*. The second group, 14-day, located on the left was characterized by the descriptor *sweet brown*.





Figure 3.9 -- Principal component plot for Brand B stored at 27°C



Figure 3.10 -- Principal component plot for Brand B stored at 38°C

Brand C

For Brand C stored at 27°C, PC 1 explained 38.3% of the variation while PC 2 explained 29.1 % of the variation (Figure 3.11). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, the samples separated into two groups. The first group located on the right side of the plot is the control and 4-week samples characterized by the descriptors *fruity* and *sulfury*. The second group is the 4, 8 and 12-week samples, characterized by the descriptor *sweet brown*.

For Brand C stored at 38°C, PC 1 explains 38.7% of the variation while PC 2 explains 25.4% of the variation (Figure 3.12). After running ANOVA and looking at pairwise comparisons with Tukey's HSD on PC1, samples did not separate. Although there was no statistical separation, the control sample was in the region characterized by the descriptor *sulfury* and the other samples are clustered together near the (0,0) coordinate where it is difficult to make a distinction of their aroma characters.



Figure 3.11 -- Principal component plot for Brand C stored at 27°C

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Figure 3.12 -- Principal component plot for Brand C stored at 38°C
Generalized Procrustes Analysis

Generalized Procrustes analysis (GPA) was performed after PCA. This analysis was done to take into account the second tier of descriptors. The first tier was also analyzed along with the second tier descriptors and results showed more separation than PCA. Through GPA, second tier descriptors are also validated by the frequency of panelist usage. GPA provides a map similar to PCA in that the farther along the axis and from (0,0) coordinate a sample lies, the more intense the characteristic descriptors of that axis direction. Therefore samples that are in close proximity to the (0,0) coordinate are hard to characterize with descriptors.

Tables 3.9-3.14 are organized under each brand/temperature condition to aid in describing the GPA maps. These tables contain a complete list of all of the descriptors that were used by panelists on the positive and negative end of dimension 1 and dimension 2. The second tier descriptors that have a larger number under the "#pan" column are good candidates to expand the first tier of the lexicon.

Brand A

For Brand A stored at 27°C, dimension 1 explains 43.97% of the variation while dimension 2 explains 12.05% of the variation (Figure 3.13). Through ANOVA followed by Tukey's HSD the samples were separated into three groups on dimension 1. One group contained the control and the 4-week samples, the next group contained the 4 and 8-week samples and the last group contained the 8 and 12-week samples. Although there is no distinction between the middle samples, 4 and 8-week, there is a clear separation between the control and the 12-week, control and 8-week, and 4 and 12-week. The control is characterized by the first tier descriptors *fruity* and *sulfury* and further refined with the second tier descriptors, *artificial fruit, perm solution, skunky*, and *cream corn*. The 12-week sample is characterized by the first tier descriptor, *sweet brown*, and further refined with the second tier descriptors *baked pineapple* and *honey*.



Figure 3.13 -- Consensus plot following Generalized Procrustes analysis for Brand A stored at 27°C

Dim 1 (negative)		Dim 1 (posi	tive)	Dim 2 (neg	Dim 2 (negative) Dim 2 (po		itive)	
Descriptor	#pan	Descriptor	#pan	Descriptor	#pan	Descriptor	#pan	
sulfury	6	swtbrown	8	swtbrown	3	overall	3	
permsol	5	bakedpine	6	bakedpine	2	pineapple	3	
fruity	4	honey	6	cookfruit	2	app/pear	2	
skunky	4	cookfruit	2	fruity	2	cr.com	2	
artfruit	3	fruity	2	honey	2	fruity	2	
cr.com	3	caramel	1	paper	2	paper	2	
paper	2	overall	1	sherry	2	permsol	2	
app/pear	1	paper	1	skunky	2	skunky	2	
cookfruit	1	pineapple	1	sulfury	2	sulfury	2	
honey	1	pru/raisin	1	app/pear	1	yeast	2	
musty	1	sherry	1	artfruit	1	cookfruit	1	
pineapple	1			caramel	1	honey	1	
swtbrown	1			cr.corn	1	musty	1	
yeast	1			permsol	1	pru/raisin	1	
				pineapple	1	swtbrown	1	
<u></u>				pru/raisin	1			

Table 3.9 Complete list of descriptors that describe GPA map for Brand A 27°C

#pan = Number of panelists who used the descriptor

For Brand A stored at 38°C, dimension 1 explains 26.9% of the variation while dimension 2 explains 20.8% of the variation (Figure 3.14). ANOVA followed by Tukey's HSD separated the samples into three groups on dimension 1. The group on the left side is the control. The second group, in the center, is five and 8-day samples while the third group, on the right, is 14 and 8-day samples. Similarly to the 27°C sample, the middle samples do not have distinguishable descriptors. The control can be characterized by the first tier descriptors, *sulfury* and *fruity*, and further refined with the second tier descriptors *skunky* and *yeast*.



Figure 3.14 -- Consensus plot following Generalized Procrustes analysis for Brand A stored at 38°C

The 14-day sample can be characterized by the first tier descriptor, *sweet brown*, and further refined with the second tier descriptors *baked pineapple* and *honey*.

Dim 1 (negative)		Dim 1 (posi	itive)	Dim 2 (nega	Dim 2 (negative) Dim 2 (positi		itive)
Descriptor	#pan	Descriptor	#pan	Descriptor #pan		Descriptor	#pan
sulfury	7	swtbrown	7	bakedpine	3	bakedpine	4
skunky	4	bakedpine	6	Fruity	3	paper	4
yeast	4	honey	4	Overall	3	honey	3
fruity	3	paper	2	permsol	3	swtbrown	3
artfruit	2	pineapple	2	pineapple	3	artfruit	2
cr.corn	2	sulfury	2	app/pear	2	fruity	2
honey	2	caramel	1	cookfruit	2	overall	2
musty	2	cookfruit	1	musty	2	permsol	2
paper	2	cr.corn	1	paper	2	pineapple	2
permsol	2	fruity	1	skunky	2	app/pear	1
cookfruit	1	permsol	1	sulfury	2	musty	1
pineapple	1	pru/raisin	1	swtbrown	2	pru/raisin	1
swtbrown	1	sherry	1	caramel	1	sherry	1
		-		cr.corn	1	skunky	1
				pru/raisin	1	sulfury	1
				pru/raisin	1	-	

Table 3.10 Complete list of descriptors that describe GPA map for Brand A 38°C

#pan = Number of panelists who used the descriptor

Brand B

For Brand B stored at 27°C, dimension 1 explains 47.1% variation while dimension 2 explains 13.2% variation (Figure 3.15). ANOVA followed by Tukey's HSD separated the samples into three groups on dimension 1; the control on the left, the 4-week sample in the center and the 8 and 12-week samples on the right. The control grouping is characterized by the first tier descriptors, *fruity* and *sulfury*, and further refined with the second tier descriptors *perm solution*, *skunky* and *artificial fruit*. The 12 and 8-week samples are characterized by the first tier descriptors, *sweet brown*, *paper* and *overall*, and further refined with the second tier descriptors *baked pineapple*, *caramel*, and *honey*. The 4 week grouping is located in the center were there is no clear distinction between descriptors.



Figure 3.15 -- Consensus plot following Generalized Procrustes analysis for Brand B stored at 27°C

Dim 1 (negative)		Dim 1 (posi	itive)	Dim 2 (nega	Dim 2 (negative) Dim 2 (positi		tive)
Descriptor	#pan	Descriptor	#pan	Descriptor #pan		Descriptor	#pan
sulfury	7	swtbrown	8	fruity 3		paper	4
permsol	6	bakedpine	6	overall	3	caramel	3
fruity	6	caramel	5	sulfury	3	fruity	2
artfruit	3	honey	5	artfruit	2	overall musty	2
skunky	3	paper	4	bakedpine	2		2
yeast	2	overall	3	paper	2	pineapple	1
app/pear	2	sherry	2	cr.corn	2	app/pear	1
cr.corn	2	pru/rai	2	permsol	2	bakedpine	1
overal	1	cookfruit	2	honey	2	cookfruit	1
bakepine	1	pineapple	2	swtbrown	2	honey	1
pineapple	1	musty	1	skunky	1	cr.com	1
				sherry	1	swtbrown	1
	. <u></u>			pineapple	1	yeast	1

Table 3.11 Complete list of descriptors that describe GPA map for Brand B 27°C

#pan = Number of panelists who used the descriptor

For Brand B stored at 38°C, dimension 1 explains 34.0% of the variation while dimension 2 explains 14.0% of the variation (Figure 3.16). ANOVA followed by Tukey's HSD separated the samples into two groups on dimension 1. The first group contains the control, 5 and 8-day and the second group contains the 5, 8, and 14-day samples. While there is no clear separation between the middle samples, 5 and 8-day, there is a clear separation between the control sample and the 14-day. The control group can be characterized by the first tier descriptors, *sulfury* and *fruity*, and can be further refined with the second tier descriptors *skunky* and *yeast*. The 14-day sample can be characterized by the first tier



Figure 3.16 -- Consensus plot following Generalized Procrustes analysis for Brand B stored at 38°C

descriptor, *sweet brown*, and further refined with the second tier descriptors *baked pineapple* and *honey*.

Dim 1 (negative)		Dim 1 (posi	m 1 (positive) Dim 2 (negative) Dim		Dim 2 (posi	itive)	
Descriptor	#pan	Descriptor	#pan	Descriptor #pan		Descriptor	#pan
sulfury	7	swtbrown	7	bakedpine 3 1		bakedpine	4
skunky	4	bakedpine	6	fruity	3	paper	4
yeast	4	honey	4	overall	3	honey	3
fruity	3	overall	2	permsol	3	swtbrown	3
artfruit	2	paper	2	pineapple	3	artfruit	2
cr.corn	2	pineapple	2	app/pear	2	fruity	2
honey	2	sulfury	2	cookfruit	2	overall	2
musty	2	caramel	1	musty	2	permsol	2
paper	2	cookfruit	1	paper	2	pineapple	2
permsol	2	cr.com	1	skunky	2	app/pear	1
cookfruit	1	fruity	1	sulfury	2	musty	1
pineapple	1	permsol	1	swtbrown	2	pru/raisin	1
swtbrown	1	sherry	1	caramel	1	sherry	1
				cr.corn	1	skunky	1
				honey	1	sulfury	1
				pru/raisin	1		

Table 3.12 Complete list of descriptors that describe GPA map for Brand B 38°C

#pan = Number of panelists who used the descriptor

Brand C

For Brand C stored at 27°C, dimension 1 explains 35.7% of the variation

while dimension 2 explains 11.53 % of the variation (Figure 3.17). ANOVA

followed by Tukey's HSD separated the samples into two groups on dimension 1.



Figure 3.17 -- Consensus plot following Generalized Procrustes analysis for Brand C stored at 27°C

The control and 4-week samples were in one group while the 4, 8 and 12-week samples were in the second group. The control clearly separates from the 8 and 12-week samples as being characterized by the first tier descriptors, *sulfury*, *fruity*, and *sweet brown*, and further refined with the second tier descriptors *perm solution*, *skunky*, *pineapple* and *baked pineapple*. The trend of the 8 and 12-week samples is characterized by the first tier descriptor, *sweet brown*, and further refined with the second tier descriptor. The trend of the 8 and 12-week samples is characterized by the first tier descriptor, *sweet brown*, and further refined with the second tier descriptor, and further refined with the second tier descriptor.

Dim 1 (negative)		Dim 1 (posi	tive)	Dim 2 (nega	ative)	Dim 2 (posi	tive)
Descriptor	#pan	Descriptor	#pan	Descriptor	#pan	Descriptor	#pan
sulfury	6	swtbrown	5	honey	6	fruity	3
fruity	4	bakedpine	4	swtbrown	5	paper	3
permsol	4	honey	4	overall	4	overall	2
skunky	4	musty	2	bakedpine	3	pineapple	2
bakedpine	3	sherry	2	musty	2	artfruit	1
pineapple	3	caramel	1	paper	2	bakedpine	1
swtbrown	3	cookfruit	1	cookfruit	1	cr.com	1
honey	2	cr.corn	1	fruity	1	musty	1
overall	2	overall	1	permsol	1	permsol	1
app/pear	1	paper	1	pineapple	1	pru/raisin	1
artfruit	1	pru/raisin	1	sherry	1	sulfury	1
cookfruit	1	skunky	1	skunky	1		
musty	1	yeast	1	yeast	1		
paper	1						
yeast	1						

Table 3.13 Complete list of descriptors that describe GPA map for Brand C 27°C

#pan = Number of panelists who used the descriptor

For Brand C stored at 38°C, dimension 1 explains 26.7% of the variation while dimension 2 explains 16.8% of the variation (Figure 3.18). ANOVA followed by Tukey's HSD could not separate the samples. Although there are no statistically significant groupings, there is a trend for the 8 and 14-day samples to be characterized by the first tier descriptors, *sweet brown* and *fruity*, and further refined with the second tier descriptors *baked pineapple* and *honey*. The control and the 5-day samples are characterized by the first tier descriptors, *overall, fruity*, and *sulfury*, and further refined with the second tier descriptor *baked pineapple*.

Dim 1 (negative)		Dim 1 (posi	tive)	Dim 2 (nega	ative)	Dim 2 (posi	tive)
Descriptor	#pan	Descriptor	#pan_	Descriptor	#pan	Descriptor	#pan
bakedpine	3	bakedpine	4	swtbrown	5	permsol	3
fruity	3	honey	4	bakedpine	3	swtbrown	3
overall	3	swtbrown	4	fruity	3	bakedpine	2
sulfury	3	fruity	3	artfruit	2	honey	2
sherry	2	app/pear	2	cr.corn	2	paper	2
swtbrown	2	caramel	2	overall	2	sulfury	2
artfruit	1	cookfruit	2	permsol	2	artfruit	1
caramel	1	cr.com	2	pineapple	2	cookfruit	1
cookfruit	1	paper	2	app/pear	1	cr.corn	1
cr.com	1	sulfury	2	honey	1	skunky	1
honey	1	artfruit	1	musty	1		
pineapple	1	musty	1	paper	1		
pru/raisin	1	overall	1	pru/raisin	1		
skunky	1	permsol	1	sherry	1		
yeast	1	pineapple	1	sulfury	1		
		sherry	1	yeast	1		
		skunky	1				_

Table 3.14 Complete list of descriptors that describe GPA map for Brand C 38°C

#pan = Number of panelists who used the descriptor



Figure 3.18 -- Consensus plot following Generalized Procrustes analysis for Brand C stored at 38°C

Discussion

First Tier: Analysis of Variance

Overall, the descriptors *sweet brown* and *sulfury* had the largest effect as the samples changed than any other descriptors. The changes that took place during storage with the descriptor *sulfury* are in congruence with the findings of Barker and others (1983) who found that the reduction of *sulfury* intensity during storage also aids in the perception of aldehydes (stale beer aromas). The *sweet brown* descriptor has not been used in beer terminology, but it is most likely to be a culmination of the aroma associated with reactions of amino acids and aldol sugars and the caramelized notes often described by brewers. The descriptor *sweet brown* and its relationship to staling aldehydes must be further investigated.

In addition to *sulfury* and *sweet brown*, the intensity of *fruity* also changed during storage. However, the changes in *fruity* occurred specifically in Brand B stored at 27°C. Therefore, the descriptor *fruity* was effective only for this brand/temperature condition. This may be a function of different ingredients (hop product, yeast strain) used and their aging process.

Samples stored at 27°C behave differently than samples stored at 38°C. For example, in Brand B stored at 27°C (Figure 3.3), the 8-week storage temperature shows a large magnitude in intensity between the descriptors *fruity* and *sulfury* and *sweet brown*, while the 8-day sample stored at 38°C (Figure 3.4) has a negligible magnitude between those descriptors. Clearly, panelists are able to use the lexicon to separate samples, as shown in the results of the MANOVA, PCA and GPA. Since there is a smaller magnitude between the *sulfury* and *sweet brown* descriptors in all of the brands stored at 38°C (Figures 3.1-3.6), samples stored at that temperature do not develop as distinct aromas as samples stored at 27°C. This supports the conclusion of Bright and others (1993), that accelerated testing at 38°C may produce misleading results due to the considerable variability in the carbonyl profile of packaged American-style lagers stored at different testing temperatures.

First Tier Descriptors: PCA

The first tier of the lexicon was very successful in distinguishing the control from aged samples. The attributes that contributed the most to the separation were *fruity*, *sulfury* and *sweet brown*, where the control was in the region characterized by *fruity* and *sulfury* and the aged samples were in the region characterized by *sweet brown* (figures 3.7-3.12). From this analysis *sulfury* and *fruity* are highly positively related. By contrast, the *sweet brown* descriptor is highly negatively related with *sulfury* and *fruity* as shown on PC1 in most of the PCA plots. This is in agreement with the analysis of variance of the first tier terms. *Paper* was the predominant descriptor on PC2 in most of the plots, but it did not have enough impact to create differences among the samples. Research on

the paper/cardboard aroma (trans-2-nonenal) in beer stored at different temperature conditions has shown that a sample stored at 60°C had a similar paper/cardboard aroma to that of a sample stored at 37°C and 18°C (Bright and others 1993; Kaneda and others 1995; Greenhoff and Wheeler 1981).

As noted above, PCA supports the results of the analysis of variance. The behavior of the samples at different storage temperatures is also in agreement with the analysis of variance. The samples stored at 27°C were more distinctly separated than samples stored at 38°C. For example, Brand B samples stored at 27°C are separated into 3 groups on PC1, while Brand B samples stored at 38°C are separated into two groups on PC1 (Figures 3.9 and 3.10). Samples stored at 38°C for Brands A and C were clustered in the (0,0) coordinate region where there was no clear distinction as to their characters. Where there is no separation in any of the brands and temperatures, the explanation may lie with the following two possibilities: (1) The lexicon developed was not applicable for that particular brand (which was unlikely because all samples tested were used in lexicon development step), (2) The particular sample does not possess any distinctive descriptor, but has a little of each of them. (This possibility is more likely).

Second Tier Descriptors: GPA

Samples analyzed using GPA had the same trends as analysis of variance and PCA, but added refined descriptors to the broad categories of the first tier terms. The first tier descriptor, *sweet brown*, was positively related with the second tier descriptors, *baked pineapple* and *honey*. The first tier descriptor *sulfury* was positively related with the second tier descriptors *perm solution* and *skunky*. The first tier descriptor, *fruity*, was positively related with the second tier descriptor, *artificial fruit* (Figures 3.13-3.18, Tables 3.9-3.14).

Conclusion

The lexicon for aged beer aroma was developed using 19 descriptors. The two most vital descriptors for separating the samples were *sulfury* and *sweet brown*. The descriptors describing staling from the literature were not as useful in describing the stored samples as the descriptors generated from the panel.

PCA was used to gain a broad understanding of how panelists used first tier descriptors over the entire range of samples within storage conditions. The second tier descriptors, analyzed by GPA, refined the broad categories of the first tier descriptors. Panelists used the descriptors *skunky* and *perm solution* to refine the *sulfury* group, the descriptors *baked pineapple* and *honey* to refine the *sweet brown* group, and the descriptor *artificial fruit* to refine the *fruity* group.

The trends of significant descriptors over storage time depended on brand and temperature condition. Evidence from samples analyzed through ANOVA, PCA and GPA reveal that commercial American lagers age differently at the two storage temperatures, 27°C and 38°C, so caution must be taken when using accelerated temperatures.

IV. ANALYSIS OF CONSUMER PREFERENCE AND DIFFERENCE FOR AGED NORTH AMERICAN LAGER

Christina Veronique Edwards van Muijen and Mina R. McDaniel

<u>Abstract</u>

From a previous study on staling of North American lagers, a trained panel found differences in aroma attributes between fresh and aged beer (Edwards van Muijen and McDaniel 2001). Those differences were mainly in *sulfury* and *sweet brown* aromas, where the fresh beer had higher *sulfury* characteristics and the aged beer had higher *sweet brown* characteristics. In view of the marketing of freshness dating, a consumer panel was implemented to determine (1) if the average North American lager consumer had a preference for fresh versus aged beer, and (2) if perceivable differences existed between the fresh versus aged samples.

A consumer test was designed, using the three North American Lagers that were tested in the trained panel. For Brand A 79 consumers participated, for Brand B 99 consumers participated, and for Brand C 93 consumers participated. The aged beer was stored at 38°C for 1 and 2 weeks, and the control was stored at 1°C for the same time period. A preference test followed by a triangle test was performed on control versus 1 week at 38°C and control versus 2 week at 38°C for each brand (2 preference and 2 triangle tests for each brand; control vs. 1 week and control vs. 2 week).

The findings suggest consumers have no significant preference for any brand or time point. Brand A had the only significant difference ($p \le 0.05$) between samples stored at 38°C for 2-weeks and the control. This research shows that a self-described group of North American lager consumers do not have a preference between aged beer and fresh beer and, for the most part, cannot tell the difference between aged beer and fresh beer.

Introduction

Aging in beer is often thought of as a detriment to the beer's flavor (Schmitt and Hoff 1979; Lynch and Seo 1987; Ogane and others 2000; Bamforth 1999; Devreux and others 1982). Hundreds of researchers have studied staling in beer, most trying to understand how to stop it. However, according to Axcell and Torline (1998), consumers do not have an adequate understanding of oxidized or stale flavors. Even if flavor changes (such as caramel-like or bready) are noticed, they may not necessarily be negative. Some people prefer a malty or grainy taste rather than a fresh sulfur note that occurs in lagers (Axcell and Torline 1998).

Guinard and others (2000), in a study involving consumer testing with commercial lager beers proposes that consumer preferences should be closely examined because they are the driving force behind product sales. Another interesting result from this study was that liking for taste was the best predictor for overall liking, which confirms previous findings of Moskowitz and Krieger (1992).

A recent study verified that a trained descriptive panel could determine differences in character between the aromas of fresh and stale lager beer (Edwards van Muijen and McDaniel 2001). This descriptive panel found that the fresh beers contained *sulfury* aromas, while the aged beers had *sweet brown* characteristics such as *baked pineapple*, *honey*, and *caramel*.

The current study proposes to determine if a consumer panel could perceive the changes found by the descriptive panel. As a result, a consumer panel was conducted using the three brands of North American lagers tested by the trained descriptive panel. The beers for this portion of the study were aged for 0, 1 and 2 weeks at 38°C. The consumer test was composed of a preference test followed by a triangle test for each brand/temperature combination. The of this study objectives were 1) to determine if consumers had a preference for fresh versus aged beer, and 2) to determine if perceivable differences existed between the fresh versus aged samples.

Materials and Methods

Samples

Three bottled North American lager brands were selected for this study on the basis of market position and brand. To ensure confidentiality, the brands will be referred to as Brand A, Brand B, and Brand C. Each brand of beer was tested on a separate day, making three days of testing. Samples were purchased from local distributors. Samples of the same brand were from the same production lot. All brands were produced within 3 days of each other based on the date coding from the imprint on the package. The age at purchase was 2 weeks, and the age at serving was 5 weeks.

Sample Storage

Brand A, Brand B, and Brand C were aged at 38°C (100°F) for one week and two weeks. Temperature and length in storage are representative of industry practices for accelerated storage testing. The samples, after purchase, were put into a controlled temperature (38°C) storage room at their respective time point so that they were removed at the same time. They were then stored at 1°C for 4 days until testing.

Sample Preparation

Samples were prepared for serving in a 5°C cold room to minimize carbonation loss. The beer was carefully poured into a stainless steel one-ounce measuring cup and then into a 7-ounce plastic glass covered with a plastic lid. When served to panelists, samples were between 5°-7°C (42-45°F).

Sample Presentation

Panelists, seated in individual testing booths lit with incandescent bulbs, were instructed first to read and sign the informed consent form (Appendix 2). They were then given the samples and recorded their test responses on the ballot (Appendix 3) provided. A preference test followed by a triangle test was presented for control versus 1-week and control versus 2-week, totaling four tests for each panelist. The order of which storage time was tested first was balanced and then randomized across panelists. Samples were presented to panelists for the preference test first. The control and the aged sample were presented simultaneously on a tray to the panelists who were instructed to recorded their preference on the ballot. When finished tasting, the panelists were given the ballot for the triangle test. Two control samples and one aged sample (or vice versa) was presented simultaneously on a tray to panelists. After tasting and recording their answer on the ballot, the panelists repeated the preference test and triangle test for the next storage time. The order in which samples were presented on trays was balanced and then randomized across panelists. After the panelist finished the last sample, he/she filled out a demographic questionnaire (Appendix 4).

Panelists

Panelists were recruited by e-mail from the Sensory Lab database by use of a screener. The criterion to pass the screener was to drink American lager style beer on at least one occasion per month (Appendix 1) and be at least 21 years old. Seventy-nine people participated in the tests for Brand A, ninety-nine people participated in the tests for Brand B, and ninety-three people participated in the tests for Brand C. Panelist demographic information is in appendix 5.

Data Analysis

Preference Test

The results of the consumer paired preference tests for 1-week and 2-week aged samples versus control were interpreted using the procedure outlined in Meilgaard and others (1991). To determine whether there was a significant preference for one of the samples, a z-statistic ($\alpha = 0.05$) was calculated and then compared to the α -critical value on a standard normal table.

The equation to determine the z-statistic is:

 $Z = (k-0.5n))/\sqrt{0.25n}$, where k is the number of correct responses and n is number of subjects.

Triangle Test

The results of the consumer triangle test for 1-week and 2-week aged samples versus control were interpreted using the procedure outlined in Meilgaard and others (1991). A difference test was used because the findings of previous research (Edwards van Muijen and McDaniel 2001), suggests that the samples to be tested are different according to a group of trained panelists. Our objective for the triangle test was to determine whether consumers could tell a difference between fresh versus aged beer. To determine if a significant difference was shown for one of the samples, a z-statistic ($\alpha = 0.05$) was calculated and then compared to the α -critical value on a student's t-distribution. The equation to determine the z-statistic is as follows:

 $Z = (k - (n/3))/\sqrt{(2n/9)}$, where k is the number of correct responses and n is the number of subjects.

Results

The results are presented in two sections according to storage time versus the control. The results of the two preference tests and two triangle tests within each brand are reported in Table 4.1.

Table 4.1 Percentages of panelist's scores for the preference tests (Pref.) and triangle tests (Δ) of 1-week versus control and 2-week versus control

Brand	Pref.	1 week	Δ 1 week		Pref.	2 week	Δ 2 week	
Scores in %	l wk	control	corr	incorr	2 wk	control	corr	incorr
A, n=79	48	52	27	73	51	49	44*	56
B, n=99	55	45	28	72	57	43	31	69
C, n=93	54	46	39	61	54	46	29	71

*Significant at p≤0.05

1-week versus Control

Paired Preference Test

For the paired preference test for samples stored for Brand A (n=79), fiftytwo percent of panelists preferred the control sample and forty-eight percent of panelists preferred the samples stored for 1-week at 38°C. For the paired preference test for samples stored for Brand B (n=99), forty-five percent of panelists preferred the control sample and fifty-five percent of panelists preferred the 1-week sample. For the paired preference test for samples stored for Brand C (n=93), forty-six percent of panelists preferred the control sample and fifty-four percent of panelists preferred the 1-week sample. There was no significant preference (p<0.05).

Triangle Test

For the triangle test for samples stored for Brand A (n=79), twenty-seven percent of panelists got the test correct and seventy-three percent of panelists were incorrect. There was no significant difference ($p \le 0.05$). For the triangle test for samples stored for Brand B (n=99), twenty-eight percent of panelists got the test correct and seventy-two percent of panelists were incorrect. There was no significant difference ($p \le 0.05$). For the triangle test for samples stored for Brand C (n=93), thirty-nine percent of panelists got the test correct and sixty-one percent of panelists were incorrect. There was no significant difference ($p \le 0.05$).

2-week versus Control

Paired Preference Test

For the paired preference test for samples stored for Brand A (n=79), fiftyone percent of panelists preferred the control sample while forty-nine percent of panelists preferred the 2-week sample. There was no significant preference ($p \le 0.05$). For the paired preference test for samples stored for Brand B (n=99), forty-three percent of panelists preferred the control sample while fifty-seven percent of panelists preferred the 2-week sample. There was no significant preference ($p \le 0.05$). For the paired preference test for samples stored for Brand C (n=93), forty-six percent of panelists preferred the control sample while fifty-four percent of panelists preferred the 2-week sample. There was no significant preference ($p \le 0.05$). For the paired preference test for samples stored for Brand C (n=93), forty-six percent of panelists preferred the control sample while fifty-four percent of panelists preferred the 2-week sample. There was no significant

Triangle Test

For the triangle test for samples stored for Brand A (n=79), forty-four percent of panelists got the test correct and fifty-six percent of panelists were incorrect. At the ($p\leq 0.05$), there was a significant difference. For the triangle test

for samples stored for Brand B (n=99), thirty-one percent of panelists got the test correct and sixty-nine percent of panelists were incorrect. There was no significant difference ($p\leq0.05$). For the triangle test for samples stored for Brand C (n=93), twenty-nine percent of panelists got the test correct and seventy-one percent of panelists were incorrect. There was no significant difference ($p\leq0.05$).

Discussion

Preference (Paired Preference Tests)

For all of the brands tested, there were no significant preferences for samples stored for 1 or 2 weeks at 38°C as compared to a control. This supports observations by Axcell and Torline (1998) even though major breweries spend copious amounts of time and money researching ways to preserve freshness, consumers still purchase beer that is stale and/or has defects in the eyes of the brewer.

Further study must be done, but this suggests that consumers are accustomed to and accept aged beer. North American lagers are always served cold (as done in this study) and consumers may not be able to distinguish, without a lengthy training period, the subtleties in the beer that the aging process may induce.

Difference (Triangle Tests)

Consumers could only distinguish a significant difference for Brand A samples stored for two weeks at 38°C versus the control. All other brand/storage conditions were not significantly different.

From both the preference and difference tests, is it clear that this group of consumers cannot distinguish the aged beer from the fresh beer (control). Although the accelerated storage times reflect those commonly practiced by industry, accelerated storage may not represent the actual aging process. Edwards van Muijen and McDaniel (2001) found that accelerated storage (38°C for 2 weeks) is not an exact portrayal of a longer storage time at lower temperature (27°C for 3 months), which represents the natural aging process of the beer. Bright and others (1993) found (American lager) staling compounds formed during storage varied considerably with test temperature, which confirms that accelerated storage testing may not give results similar to results obtained from actual shelf-life testing.

Conclusions

The consumers in this study did not have a preference across all brands for samples stored for 1 week at 38°C versus control nor for samples stored for 2weeks at 38°C versus control. The consumers only perceived a difference for Brand A samples stored for two weeks out of all the other brands and storage conditions. Further study needs to be performed to examine if consumers can perceive a difference with beer stored at conditions closer to the natural aging process.

V. THESIS SUMMARY

In the first study, a lexicon for aged beer aroma was successfully developed and tested with two major categories: *sulfury* and *sweet brown*. PCA was used to gain a broad understanding of how panelists used first tier descriptors over the entire range of samples within storage conditions. The second tier descriptors, analyzed by GPA, refined the broad categories of the first tier descriptors. Panelists used the descriptors "*skunky*" and "*perm solution*" to refine the *sulfury* group, the descriptors "*baked pineapple*" and "*honey*" to refine the *sweet brown* group, and the descriptor "*artificial fruit*" to refine the *fruity* group.

The trends of significant descriptors over storage time depended on brand and temperature condition. Evidence from MANOVA, ANOVA, PCA and GPA reveal that domestic lagers within the same brand at the two storage temperatures, 27°C and 38°C, behave differently. Although the 38°C storage may induce stale aromas, it is not an appropriate temperature to represent the natural aging process in beer. This study shows that when the beer is stored at 38°C for 8 days, the variation in the aroma intensity is considerably less than when the beer is stored at 27°C for 8 weeks.

In the second study, consumers did not indicate preferences across all brands for samples stored for 1 week at 38°C versus control or samples stored for 2-weeks at 38°C versus control. The consumers only perceived a difference for Brand A samples stored for two weeks out of all the other brands and storage conditions. Further study is needed to provide insights into whether consumers can perceive a difference with beer stored at conditions closer to the natural aging process.

BIBLIOGRAPHY

Axcell B and Torline P. 1998. Some alternate views on beer flavour. *Master* Brewers Association of the Americas Technical Quarterly 35 (2): 91-4

Back W, Forster C, Krottenthaler M, Lehmann J, Sacher B, Thum B, Weihenstephan. 1999. New research findings on improving taste stability. *Brauwelt International.* 5: 394-405

Bamforth CW. 1998. *Tap into the art and science of brewing*. New York, New York: Plenum Press. 245 pp.

Bamforth CW. 1999. The science and understanding of the flavour stability of beer: a critical assessment. *Brauwelt International* 2: 98-110

Barker RL, Gracey DEF, Irwin AJ, Pipasts P, Leiska E. 1983. Liberation of staling aldehydes during storage of beer. *Journal of the Institute of Brewing* 89: 411-5

Bright D, Patino H, Schroedl D, Nyarady S. 1993. Beer flavor stability improvement and correlations to carbonyl profile. *European Brewery Convention Congress* 413-420

Caul J. 1957. The profile method of flavor analysis Adv. Food Res 7: 1-40

Civille G and Lawless H. 1986. The importance of language in describing perceptions. *Journal of Sensory Studies* 1: 203-215

Civille G and Lyon B. 1996. Aroma and flavor lexicon for sensory evaluation. *ASTM data series publication DS 66* West Conshohocken, PA. 158 pp.

Clapperton JF. 1974. Profile analysis and flavour discrimination. Journal of the Institute of Brewing 80: 164-73

Clapperton JF. 1976. Ribes flavour in beer. Journal of the Institute of Brewing 82: 175-6

Dalgliesh CE. 1977. *Flavour stability*. Presented at European Brewery Convention Congress, Amsterdam, Netherlands

DeClerk J. 1934. Journal of the Institute of brewing 40: 4

DeMets M and Verzele M. 1968. The aroma of hops I. Origin and identification of hop aroma substances. *Journal of the Institute of Brewing* 74: 74-81

Devreux A, Blockmans C, van de Meerssche J. 1982. Carbonyl compounds formation during aging of beer. Presented at European Brewery Convention Congress, Copenhagen

Drost BW, van Eerde P, Hoekstra SF, Strating J. 1971. Fatty acids and staling of beer. Presented at European Brewery Convention Congress

Drost BW, Duidam J, Hoekstra SF, Strating J. 1974. Role of individual compounds in beer staling. *Master Brewers Association of the Americas Technical Quarterly* 11(2): 127-34

Drost BW, van den Burg R, Freijee FJM, van der Velde EG, Hollemans M. 1990. Flavor stability. American Society of Brewing Chemists 48(4): 124-31

Edwards van Muijen CV and McDaniel MR. 2001. Development of a lexicon for staling aromas in North American lager and analysis of consumer preference and difference. MS thesis. Oregon State University, Corvallis. 104 pp.

Evans D, Schmedding D, Bruinje A, Heideman T, King B, Groesbeek N. 1998. *Flavour impact of aged beers*. Institute of Brewing (Great Britain). Austrailian and New Zealand section, 25th convention

Fix G. 1989. Principles of brewing science. Boulder: Brewers Publications. 246 p.

Furusho S, Kobayashi N, Nakae N, Takashio M, Tamaki T, Shinotsuka K. 1998. A developed descriptive sensory test reveals beer flavor changes during storage. *Master Brewers Association of the Americas Technical Quarterly* 36(2): 163-6

Greenhoff K and Wheeler RE. 1981. Evaluation of stale flavour and beer carbonyl development during normal and accelerated aging utilizing liquid and high pressure liquid chromatography. Presented at European Brewery Convention Congress, London

Grigsby JH, Palamand SR, Davis DP, Hardwick WA. 1972. Studies on the reactions involved in the oxidation of beer. *American Society of Brewing Chemists*: 87-92

Guinard J-X, Uotani B, Mazzucchelli R, Taguchi A, Masuoka S, Fujino S. 2000. Consumer testing of commercial lager beers in blind versus informed conditions: Relation with descriptive analysis and expert quality ratings. *Journal of the Institute of Brewing* 106(1): 11-9

Guinard J-X, Yip D, Cubero E, Mazzucchelli R. 1999. Quality ratings by experts, and relation with descriptive analysis ratings: a case study. *Food Quality and Preference* 10: 59-67
Hashimoto N. 1972. Oxidation of higher alcohols by melanoidins in beer. Journal of the Institute of Brewing 78: 43-51

Hashimoto N and Eshima T. 1977. Composition and pathway of formation of stale aldehydes in bottled beer. *Journal of the American Society of Brewing Chemists* 35(3): 146-50

Hashimoto N and Eshima T. 1978. Oxidative degradation of isohumulones in relation to flavour stability. *Journal of the Institute of Brewing* 85: 136-40

Hunter B. 1996. More informative beer labels. *Consumers' Research Magazine* 79(10): 10-15

Irwin AJ, Barker RL, Pipasts P. 1991. The role of copper, oxygen, and polyphenols in beer flavor stability. *Journal of the American Society of Brewing Chemists* 49(3): 140-9

Kaltner D, Thum B, Forster C, Back W, Freising-Weihenstephan. 2001. Investigations into technological and flavor effects in beer. *Brauwelt International* 19(1): 40-45

Kamimura M and Kaneda H. 1993. Shelf-life of beer. *Shelf-life studies of foods and beverages*, ed. G Charalambous, pp. 821-89. Amsterdam: Elsevier Science Publishers B.V.

Kaneda H, Kobayashi N, Furusho S, Sahara H, Koshino S. 1995. Reducing activity and flavor stability of beer. *Master Brewers Association of the Americas Technical Quarterly* 32(2): 90-94

Khermouch G, Jensen T. 1996. A-B pushes freshness. Adweek Eastern Edition 37(34): 4

Kunze W. 1996. Technology brewing and malting. Berlin: VLB

Lermusieau G, Noel S, Liegeois C, Collin S. 1999. Nonoxidative mechanism for development of *trans*-2-nonenal in beer. *Journal of the American Society of Brewing Chemists* 57(1): 29-33

Lewis M and Young T. 1996. Brewing. London: Chapman & Hall. 260 pp.

Lindsay RC. 1974. Sampling designs and experimental procedures for staling flavor studies. *Master Brewers Association of the Americas Technical Quarterly* 11(2): 101-3

Lynch PA and Seo CW. 1987. Ethylene production in staling beer. *Journal of Food Science* 52(5): 1270-2

Meilgaard M. 1972. Stale Flavor Carbonyls in Brewing. *The Brewers Digest*: 48-57

Meilgaard M. 1975a. Flavor chemistry of beer part 1: Flavor interaction between principal Volatiles. *Master Brewers Association of the Americas Technical Quarterly* 12(2): 107-17

Meilgaard M. 1975b. Flavor chemistry of beer part 2: Flavor and threshold of 239 aroma volatiles. *Master Brewers Association of the Americas Technical Quarterly* 12(3): 151-68

Meilgaard M. 1982. Prediction of flavor differences between beers from their chemical composition. Journal of Agriculture and Food Chemistry 30: 1009-17

Meilgaard M. 1991. The flavor of beer. Master Brewers Association of the Americas Technical Quarterly 28: 132-41

Meilgaard M, Ayma M, Ruano JI. 1971. A study of carbonyl compounds in beer. IV. Carbonyl compounds identified in heat treated wort and beer. *American Society of Brewing Chemists*: 219-29

Meilgaard M, Dalgliesh CE, Clapperton JF. 1979. Beer flavor terminology. American Society of Brewing Chemists 37(1): 47-52

Meilgaard M, Reid DS, Wyborski KA. 1982. Reference standards for beer flavor terminology system. *American Society of Brewing Chemists* 40(4): 178-87

Meilgaard M and Muller JE. 1987. Progress in descriptive analysis of beer and brewing products. *Master Brewers Association of the Americas Technical Quarterly* 24: 79-85

Meilgaard M, Civille G, Carr BT. 1991. Sensory Evaluation Techniques. Boca Raton, Florida: CRC Press

Mercredy JM, Sonnemann JC, Lehmann SJ. 1974. Sensory profiling of beer by a modified QDA method. *Food Technology*: 36-41

Moskowitz H and Krieger B. 1992. What sensory characteristics drive product quality? An assessment of individual differences. *Journal of Sensory Studies* 8: 271-282

Noonan GJ. 1996. *New Brewing Lager Beer*. Boulder, Colorado: Brewers Publications, division of the association of Brewers

Ogane O, Yokoyama F, Hirano T. 2000. Quantification of beer freshness, based on the original freshness scale. *Master Brewers Association of the Americas Technical Quarterly* 37(1): 69-72

Palamand SR, Hardwick WA, Davis DP. 1970. Studies of some carbonyl-amine reactions and their significance to beer flavor. *American Society of Brewing Chemists*: 56-62

Papazian C. 1993. *Evaluating Beer*. Boulder, Colorado: Brewers Publications. 238 pp.

Peacock VE, Dienzer ML, McGill LA, Wrolstad RE. 1980. Hop Aroma in American Beer. Journal of Agriculture and Food Chemistry 28(4): 774-7

Peacock VE, Deinzer ML, Likens ST, Nickerson GB, McGill LA. 1981. Floral hop aroma in beer. *Journal of Agriculture and Food Chemistry* 29: 1265-9

Sakuma S, Amano H, Ohkochi M. 2000. Identification of off-flavor compounds in beer. *American Society of Brewing Chemists* 58(1): 26-9

Sanchez NB, Lederer CL, Nickerson GB, Libbey LM, McDaniel MR. 1992. Sensory and analytical evaluation of beers brewed with three varieties of hops and unhopped beer. In *Food Science and Human Nutrition.*, ed. Charalambous, pp. 403-26. New York: Elsevier Science Publishers

Schmitt DJ and Hoff JT 1979. Use of graphic linear scales to measure rates of staling in beer. *Journal of Food Science* 44(3): 901-4

Senstools v. 2.3 1997. Guelph, Ontario, Canada.

Sharpe FR and Laws DR. 1981. The essential oil of hops: A review. Journal of the Institute of Brewing 87: 96-107

Siebert KJ. 1994. Sensory analysis of hop oil-derived compounds in beer; Flavor effects of individual compounds. Quality control. Presented at the European Brewery Convention Hop Symposium, Zoeterwoude, May 31-June 1.

Siebert KJ, Ramus SA, Peppard TL, Guzinski JK, Stegink LJ. 1989. An investigation into hop fraction leading to hoppy aroma in beer. *Master Brewers* Association of the Americas Technical Quarterly 26: 62-9

SPSS v. 10.1. 2000. SPSS Inc., Chicago, IL

Stenroos LE. 1973. The effects of time, temperature, and air on various finished beer components. *American Society of Brewing Chemists Proceedings*: 50-6

Stone H and Sidel J. 1993. Sensory Evaluation Practices. Academic Press 338pp.

Stone H, Sidel J, Oliver S, Woolsey A, Singleton RC. 1974. Sensory evaluation by quantitative descriptive analysis. *Food Technology* 28(2) 24-34

Stucky G. 1996. Aroma Qualities of Raw Hops and Hops in Beer By trained Panel Free-Choice Profiling. MS thesis. Oregon State University, Corvallis, OR. 104 pp.

Stucky G and McDaniel M. 1997. Raw hop aroma qualities by trained panel freechoice profiling. *Journal of the American Society of Brewing Chemists* 55(2): 65-72

Tressl R, Friese L, Fendesack F, Koppler H. 1978. Gas-chromatographic-mass spectrometric investigation of hop aroma constituents in beer. *Journal of Agriculture and Food Chemistry* 26(6): 1422-32

Uchida M and Ono M. 1996. Improvement for oxidative flavor stability of beer-role of OH-radical in beer oxidation. *Journal of the American Society of Brewing Chemists* 54(4): 198-204

Whitear AJ, Carr BT, Crabb D, Jaques D. 1979. *The challenge of flavor stability*. Presented at European Brewery Convention Congress, Berlin

Willaims A and Langron S. 1984. The use of free-choice profiling for the evaluation of commercial ports. J. Sci. Food Agric. 35:558-568

Williams RS and Wagner HP. 1979. Contribution of hop bitter substances to beer staling mechanisms. *American Society of Brewing Chemists* 37(1): 13-9

Yang X, Lederer C, McDaniel M, Deinzer M. 1993. Chemical Analysis and sensory evaluation of hydrolysis products of humulene epoxides II and III. *Journal of Agriculture and Food Chemistry* 41: 1300-1304

APPENDICES

More taste test news from the Sensory Science Lab at OSU!

YOU MUST BE OVER 21 YEARS OF AGE TO PARTICIPATE IN THIS STUDY. This test will run Monday, April 23rd, Wednesday, April 25th and Friday April 27th from 10am until 5pm in 20 minute increments. If you qualify you may sign up for all three days. Please include the days and times you prefer with your response. You will receive a gift certificate to either New Morning Bakery, The Great Harvest, StarBucks Cafe, The Beanery, Tarntip Thai Restaurant, Regal Cinemas, Togo's or American Dream Pizza, Noah's Bagel, Crystal's, Hollywood Videos. (Testing will last about 20 minutes.)

Would you like to participate? If yes, please reply to the screener below by using the 'reply' function on your e-mail and type a YES response after those choices that apply. (If you prefer to use the telephone, please call us at 737-6508 or 737-6506 and you will be screened over the phone.) We will contact you to let you know whether or not you qualify.

Remember choosing ALL the answers may DISQUALIFY you from this test.

Thank you for your time Sensory Lab

YOUR NAME: DAY-TIME PHONE #:

- 1. Do you drink beer?
 - a) YES
 - b) NO
- 2. What style(s) of beer do you drink most often? (Choose a maximum of 3)a) Ale
 - b) Lager
 - c) Porter
 - d) Stout
 - e) Light Beer
 - f) Non-Alcoholic
 - g) Other_____

APPENDIX 1 SCREENER (continued)

- 3. Which brands of beer do you drink? How many times per month? Please indicate by writing the number of occasions per month beside the brand(s) you drink.
 - a) Corona
 - b) Budweiser
 - c) Deschutes
 - d) Widmere
 - e) Miller
 - f) Guinness
 - g) Sierra Nevada
 - h) Coors
 - i) Full Sail
 - j) Pabst Blue Ribbon
 - k) Milwaukee's Best
 - 1) King Fischer
 - m) Pacific Crest
 - n) Henry Weinhardts
 - o) Other
- 4. What is your present age?
 - a) 21-30 years
 - b) 31-40 years
 - c) 41-50 years
 - d) 51-60 years
 - e) Over 60 years

If you qualify please let us know what time you would like to come between 10am and 5pm in 20 minute increments: (write in time preference e.g. 10-10:20, 10:20-10:40, 10:40-11:00)

Monday April 23 rd	a)	b)
Wednesday April 2	$(5^{\text{th}} a)$	b)
Friday April 27 th	a)	b)

PLEASE BE SURE TO BRING IDENTIFICATION OR YOU WILL NOT BE ALLOWED TO PARTICIPATE.

APPENDIX 2 CONSENT FORM

Oregon State University Department of Food Science & Technology Sensory Science Lab

Alcoholic Beverage Consent Form

Project Director: Dr. Mina McDaniel (541) 737-6507

Product:American Lager BeerIngredients:Water, malted barley, hops, yeast, wheat, corn, rice

The Sensory Science Lab at Oregon State University engages in product testing and evaluation research. As a participant in such research, you may be offered the opportunity to sample small quantities of alcoholic beverages. You are never required to sample such beverages, nor are you required to finish any beverage you elect to sample. Indeed, the decision as to whether to sample any product offered during the research is yours alone, and you alone should determine how much of the sample you wish to consume. You may withdraw from this study at anytime without penalty. You will be assigned a subject number in order to keep your identity confidential.

The Sensory Science Lab considers the health and safety of research participants and the public to be of utmost importance. Therefore, you should refrain from sampling any alcoholic beverage offered as part of this research if you have been advised by your doctor or if you have any medical reason to refrain from consuming alcoholic beverages (beer, wine or distilled spirits.)

You should also refrain from sampling any alcoholic beverage on a given day if:

- You have consumed any beer, wine or distilled spirits on that day.
- You are taking any prescription or over-the-counter (non-prescription) medication and you have been advised by your doctor or the label, or instructions state that you should refrain from consuming alcoholic beverages while taking the medication.

In addition, Federal Law requires that alcoholic beverage labels contain the following statement:

GOVERNMENT WARNING:

- 1. According to the Surgeon General, women should not drink alcoholic beverages during pregnancy because of the risk of birth defects.
- 2. Consumption of alcoholic beverages impairs your ability to drive a car or operate machinery and may cause health problems.

Furthermore, you should follow your doctor's advice if you are pregnant, attempting to become pregnant or nursing.

You may be asked by the person conducting the research in which you are participating or by another facility personnel to remain at this facility for a period of time after your last sampling of an alcoholic beverage. Moreover, if you appear to be impaired at the end of such time period, you will be provided with an alternative means of transportation to your home and arrangements will be made for your to return at a later date for your car, at the Sponsor's expense.

APPENDIX 2 CONSENT FORM (continued)

By signing this you acknowledge that you have read the ingredient list and certify that you have no known allergies to the ingredients listed above. You acknowledge and agree that you have read this Statement of Policy and fully understand its contents. You further acknowledge that you are 21 years or older. Please contact the project director with any questions concerning this study or specific procedures. Please contact the Institutional Review Board (IRB) Coordinator with any questions concerning your rights as a research participant at (541) 737-3437 or via e-mail at mailto:IRB@orst.edu<>. My signature below indicates that I have read and understand the procedures described above and give my informed and voluntary consent to participate in this study. I understand that I will receive a signed copy of this consent form.

Name of Participant: (please prin	nt)Age:
Signature of Participant:	Date:

APPENDIX 3 PREFERENCE AND TRIANGLE TEST BALLOTS

Panelist #_____

Date_____

PREFERENCE TEST

Please taste the product on the left first. Taste the product on the right second.

Now that you've tasted both products, which one do you prefer? Please circle the three digit code associated with the sample you prefer.

THANK YOU!

Please open door or notify server that you are ready for the next samples.

Panelist #_____

Date_____

TRIANGLE TEST

Taste the samples from left to right. Two samples are identical; one is different. Circle the three digit code associated with the ODD/DIFFERENT sample.

THANK YOU!

Please open door or notify server that you are ready for the next samples.

Demographic Questionnaire

- 1. 1. What style(s) of beer do you drink most often? (Choose a maximum of 3) □ Ale

 - □ Lager D Porter
 - □ Stout

 - □ Light Beer
 - □ Non-Alcoholic
 - Other
- 2. Which brands of beer do you drink? How many times per month? Please indicate by writing the number of occasions per month beside the brand(s) you drink.

Br	ands_	# Occasions per month
	Corona	
	Budweiser	
	Deschutes	
	Widmere	
	Miller	
	Guinness	
	Sierra Nevada	
	Coors	
	Full Sail	
	Pabst Blue Ribbon	
	Milwaukee's Best	
	King Fischer	
	Pacific Crest	
	Henry Weinhardts	
	Other	

- 3. What is your present age?
 - □ 21-30 years
 - □ 31-40 years
 - □ 41-50 years
 - □ 51-60 years
 - Over 60 years
- 4. Please indicate your gender:
 - □ Male
 - **D** Female

Panelist Number

APPENDIX 5 DEMOGRAPHIC INFORMATION

For Brand A, there were 79 consumer panelists, 61 (77%) males and 18 (23%) females. Table 5.1 shows the age ranges, number of panelists in the age range and percentage of total.

Table 5.1 Panelist age information for Brand A

Age Range	# of panelists	% of total
21-30	52	66
31-40	11	14
41-50	10	12
51-60	5	6
61 +	0	0

For Brand B there were 99 panelists, 58 (58%) males, 40 (40%) females and three panelists who did not volunteer their personal information. Table 5.2 shows the age ranges, number of panelists in the age range and percentage of total.

Table 5.2 Panelist age information for Brand B

Age Range	# of panelists	% of total
21-30	47	47
31-40	16	16
41-50	18	18
51-60	12	12
61 +	3	3

APPENDIX 5 DEMOGRAPHIC INFORMATION (continued)

For Brand C there were 93 panelists, 58 (62%) males, 33 (35%) females and two panelists who did not volunteer their personal information. Table 5.3 shows the age ranges, number of panelists in the age range and percentage of total.

Table 5.3 Panelist age information for Brand C

Age Range	# of panelists	% of total
21-30	49	53
31-40	12	13
41-50	18	19
51-60	12	13
61 +	0	0