

## ABSTRACT

### **KRAUSE, ANDREA JEAN. Evaluation of Consumer Acceptance and Storage Stability of Butter. (Under the direction of Dr. MaryAnne Drake.)**

Consumers value the rich flavor and smooth texture of butter. With variations due to processing, storage conditions, addition of starter culture, and salt, there are many diverse flavor profiles, textures, and colors of butter. A better understanding of the key drivers of butter and vegetable oil spread purchase may aid in identification of marketing strategies. Sweet cream butter in the United States is often stored for up to two years in refrigerated or frozen storage in bulk (25 kg) and 454g packages of four sticks (butter quarters). Deterioration of flavor and texture may occur during this time. No comprehensive studies have been published that compare bulk and stick over time using sensory analysis and analytical techniques. The objectives of this study were to explore consumer preferences for butters and margarines/spreads and to evaluate the flavor and texture stability of bulk and stick butter across frozen (-20C) and refrigerated (5C) storage.

To understand consumer acceptance of butter, a trained descriptive panel evaluated 29 commercial butters and spreads using a defined sensory language. Two focus groups were conducted with butter consumers to gain an understanding of usage and consumption habits. Eight representative butters and spreads were selected for consumer acceptance testing. Both internal and external preference mapping techniques were applied to interpret consumer data. Five consumer clusters with distinct butter/spread likes and dislikes were identified. Butter acceptability varied among consumers and butters with specific sensory characteristics could be marketed to specific target market segments.

Currently no industry specifications exist for butter storage. In the storage study, butter stability at 5C and -20C over a 15 month period was examined. Changes were monitored through descriptive sensory analysis of flavor, texture, and color by a trained panel using a defined sensory language. Additionally, chemical changes were evaluated by oxidative stability index, peroxide value, free fatty acid value, vane, instrumental color, differential scanning calorimetry, and oil turbidity.

The data indicated that refrigerated butter quarters showed the fastest decline in quality; refrigerator/stale flavor was detected after 6 months. When frozen at -20C, sticks can be stored for up to 12 months. It is still advantageous for manufacturers to continue to store butter in large blocks. For bulk butter in refrigerated conditions, flavor quality is maintained for at least 9 months. In frozen storage, bulk butter can be stored in excess of 15 months without flavor detriment. While freezing may not completely stop lipid oxidation, it will maintain the flavors of freshly produced butter (milkfat, cooked/nutty) longer. Since butter is such a highly prized fat source in terms of its flavor and textural properties, it is important that manufacturers understand how long their product can be stored before negative attributes develop. These off-flavors could potentially carry-through to applications and negatively impact consumer perception.

**EVALUATION OF CONSUMER ACCEPTANCE AND STORAGE  
STABILITY OF BUTTER**

by

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of North Carolina State University in partial fulfillment of  
the requirements for the Degree of Master of Science.

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## **BIOGRAPHY**

Andrea Jean Krause was born in Madison, Wisconsin on June 30, 1982 to James and Judith Krause. She has one older sister, Jennifer.

Andrea graduated from McFarland High School in 2000. She had always had a keen interest in food, mixing ingredients and spices in her mother's kitchen for fun for as long as she can remember. Her interest in food as a legitimate science was piqued as a freshman at the University of Wisconsin—Madison when she took an introduction to Food Science course. Andrea became very involved in the Food Science Club after declaring Food Science her major sophomore year. She also worked as a lab assistant in both the Animal Science and Food Science departments, and attributes these experiences as driving forces in her desire to continue on to graduate school. Andrea completed two industry internships with McCain Snackfoods Inc. in Appleton, WI in 2003 and Kraft Foods Inc. in Glenview, IL in 2004.

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**CHAPTER 1**

**LITERATURE REVIEW**

## INTRODUCTION

The existence of butter has been recorded as early as 2000 BC (Douglas, 2004). This concentrated milkfat product has also become an important product in modern times. Recent concerns over obesity, dairy prices, and other factors have resulted in per capita consumption as well as production to remain relatively constant from 2000-2004 (IDFA, 2005).

The standard of identity of butter stipulates that it must contain at least 80% milkfat (USDA, 1989). This milkfat component has many unique properties. It contains more than 400 fatty acid varieties, of which 66% are saturated, 30% are monounsaturated, and 4% are polyunsaturated. Stages of lactation, diet, dietary supplementation, season of the year, and disease all have an impact on the fatty acid composition of milkfat. Stability, color, nutritional, and rheological properties of the final product can all vary depending on the fatty acid composition (Fox, 2000, Hawke and Taylor, 1994, Nickerson, 1995).

Texture is one of the valued properties of butter. Size and shape of fat crystals, interactions between crystallized fat and liquid oil, fatty acid composition, and temperature treatment of the cream during processing all have an effect on the texture profile of butter. Blending and work-softening, and milkfat fractionation can be used to modify the texture of butter for increased cold spreadability. A plethora of analytical techniques have been used to evaluate the texture of butter and margarine-type products. These methods include: penetrometry, sectility, compression, extrusion, vane, dropping-point, DSC, and texture profile analysis (Fearon and

Johnston, 1989, Voisey, 1976). Sensory evaluation of butter texture has also been conducted.

Aside from its texture, butter is prized for its rich flavor. Many of the compounds that play a role in the characteristic flavor of butter have been identified although there is currently no chemical mixture that has been able to flawlessly replicate the aroma of butter (Peterson and Reineccius, 2003). Diacetyl, lactones, short-chain fatty acids, lipid-derived aldehydes, and dimethyl sulfide are all known to contribute to butter flavor. Many instrumental techniques have been utilized to isolate compounds associated with butter flavor (Wampler 2002, Povolo and Contarini, 2003, Reid, 2003, Peterson and Reineccius, 2003).

Sensory analysis and USDA grading have been employed as other methods to evaluate butter flavor. Previous studies have relied on grading, product specific scaling, and vague terminology to compare products or evaluate quality of butter and margarine/spread products. No published lexicon currently exists for descriptive sensory analysis of butter flavor. The Spectrum<sup>TM</sup> method was developed to allow attributes and products to be compared on a universal 15-point scale by trained panelists. It has been used to characterize the flavors of many products including cheese, peanut butter, chocolate milk and many others (Young, Drake, Lopetcharat, & McDaniel, 2004; McNeill, Sanders, & Civille, 2002; Thompson, Drake, Lopetchararat, & Yates, 2004). There have been no published studies, which use the Spectrum<sup>TM</sup> method to characterize butter flavor using a defined sensory language.

The wide varieties of butters and margarines/vegetable oil spreads on the market have very different flavor profiles, and consumers use them differently.

Numerous studies have evaluated consumer acceptance of margarine spreads and butter/oil blends (Avramis, et al., 2003, Chen, et al., 2004, Kim, et al., 2005, Michicich, et al., 1999). No current studies have attempted to link consumer attitudes toward the varying flavors exhibited among butters and compare them to margarine or vegetable oil spreads using a descriptive sensory language. Understanding what flavor attributes certain segments of butter and margarine/vegetable oil spread consumers prefer will help manufacturers develop marketing strategies and products to best accommodate these market segments.

Dairy manufacturers produce large amounts of butter in the winter months due to a surplus of milkfat. It is often necessary to store this butter for extended periods of time until there is a demand for it. During refrigerated and/or frozen storage, degradation of quality may occur. This is an important issue when companies develop specification sheets for butter suppliers or when butter suppliers design storage regimes. Butter is commonly stored for extended periods in blocks (25 kg) which are subsequently re-worked into quarter-pound sticks. However, retail packages (sticks or quarters as they are referred to by industry) are often stored for extended periods.

A variety of studies have been conducted stability of butter in various wrapping materials (Emmons et al., 1986; Tomlinson and Dixon, 1977; MacBean, 1974; Downey and Murphy, 1968; Pont, 1961). Recent studies have not addressed butter stability in both refrigerated and frozen storage. Further, stability of bulk and stick butter have not been compared. The objectives of this study were two-fold. In the first objective, the consumer preferences for butters and margarines/spreads were

explored through preference mapping. The second objective was to evaluate the flavor and texture stability of bulk and stick butter across frozen (-20C) and refrigerated (5C) storage. Descriptive sensory analysis, which has not been previously applied to butter, was used to monitor flavor and texture. Instrumental methods were also used to evaluate chemical and texture changes.

## LITERATURE REVIEW

### 1.1 Milkfat

Fresh bovine milk is composed on average of 4% milkfat (Walstra, et al., 1999). Butter, fluid milk, and other milkfat products contain varying levels of milkfat (Dairy Management Inc., 1996). The bulk of milkfat composition (98%) is triglycerides (Fox, 2000, Fox and McSweeney, 1998, Walstra, et al., 1999). The remaining portion is comprised of phospholipids, minor amounts of diglycerides, monoglycerides, cholesterol, cholesterol esters, minute amounts of fat-soluble vitamins (A, D, E, and K) and other lipids (Fox, 2000, Fox and McSweeney, 1998, Riel, 1985). In recent times, diets high in lipids have been implicated in cardiovascular diseases (Berner, 1993, Mondello, et al., 2004), and milkfat has been closely examined since it is a major contributor to the saturated fat in American diets. In 1988, butter alone was thought to contribute 3.4% total fat, 6.3% saturated fat, and 2.9% cholesterol in the overall diet (Berner, 1993).

#### 1.1.1 Properties

Physical properties of milkfat are a result of the interactions between the solid and liquid fat phases and are largely dependent on temperature (Dairy Management Inc., 1996). These properties are important for the formulation of products which utilize milkfat as an ingredient (Neville and Jensen, 1995). Below  $-40^{\circ}\text{C}$ , milkfat is entirely crystallized. Up to  $40^{\circ}\text{C}$  the fat is a mixture of oil and fat crystals (Jensen and Clark, 1988). In some cases, a crystal inside the globule becomes so large that it pierces the membrane. If this globule bumps into another and that membrane is pierced by the protruding piece of fat crystal, the two will become partially coalesced.

Since it is unable to fully coalesce, it is called a granule (Walstra, et al., 1999). The majority of fat globules are from 1 to 5 $\mu$ M in diameter (Jensen and Clark, 1988, Keenan and Patton, 1995). In addition to a unique melting profile, milkfat is used in foods to function as an emulsifier, contributes to color, body, viscosity, aeration, texture, and provides structure (Dairy Management Inc., 1996).

### **1.1.2 Milk Fat Globule Membrane**

In raw milk, droplets of milkfat are surrounded by a membrane composed of proteins and phospholipids referred to as the milkfat globule membrane (MFGM) (Table 1.1) (Corredig and Dalgleish, 1998, Keenan and Patton, 1995). Milk is secreted from secretory cells in the mammary gland. The surrounding MFGM is derived from the apical membrane of this secretory cell (Kanno, 1989, Muir, 1998). The outer structure of the membrane is thought to have a lipid bilayer with scattered proteins that jet out into the milk plasma (the remaining portion of the milk that is not fat). Below this bilayer, there is a covering of proteins and then another lipid layer (Figure 1.1). The precise structure of the MFGM is not clearly understood because a large part of the structure is lost during and after the milk is secreted as well as during contact with air. It is important to note that the composition of the MFGM is more similar to that of a cell membrane than to that of milkfat or milk plasma. The MFGM contains a high level of phospholipids (50%) in addition to a large amount of triglycerides (McPherson and Kitchen, 1983). In comparison to milkfat triglycerides, those in the MFGM contain more long-chain fatty acids. The membrane also contains many unsaturated fatty acids and as many as 10 major constituents and

several minor components (Keenan and Patton, 1995, Walstra, et al., 1999) (Table 1.2).

Although it only composes 1% of milk proteins (Innocente, et al., 1997, McPherson and Kitchen, 1983), MFGM proteins are essential for the stability of the fat globules (Fox and McSweeney, 1998, Keenan and Patton, 1995). Because of its composition, the MFGM plays an important role in reducing the interfacial tension between the fat and the aqueous phase of milk to prevent coalescence. Therefore, it acts as an emulsifier which enables the fat globules to maintain membrane shape while remaining suspended in the aqueous phase of the milk (Corredig and Dalgleish, 1998, Fox and McSweeney, 1998, Innocente, et al., 1997, McPherson and Kitchen, 1983). The membrane also fulfills another important function-- it guards the inner fat droplet against lipolysis by native milk lipoprotein lipase (Fox and McSweeney, 1998). Even if there is slight damage to the membrane, the droplet can remain intact. But if this damage occurs, lipoprotein lipase may hydrolyze the milk fat resulting in the release of free fatty acids which cause rancid off-flavors and odors (Muir, 1998). In the process of butter-making, which is an intentional destabilization of the fat globule, the result is the coalescence of the fat into a solid mass. A large majority of the MFGM present is flushed away with the buttermilk (Corredig and Dalgleish, 1998).

Due to its ability to keep milkfat suspended in the aqueous phase of milk (contains both hydrophilic and hydrophobic parts), MFGM has been isolated and used to stabilize emulsions (Kanno, 1989, Riel, 1985). The MFGM can be used in some foods as an emulsifying agent (Innocente, et al., 1997). In addition, the phospholipids

in the membrane are touted as having health benefits. The buttermilk that results from butter manufacture is an excellent source of this beneficial fat (Corredig, et al., 2003, Riel, 1985). Until recently this was an untapped source, but through the use of membrane filtration, concentration of MFGM has become of interest (Corredig, et al., 2003).

### **1.1.3 Fatty Acid Composition**

A wider variety of fatty acids can be obtained from milkfat than from any other source (Christie, 1994). Many of the more than 400 fatty acids in milkfat from cow's milk exist only in minor amounts (Douma, 2004, Fox, 2000, Fox and McSweeney, 1998, Jensen and Clark, 1988) (Table 1.3). Six million triglyceride combinations can be made from these 400 fatty acids (Jimenez-Flores, 1997). Of these 400, about 66% are saturated, 30% monounsaturated, and 4% polyunsaturated (Baer, 1996). Bovine milk contains a greater variety of fatty acids than other mammary species. Human milk, in comparison, contains 184 fatty acids (Fox and McSweeney, 1998).

There are many things that can impact the variety of fatty acids and in turn change properties of the final product made from the milkfat (Fox, 2000, Nickerson, 1995). Stage of lactation, diet, dietary supplementation, season of the year, and disease, all have an impact on the fatty acid composition of the milkfat. The stability, color, nutritional, and rheological properties of the final product all can vary depending on the fatty acid composition (Fox, 2000, Hawke and Taylor, 1994, Nickerson, 1995). Although it would be desirable for a consistent product, it is

virtually impossible to standardize the fatty acid composition of the milkfat because so many factors affect its makeup (Fox, 2000).

The main long-chain fatty acids in milkfat are C16:0 (palmitic acid), C18:0 (stearic acid), and C18:1 (oleic acid) (Fox, 2000, Harding, 1995). Bovine milkfat also contains moderate amounts of medium-chain fatty acids, C10:0 (capric acid), C12:0 (lauric acid), and C14:0 (myristic acid) (Riel, 1985). Ruminant milkfat is unique in that it contains short-chain fatty acids, butyric acid (C4:0) is the most prevalent of the short-chain fatty acids and often identified with butter due to its characteristic impact on flavor (Douglas, 2004). In comparison to plant oils, milkfat contains a low amount of polyunsaturated fatty acids (PUFA), (Fox, 2000) and monounsaturated fatty acids (MUFAs) (Smith, et al., 1978).

Oleic acid (C18:1) is both the most prevalent monounsaturated fatty acid (MUFA) and the overall most prevalent fatty acid in milkfat, comprising 20-30% of the total by weight (Fearon, 2001, Goff and Hill, 1993, Harding, 1995). Aside of increased oxidative stability, MUFAs have potential health benefits—lowering low density lipoprotein (LDL) while unchanging high density lipoprotein (HDL) levels (Fearon, 2001). Many studies have shown that a diet of unprotected (non-encapsulated) lipids, including, but not limited to: coconut oil, cottonseed oil, marine oil, oleic acid, safflower seeds, soyabean oil, soyabeans, and sunflower seeds, increases the amount of beneficial MUFAs contained in milkfat (Baer, 1996, Fearon, 2001, Hawke and Taylor, 1994). Oleic acid and another prevalent triglyceride, stearic acid (C18:0), are both manifested from blood chylomicrons (Fox, 2000).

Even though the diet of a ruminant animal is high in polyunsaturated fatty acids (PUFAs), a low level is found in the milkfat due to microorganisms present in the gut of the animal (Fox, 2000). Only 5% of milkfat fatty acids are of the PUFA variety, and the majority of these come from phospholipids (Jensen and Clark, 1988). Such a small amount of PUFAs make it through to the milk itself because organisms in the rumen hydrogenate the fatty acids. Contrary to the dietary method used to increase MUFAs, fat encapsulation is the only known way to avoid hydrogenation and increase the amount of unsaturated fats in milkfat (Fox and McSweeney, 1998, Jensen and Clark, 1988). The encapsulation allows the fatty acids to be absorbed in the intestine and avoid hydrogenation (Jensen and Clark, 1988). A benefit aside from the health aspects of PUFAs, is the reduced melting point in butter that comes with increased PUFA concentrations (Fox and McSweeney, 1998, Jensen and Clark, 1988).

One specific PUFA that has recently been shown to have potential health benefits is conjugated linoleic acid (CLA, C18:2). The specific isomer of interest is *cis-9, cis-12*-octadecadienoic acid (MacDonald, 2000, Parodi, 1999). Fat from ruminant animals contains larger amounts of CLA than that of non-ruminant animals. Furthermore, lipids from animals that are pasture-fed have a greater amount of CLA than animals that are fed both grain and from pasture (Fearon, 2001). CLA is produced in the rumen as a result of bacteria, which hydrogenate linoleic acid using linoleic isomerase (MacDonald, 2000, Parodi, 1999). Health benefits of CLA are still under investigation, but animal studies have shown promising results (Fearon, 2001, MacDonald, 2000, Parodi, 1999). CLA may act as an antioxidant to inhibit cancerous

tumor growth and slow the development of fatty lesions in arteries (Parodi, 1999). Research is also being conducted to determine if the primary method for anti-cancer effects of CLA is a boost to the immune system. CLA has also been touted to reduce body fat and increase bone mass (Baer, et al., 2001, Fearon, 2001, MacDonald, 2000). It is important to note these findings have not been studied extensively in humans and that many good sources of CLA (beef, milkfat, lamb) are high in fat which may pose other health risks (Fearon, 2001, MacDonald, 2000).

Only ruminant animals produce milkfat that contains the short-chain fatty acid butanoic acid. This fatty acid in the milkfat results from the breakdown of carbohydrates by bacteria in the rumen. Because of this, the butanoic acid content of the fat depends largely on the diet of the animal. The resulting carbohydrate breakdown product,  $\beta$ -hydroxybutyrate becomes incorporated into the milkfat (Fox, 2000, Fox and McSweeney, 1998). The presence of butanoic acid and other short-chained fatty acids is used as a way to test for adulteration of butter with other fats (Fox and McSweeney, 1998, Harding, 1995). Butanoic acid is also most often associated with fatty-acid type flavors in butter (Douma, 2004).

## **1.2 Milkfat Products**

### **1.2.1 Butter**

Butter is defined by the USDA (1989) as the product that is “made exclusively from milk or cream, or both, with or without common salt, and with or without additional coloring matter, and containing not less than 80% by weight of milkfat.” Twenty-one pounds of fresh milk are necessary to produce one pound of butter (Douma, 2004). The churning and working of milkfat results in the disruption of the

MFGM which in turn destabilizes the fat globules and results in their coalescence (Fox, 2000). Eventually, a partially crystalline mass of aggregated fat results (Walstra, et al., 1999). The resulting butter is a water in oil emulsion--an inversion of the cream (oil droplets suspended in water). It is important that the water droplets be smaller than 10 $\mu$ m and well dispersed or adverse effects to the quality and microbial growth could result (Precht, 1988). Butter was largely manufactured using a batch method up until the 1940's when continuous methods became more economically sound (Bruhn, 2004). Although butter consumption per capita is relatively stagnant due to increasing concerns about obesity, dairy prices, and other factors, butter production in the United States rose 9.2% in 1999 to total 1.28 billion pounds (Bruhn, 2004).

### **1.2.2 Anhydrous Milkfat (AMF)**

Anhydrous butter oil, anhydrous butterfat, and anhydrous milk fat (AMF) can all be used interchangeably to describe the product that consists of highly concentrated milkfat from either butter or cream (Caric, 1994, Whittier, 1970). This product is relied upon heavily in hot climates due to its extended shelf life compared to butter as well as in chocolate manufacture, processed foods, ice cream, and baby food (Caric, 1994). The International Dairy Federation is responsible for the quality standards for milkfat composition (Caric, 1994). The milkfat content of AMF must be at least 99.8%, it must contain no more than 0.1% moisture, and 0.3% free fatty acids expressed as oleic acid (Caric, 1994, Whittier, 1970). In addition, there are strict standards for copper, iron, peroxide value, neutralizing agents, and microorganisms (Caric, 1994). If the cream is well pasteurized and properly cooled,

rancidity can be prevented. Off-flavors from oxidation can be avoided by excluding copper, iron, light, and air from the product (Boudreau and Saint-Amant, 1985).

Anhydrous butter oil and AMF are made via two different processes.

Anhydrous butter oil is made from butter that has been melted back down into oil (Caric, 1994, Walstra, et al., 1999, Whittier, 1970). After the butter is melted (salted butter is washed and overly acidic butter is neutralized), it is further heated.

Afterward, it goes into a holding tank to further separate the fat, remove air, and agglomerate the proteins (Boudreau and Saint-Amant, 1985, Caric, 1994). A separator is employed to remove the water phase and vacuum drying is used to reduce the water content even further. The milkfat (butter oil) is then cooled (Caric, 1994).

Another method to make AMF is a process that begins with whole milk. The milk is pasteurized and then sent to a machine called the Centrifactor (Caric, 1994). A Centrifixator is used commercially to centrifuge the milk to concentrate it from 35-45% cream to 80-85% cream, as well as homogenize it (Whittier, 1970). In this step, the cream is heated further and the fat globules are interrupted mechanically (Caric, 1994, Douglas, 2004). A serrated disc that homogenizes the milk also breaks the fat globules down and inverts the emulsion (Caric, 1994, Whittier, 1970). This is done at high temperatures, in order to create droplets of oil instead of butter granules (Whittier, 1970). The liquid oil leaves the machine with drops of water and buttermilk dispersed in it and as a result, the fat must be concentrated in a solids-ejecting separator (Douglas, 2004). The AMF is often vacuum-dried to drive the remaining water content to less than 0.1% and remove some of the oxygen present. This process serves to increase the shelf life (Walstra, et al., 1999).

The fatty acids in AMF can be modified into sucrose polyesters (SPE). These compounds are formed via the hydrolysis of triglycerides and the attachment of six to eight fatty acids to the hydroxyl groups of a sucrose molecule (Drake, et al., 1994). SPEs with a wide range of physical properties can be produced since milkfat has a wide range of properties and types of fatty acids (Kaylegian, 1995). SPE are not digested and can potentially be used as fat substitutes because they maintain the desired textural and flavor qualities of full caloric fats (Drake, et al., 1994).

### **1.2.3 Milkfat fractions**

Milkfat fractionation techniques have the potential to enhance desirable characteristics in milkfat products (Bruhn, 2004, Hartel and Kaylegian, 2001). Fractionation involves separation based on the physical or chemical properties of the natural milkfat components present. Using separation, desirable properties can be isolated, and as a result value-added ingredients can be obtained from lesser quality ones (Hartel and Kaylegian, 2001). The composition and properties of the fractions (fatty acid composition, melting point) depend on the makeup of the original milkfat (Shukla, 1995).

Milkfat is fractionated based on some of its inherent physical properties such as crystallization, solubility, and volatility (Kaylegian and Lindsey, 1995). Several fractionation methods have been used to separate milkfat. Crystallization is attractive for milkfat fractionation because milkfat contains a wide range of components which encompass a large melting point span. Milkfat is said to be composed of three main parts in regards to melting. The long-chain fatty acids (high molecular weight) which melt at a temperature around 50C and compose only 5-10% of the fat present,

constitute the high-melting fraction (HMF) (Barbano and Sherbon, 1975, Hartel and Kaylegian, 2001, Sherbon and Dolby, 1973). The middle-melting fraction (MMF) has a melting point between 35C and 40C and makes up approximately 25% of the fat composition. Fatty acids generally composed of two short chain or cis-unsaturated fatty acids and one saturated fatty acid have a melting point of less than 15C. This low-melting fraction (LMF) makes up the bulk of the fat composition (65-70%) (Hartel and Kaylegian, 2001).

Crystallization separation is very commonly used for butter (Hartel and Kaylegian, 2001). Fractionation is extremely desirable because it can be used to enhance the spreadability of butter. Butter with higher amounts of short and medium chain fatty acids can be separated via a distillation method of fractionation. This modified fatty acid composition utilizes the LMF of the milkfat to make the butter more spreadable (Bruhn, 2004). Another advantage to fractionation is the potential to blend fractions and create an expansive range of products that are extremely uniform in their properties (Deffense, 1993)

One method of fractionation, known as the Alfa-Laval process, is used to create a softer more spreadable butter product. It is a relatively simple separation based on melting point of fat crystals. Butterfat is heated and then cooled slowly. The fat that melts at the highest temperature settles to the bottom and the low temperature melting fat can be separated (King, 1974). The solid fat that settles out still contains a small amount of liquid fat between the crystals or adhered to their surface (Kaylegian, 1995). Refractionation can be used to obtain additional liquid fat.

Because more of the lower melting liquid fat is removed, the remaining solid portion will be highly functional (Kaylegian, 1995, Sherbon, et al., 1972).

While the LMF is utilized in butter applications, the HMF is beneficial to other products. Chocolate that contains a HMF of milkfat is less likely to exhibit chocolate bloom than chocolate that contains regular milkfat (Lohman and Hartel, 1994). The use of the HMF may also discourage softening of the chocolate due to a greater compatibility between the cocoa butter and the HMF. When used in pastries, butter with high levels of HMF creates flaky pastries by providing a solid and brittle structure that allows the layers to hold up (Deffense, 1993, Nor Hayati, et al., 2002). Normal butter is too weak at high temperatures to resist the forces in the dough and will collapse under the forces of the gluten (Shukla, 1995).

High-melting fractions of milkfat also have other potential uses. Shellhammer and Krochta (1997) tested the use of HMFs in edible films. A mixture that is highly crystalline is a better barrier to water because water transport is slower through this matrix. Because higher-melting fractions have a higher solid to liquid fat ratio, they will be less permeable to water vapor. Potentially these types of coatings could be used to improve shelf-life and reduce the need for polymeric packaging (Shellhammer and Krochta, 1997).

Monoglycerides, a commonly used emulsifier, and diglycerides are two more examples of the application of milkfat fractionation technology. Mono and diglycerides are produced by a reaction called lipid-catalyzed glycerolysis. In addition to emulsifying properties, mono and diglycerides also impart flavor characteristics to foods (Kaylegian, 1995).

Though separation of milkfat can be beneficial in many cases, it can also have its drawbacks. Fractionated milkfat may have different functional and nutritional properties, so it is sometimes difficult to incorporate these ingredients into products and yield a similar product to one that contains normal milkfat (Scott, et al., 2003). In addition, the composition of milkfat varies throughout different times of the year and different geographic locations. The fatty acid composition also depends heavily on the diet of the animal. Variable milk composition can interfere with fractionation efforts during some seasons of the year (Fox, 2000, Hartel and Kaylegian, 2001, Hawke and Taylor, 1994, Nickerson, 1995).

### **1.3 Butter making**

The art of concentrating milkfat to make butter has been recorded as early as 2000 BC. Though little is known about the techniques used in ancient times, what evidence exists points to the Arab and Syrian use of a crude churn made out of goat skins (Douglas, 2004). The preservation technique of making butter was practiced in households which had animals that produced milk, which made it a luxury. Ancient Romans used it on open wounds and applied it to their face as a skin treatment (Board, 2004). Wooden or metal churns were used in-home to produce batches of butter that were relatively consistent in flavor, texture, and appearance. The pivotal invention of the centrifugal cream separator largely removed buttermaking from the home (Douglas, 2004).

While the exact events during churning which destabilize the liquid phase emulsion and cause the fat to coalesce are unknown, a theory known as “auto-flotation” is commonly cited (Bruhn, 2004, Webb, et al., 1974). The theory states that

as the cream is churned, small air bubbles become integrated into it. Some of the fat globules, which have been burst by the agitation, begin to gather in small groups surrounding liquid fat that has been exuded by the burst fat globules (Bruhn, 2004, Walstra, et al., 1999, Webb, et al., 1974). This is the process that reverses the natural emulsion in the milk (oil-in-water) to butter, which consists of fine water droplets dispersed in an oil continuous phase.

### **1.3.1 Batch Process**

For centuries, butter making was done in a batch process which involved the churning of 30 to 35% milkfat cream in wooden or metal churns (Webb, et al., 1974). Cream is first pasteurized to 95C to denature enzymes and kill microorganisms that could be pathogenic, cause spoilage, or undesirable off-flavors (Douglas, 2004). In some European butters, a starter culture is added so desirable acid levels and flavors can be achieved. Regardless, pre-crystallization or ripening of the butterfat is necessary prior to the churning process. In this aging step, the cream is exposed to a cooling process that gives it the desired crystalline structure. Without aging, excessive fat loss in the buttermilk occurs because too little solid fat is formed (Walstra, et al., 1999). After 12-15 hours in the aging tank, the cream is pumped to a churn (Douglas, 2004). The agitation whips air into the liquid and creates butter grains, which increase in size throughout the churning process. Eventually, clumps of butter are formed with the remaining liquid being buttermilk. The buttermilk is drained off and the result is a water-in-oil emulsion. Washing the butter granules is one way to control the firmness of the butter. This is done with smaller butter grains and results in reduced dry matter content of the butter (Walstra, et al., 1999). From

this point, salt is added (for salted butter) at 1-3% (w/v) and the product is worked to ensure even distribution of the salt (Douglas, 2004). Working makes the butter into a continuous mass and finely disperses the small amount of water present throughout it. This can be achieved by passing the butter through rollers or a fall from a height (Walstra, et al., 1999). It is at this time that globular fat becomes free fat. Working also influences the properties upon which butter is graded-aroma, taste, shelf-life stability, appearance, and color (Douglas, 2004). During the working step, additional water may be added to meet the industry standard of identity (Walstra, et al., 1999). From this point, the butter is packaged and cooled. As the butter cools, the milkfat crystallizes and makes the product more firm (Douglas, 2004). Milkfat hardens slower than other fats because it contains such a variety of fatty acids (Van Aken and Visser, 2000).

### **1.3.2 Continuous Process**

Butter processing can be operated in a continuous fashion in the “Fritz” process (Figure 1.2) where machines can run up to 10,000kg of butter per hour (Kimenai, 1986, Walstra, et al., 1999). This process is commonly used in the United States. Prior to the actual churning event, the incoming milk is separated via a centrifuge and the skim milk is sent on for further processing. The cream is pasteurized at 185F for 25 seconds (Thompson, 2004). This serves to both inactivate microorganisms and inactivate enzymes which can speed up oxidation (Lane, 1998). Cream enters the first chamber and is turned aggressively by the beater (Walstra, et al., 1999). Fine butter grains result from this intense churning, which only lasts for a few seconds, an extremely short time in comparison to the batch process (Jebson,

1994). Following this, in the separating cylinder, the butter grains are churned into larger ones and the buttermilk is separated (Walstra, et al., 1999). The butter grains fall into a working section where a low-speed screw kneads them together or it is forced through holes in orifice plates (Jebson, 1994, Kimenai, 1986). In this step, lingering buttermilk is drained off. The vacuum chamber serves to reduce the amount of air in the butter. A second more rigorous working step follows this to ensure that the water droplets are finely dispersed (Kimenai, 1986).

The Cherry-Burrell process was used frequently in the United States until the late 1950's when costs and reduced quality butter made it less attractive than the Fritz process. This process uses concentrated cream with a higher milkfat content, (30-50%) (Munro, 1986). The 30%-50% milkfat cream is destabilized by pre-churners which whip air into it. Following this, the cream is separated via a centrifugal separator at 55C to yield cream at 82% milkfat, called plastic cream (Walstra, et al., 1999). Plastic cream differs from butter in that it is still an oil in water emulsion although it exhibits similar textural properties to butter (Chandon, 1997). The cream is pasteurized and then subjected to a steam vacuum stripper. It is then pumped to a tank where salt, water, and neutralizer are added to achieve the desirable composition and pH (Munro, 1986). It is cooled to 4-6C in a scraped surface heat-exchanger (Douglas, 2004). Cystallization is initiated here and at this temperature, fat crystals, residing in the milkfat globules, break open the MFGM and cause the still present liquid fat to flow out and rapidly invert the emulsion (Walstra, et al., 1999). Since the fat has been concentrated, no buttermilk is drained from the product as it is converted to butter. The phospholipid content of butter made using this method is higher

because none of the membrane material has been flushed out. The butter is further worked in the “Texturator” by passage through perforated plates. The butter is then packaged (Douglas, 2004). Butter produced using the Cherry-Burrell method has a greater tendency to oil-off (at 25 and 28C) than conventionally churned butter (Munro, 1986).

A phase separation method can also be employed in the continuous manufacture of butter from anhydrous milkfat. Similar to the Cherry-Burrell method, plastic cream is obtained (>80% fat) (Chandon, 1997, Douglas, 2004). The cream is heated and continuously agitated. This results in the destabilization of the emulsion and the formation of a distinct aqueous and oil phase. After separation, the remaining oil has been concentrated to 98% fat (Douglas, 2004). Water, milk solids, and salt are added back to the butter oil in an emulsion pump. Crystallization is initiated upon cooling in a scraped surface heat exchanger. The butter is worked further to allow growth of crystals and texture development (Bruhn, 2004, Douglas, 2004). Phospholipid content of phase separated butter is much less than that of the concentration process. This process is similar to the manufacture of margarine (Douglas, 2004).

#### **1.4 Butter Texture**

The unique fatty acid and triglyceride profile of butter, in addition to the structure of the fat and aqueous phases, results in a melting profile and mouthfeel that compliments the flavor of butter and makes it hard to imitate (Lane, 1998) (Figure 1.3). For this reason, a desirable butter texture is an extremely important characteristic to consumers (Mortensen and Danmark, 1982b, Rohm and Weidinger,

1993) due to texture's influence on spreadability, taste, mouthfeel, and impact on type of usage (Wright, et al., 2001). One of the main goals of the industry is to make butter more spreadable (Frede, 1997), which is complicated by the fact that the fatty acid composition and physical properties of the milkfat directly influence rheological properties (Mortensen and Danmark, 1982b, Wright, et al., 2001). A rheological property of a substance is a descriptor of how it reacts to stress and strain and its ability to flow as a fluid (Daubert and Foegeding, 1998).

Texture is a sensory attribute that cannot be quantified exclusively by measures of rheological properties; it is a combination of parameters. The complete description of the texture of a food item takes into account mouthfeel, tactile sense, and sounds that result from chewing as heard in the ears (Drake, 2000). Food texture can be classified into three categories: mechanical geometrical, and other characteristics (mouthfeel qualities related to the fat and moisture components) (Larmond, 1988). In butter, there are four principle components: free oil, fat crystals, water droplets, and fat globules. Only free oil and fat crystals are believed to have an effect on textural properties (de Man and Beers, 1988).

The interactions between the fat crystals in butter play an important role in the textural properties exhibited in the product. Van der Waals forces act between fat crystals to hold the butter matrix together (de man and Beers, 1988). Several types of bonds exist in butter structure. Very strong primary bonds are formed when fat crystals in close proximity grow together. Weaker and reversible secondary bonds exist between crystals (Precht, 1988). Both primary and secondary networks are able to form because no electrostatic or steric repulsion exists between the fat crystals

(Rohm and Weidinger, 1993). Manipulation of these bonds is one way to control the texture of butter. Rheological behavior can also be partially attributed to the interactions between the 3-dimensional network of fat crystals and the liquid oil present in the butter (de man and Beers, 1988).

Keeping other variables constant (i.e. temperature), the size and shape of the fat crystals in a sample will have the largest impact on butter texture (Kawanari, et al., 1981). This size is partially dependent on the fatty acid composition and arrangement of the fat that makes up the crystal. A range of melting points can be seen in different fatty acids in milkfat. While the majority will melt below 33C, some long-chain fatty acids can have a melting point upwards of 40C. There are some short-chain molecules that can still exist in liquid form at 0C (Prentice, 1992). The amount of these fatty acids present in the milkfat is influenced by both diet of the animal and season of production (Bornaz, et al., 1993, de Man and Wood, 1958). The firmness of the butter can be changed by manipulation of the type of fat in the cow's diet. Milkfat from cows that are fed high amounts of unsaturated fats are softer. A diet too rich in polyunsaturated fatty acids can result in butter that "oils off" at high temperatures because it is too fluid (Chen, et al., 2004).

Since the manufacturing method (continuous and batch) has an impact on the crystal structure, it also has an impact on the firmness of the butter. The crystalline structure produced by the two methods is strikingly different. One reason for this is that the butter made via a batch churn process undergoes a "setting" stage once it is removed from the churn and placed into cold storage (de Man and Wood, 1958, Prentice, 1992). Crystallization is essentially finished at this stage, but the crystals

align in the storage following manufacture (during the first week). Conversely, continuously made butter continues to crystallize after the manufacturing process is finished. As a result, the batch-made butter has a softer texture than butter made using a continuous process (de Man and Wood, 1958).

Temperature has a large impact on firmness of butter (Mortensen and Danmark, 1982b, Prentice, 1992). Temperature treatment of the cream (8C-20C-12C) and higher iodine value, an indication the butterfat contains more unsaturated glycerides (Prentice, 1992), caused a substantial reduction in firmness (Mortensen and Danmark, 1982b). Along with temperature of production, storage temperature is also important for the texture. A product that is slowly chilled will have a large fat crystal size and a grainy and soft texture (de Man and Beers, 1988). With such a wide range of melting triglycerides, any small change in temperature will result in an alteration of the solid fat content (Mortensen and Danmark, 1982b, Prentice, 1992) (Figure 1.4). If a butter sample is heated and then re-cooled, the recrystallization of the fat will fall behind the decreasing temperature resulting in a hysteresis curve (Figure 1.5). Since temperature is such a key variable in butter texture, it is important that it be monitored closely when measurements are taken to quantify texture (Prentice, 1992).

One of the problems that has been encountered in development of textural analysis techniques has been the debate over the classification of butter—whether it can be considered plastic and have a yield value or if it is really just a highly viscous liquid (Wright, et al., 2001). The current consensus is that butter does indeed behave

in a plastic fashion, and it is a non-Newtonian substance which can have a yield value (de Man and Beers, 1988).

One important textural characteristic of butter is that it exhibits thixotropic properties. Precht (1988) defines this term as the ability of a colloidal dispersion to form a reversible gel-like state due to Brownian motion or van der Waals forces. It is this phenomenon that is responsible for butter softening via working. Working butter after storage at low temperatures yields butter with decreased firmness (Mortensen and Danmark, 1982b, Prentice, 1992). The softening effect is the greatest on moderately hard butters at temperatures of 10-15C. This process is often used in manufacturing as well as in the blending of multiple types of butter. Firmness of the butter slowly returns after the working process because the crystals reorganize and then begin to slowly re-crystallize (Precht, 1988, Prentice, 1992, Van Aken and Visser, 2000). But if working is done after the completion of the setting step, considerable softening of the butter will be achieved (Mortensen and Danmark, 1982b). Because continuously made butter is thought to have more irreversible bonds between crystals than batch churned butter, this butter does not regain hardness as quickly (Vasic and de Man, 1967).

Another method that results in decreased firmness of butter is blending or the addition of other oils (Fearon and Johnston, 1988, Mortensen and Danmark, 1981). Blending butter with other oils may be impractical for marketing because the addition of other oils does not fit with the standard of identity for butter (Mortensen and Danmark, 1982b). Some success has been achieved by blending softer winter butter

with summer butter. Incorporation of gases in butter through whipping has also been shown to reduce firmness and increase spreadability (Fearon and Johnston, 1988).

#### **1.4.1 Instrumental Methods**

There are currently many techniques to evaluate butter texture (Fearon and Johnston, 1989, Voisey, 1976). Voisey (1976) described three types of tests to evaluate texture: fundamental, empirical, and imitative. Fundamental tests examine the exact physical properties of the sample in terms of well-defined rheological parameters. Empirical tests use multiple stresses, which relate to the property of interest, but are not clearly defined in rheological measures like fundamental tests. Empirical tests relate closely to the property of interest of the food system. This type of test is very commonly used because it is hard to quantify many food systems with just one specific rheological property. Imitative tests are designed to mimic actual processes that foods go through such as grinding or chewing. Few of these tests exist since the wide variety of stresses placed on a product make the analysis difficult and in some cases virtually impossible.

One commonly used fundamental technique to evaluate butter texture is the penetrometer (de Man, 1976, de Man and Beers, 1988, Mortensen and Danmark, 1981, Wright, et al., 2001). This method involves using a cone, needle, or sphere to penetrate the sample. The instrument is released for a specific interval of time and the depth or the rate at which it permeates when it is released is the desired measurement (Wright, et al., 2001). The point at which the structure of the product is broken is the yield stress (Mortensen and Danmark, 1981). Exceeding the material's minimum flow value will cause the product to flow (Fearon and Johnston, 1989).

Cone penetrometer data has been converted to hardness. Hardness is the ratio of the force necessary to make an impression on the sample to the area of the impression (de Man, 1976). This is a relatively simple technique that has been shown to give consistent measurements between samples. Data from cone penetrometers has been frequently correlated with sensory data for butter (Dixon, 1974, Mortensen and Danmark, 1982a).

Another method of analysis done on plastic fats to quantify firmness, measures the force required for a metal wire to cut through the sample at a constant rate (Dixon and Williams, 1977, Wright, et al., 2001). These sectility measurements originally were done by adding weights until the force was large enough to cut through the sample at the desired speed (Prentice, 1972). A modern, more commonly used variation, of this method measures the counteracting force on the wire as it slices through the sample at a constant speed (Frede, 1997, Wright, et al., 2001). This method is extremely simple and can be performed with minimal preparation to the sample (Prentice, 1972).

A compression technique can also be used to evaluate the deformation of butter over time (Dixon, 1974, Wright, et al., 2001). This is done with a sample, typically prism-shaped or cylindrical, which is placed between 2 flat platens. One method involves applying uniform stress from the top and bottom and recording the deformation of the sample over time (Prentice, 1972, Wright, et al., 2001). Another similar method that is extremely useful in butter analysis compresses only the top of the sample at a constant rate of speed. Yield stress, firmness, and plasticity can be determined from the resulting deformation from the downward force (Wright, et al.,

2001). Both compression methods are useful in examining butter spreadability because the test examines the ability of the sample to flow. From compression measurements two things can be derived: a firmness measure expressed as shear strength, and the percent deformation at the maximum shear called the plasticity (Dixon, 1974).

Although compression is used to evaluate spreadability, it does not simulate all of the factors involved in this motion. Since it is extremely difficult to quantitatively evaluate spreadability, an extrusion device can more accurately mimic the action of spreading (de Man, 1976, Dixon, 1974, Prentice, 1972, Wright, et al., 2001). The force that is necessary to propel a sample through an opening at a constant speed and thrust is one of the forces measured in the extrusion test (Prentice, 1972). The other measurement is the friction of the sample along the barrel of instrument. When almost all of the sample has been pushed out, frictional forces are approaching zero, and at this point only the thrust is acting on the sample (Prentice, 1972, Wright, et al., 2001). Using extrusion may more accurately represent the flow that occurs when butter is spreading. Spreading is a rapid deformation of the product; other testing methods evaluate slow deformation so these may not accurately represent the spreadability of the sample. Extrusion results have been inversely correlated with penetrometer test results. In addition, it is thought that frictional force is proportional to stickiness (Wright, et al., 2001).

Another method that is used to assess spreadability is the vane method. This technique allows the yield stress to be calculated from the torque necessary to rotate a 4-8 bladed vane instrument through the sample (Daubert, et al., 1998). As the vane is

rotated slowly through the sample, the largest torque is exerted on the instrument as the sample is the yielding moment (Dzuy and Boger, 1985). The vane test also provides information to determine the maximum amount of force a product can withstand before it deforms. This yield strain can be plotted against yield stress to create a “spreadability map” of the product. The texture map is helpful because the effect of different processing conditions on spreadability can be conceptualized (Daubert, et al., 1998).

Aside from spreadability and firmness, another important attribute for the consumer is the ability of butter to melt. This is especially important in cooking applications (Borwankar, et al., 1992). Though few studies have been conducted on melting properties of butter, some methods have been developed. Most of these methods are used to analyze margarine and table spreads in an attempt to replicate the desirable melting profile of butter. A desirable melt is not waxy or greasy and has well released flavor (Borwankar, et al., 1992).

One simple method used to evaluate melt is the dropping point method. A small amount of sample is placed in a cup with a hole in the bottom. The cup and sample are heated slowly (often increasing 1°C/min) (Borwankar, et al., 1992, Papalois, et al., 1996). The temperature when the sample begins to flow and the material falls through is called the drop point. The measurement is considered a determination of rheological properties more so than of thermodynamic ones. Since butter is not purely fat, melting point of the fat can not be determined from this test (Borwankar, et al., 1992).

Obtaining a more sophisticated melting profile can be done using differential scanning calorimetry (DSC) (Fearon and Johnston, 1988, Tan and Man, 2000). This technique utilizes a small amount of sample on which the DSC runs a set temperature profile (preset by the user) and the instrument measures the thermal energy absorbed or evolved. The energy results from the physical changes caused by the temperature fluctuation (Fearon and Johnston, 1988) and a unique profile is created (Figure 1.6). DSC is widely used and valued because it gives more detailed information on melting and crystallization temperatures and heat of fusion (Schaffer, et al., 2001, Tan and Man, 2000).

Texture profile analysis (TPA) has been used successfully with other products to create a unique texture map of each individual product (Truong, et al., 2002). This technique has allowed textural differences between similar samples to be quantified. The instrument stresses the sample in an attempt to resemble the forces placed on it by the human jaw. TPA can be configured to measure fracturability, hardness, adhesiveness, cohesiveness, springiness, chewiness, and gumminess. The first five are measured and the last two are calculated (Bourne, 1978). The creation of these categories is an attempt to classify texture terms and relate rheological properties and common sensory language (Larmond, 1988).

#### **1.4.2 Sensory Evaluation of Butter Texture**

In addition to analytical techniques, sensory analysis has been conducted on food products to evaluate texture. Texture is ultimately a sensory parameter and thus sensory evaluations are important for texture evaluation. Texture is a property difficult to evaluate with the use of a machine because it can only quantify the

textural parameters in terms of a few specific characteristics. Because it is a multi-parameter characteristic, detected by several senses, and derived from the food structure, texture is evaluated well by individuals who can perceive and describe all attributes of a product's texture (Szczesniak, 2002). Specifically for butter, sensory tests are conducted to evaluate spreadability, firmness and other attributes that are difficult to assess with an analytical instrument because they can not be defined by only one rheological property.

### **1.4.3 Sensory Methods**

The evaluation of texture using sensory techniques relies mostly on trained panelists. Discrimination tests require 20 to 40 people and can only be used to determine if differences exist. More advanced descriptive analysis involving texture profiling or an attribute test involves a trained panel that has 8 to 14 participants (Drake, 2000, Lawless and Heymann, 1998). Trained panelists are taught the definitions of the terms they will be asked to evaluate so they will be consistent with the attribute they are judging (Lawless and Heymann, 1998). Becoming familiar with the scale is another important part of training. Linear (line) or numerical scales are used to quantitatively measure panelist responses. It is important that panelists are able to use the scale effectively and consistently. Identifying and recognizing texture references and their place on the scale is another key skill panelists must be versed in. In addition, training serves to make the group better synchronized and perform better statistically (Drake and Civille, 2002)

Untrained panels are not relied upon much for textural evaluations. They can only give information about preference and liking; they are unable to shed light on

product differences, texture attributes, and intensities of attributes the product exhibits since untrained panelists have no frame of reference and the results would not be comparable (Drake, 2000, Meullenet, 2004). Consumers have been found to be adequate for producing preliminary texture profiles when large numbers of panelists are used (Drake, 2000, Lawless and Heymann, 1998, Meullenet, 2004).

One way that spreadability has been measured by sensory analysis is by spreading a consistent amount of butter on a piece of filter paper. Rohm and Ulberth (1990) trained sensory panelists do this process and then score the sample in comparison to a margarine reference that was defined as a 10 or “most spreadable.” Similar tests on other solid fats have been done by spreading the test sample on a piece of bread and evaluating it on a graphic 150 mm scale (Pokorny, et al., 1985). Dixon and Parekh (1980) found reasonable agreement among trained panelists in butter that was considered “hard,” but as the butter became softer, there was a significant variation in panel response. Hardness is often evaluated by cutting the sample with a knife. Panelists were asked to evaluate the amount of force necessary to cut through a butter sample (Rohm and Ulberth, 1990). Prentice (1972) found that untrained panelists had a difficult time distinguishing firmness on a seven-point scale.

Often used in the textile industry, hand evaluation has also been used to assess food texture (Lawless and Heymann, 1998). Drake, et al. (1999) found that mouth and hand evaluation of cheese produced comparable results for some specific textural attributes, including: firmness, stickiness, and slipperiness. This type of work has not been conducted with butter.

#### **1.4.4 Correlating Sensory and Analytical Measurements**

Szczesniak (1987) stated that making correlations between sensory and instrumental methods is important for several reasons. In quality control, correlations between sensory and analytical methods are helpful because they allow for the use of faster instrumental tests to determine if products are texturally consistent. Extensive work must be done on a commodity to evaluate which parameters contribute to consumer acceptance and the relationship between these and empirical measurements. This data can potentially be used to predict consumer satisfaction. Linking sensory attributes and specific forces is very important to understanding what is sensed in the mouth when texture is being perceived. A strong relationship between sensory and analytical techniques will also help to advance the optimization of instruments to better simulate human perception.

In order for strong links to be made between mechanical instrumentation and sensory results, the physical parameters of the two tests must be similar. This can be difficult because the characteristic being examined can vary widely between products. Hardness, for example, can be measured by viscosity and consistency for things like pudding and whipped cream. On the other hand, flexibility and puncture are used for harder commodities like carrots. Oral sensory evaluations also involve subjecting the sample to more than one force (Szczesniak, 1987). To complicate the matter, deformation properties differ between food products and this property plays a vital role in the perception of texture. The complex nature of sensory terms can sometimes make the results difficult to correlate with empirical measurements. Because many of the sensory attributes that are evaluated (i.e. consistency) are composed of several sensations in the mouth, breaking these terms down into component parts can make

them more comparable to a mechanical testing (Bourne, 1983). Instrumental and sensory measurements will align best when the analytical method mimics the deformation of the product closely (Lawless and Heymann, 1998, Szczesniak, 1987).

Some parameters correlate better to sensory data better than others. Empirical tests for hardness have consistently correlated well with sensory studies (Lawless and Heymann, 1998, Szczesniak, 1987). More subjective measures, like cohesiveness have less correlation with empirical measurements. This could be due to the difficulty profiling these subjective components through sensory panels or the lack of adequate instruments to empirically analyze these properties (Szczesniak, 1998). Mortensen and Danmark (1982) found a good correlation between penetrometry and sectility measures of yield stress and sensory values of spreadability for butter. Shear, compression, and penetrometer tests were shown by Kawanari et al. (1981) to coincide with sensory data. In Cheddar cheese, TPA and compression tests were used successfully to predict sensory attributes (Truong, et al., 2002). Texture evaluation of butter is somewhat difficult because temperature has such an effect on the softness of butter. In order to obtain reliable results, tests must be done at a consistent temperature under controlled conditions. Both spreadability and hardness have a linear relationship with temperature (Rohm and Ulberth, 1990).

### **1.5 USDA Grading**

The United States has set standards for the sensory quality of butter. U.S. Grades AA, A, and B are awarded based on the body, color, and salt characteristics of the product. A sample of butter is taken with a trier, a two-edged curve-bladed tool. The trier is inserted into the sample and turned half of a rotation to pull a core out

from the block of butter. This sample is then evaluated (Bodyfelt, et al., 1988). Flavor and texture attributes are classified “slight, definite, or pronounced” (United States Department of Agriculture, 1989). The aroma that is initially present when the trier sample is taken should be correlated with the taste. Top quality butter should impart a mild, pleasant, sweet, and clean flavor in the mouth (Bodyfelt, et al., 1988). There are many off-flavors that can be detected in butter. The USDA defines 17 of these flavors which impact the final quality grade (Table 1.4). Major sensory texture defects in butter have been identified for USDA grading of butter. Textural defects can be classified as: crumbly, gummy, leaky, mealy/grainy, ragged boring, salvy or short, sticky, and weak (USDA, 1989) (Figure 1.5).

## **1.6 Flavor**

Odor, taste, and mouthfeel all contribute to the perception of flavor. The lipid component of foods specifically has a large impact on how flavors are perceived. Lipids can impart their own flavors, act as flavor precursors and flavor carriers, and modify flavor perception by changing the threshold concentration and retention (Kinsella, 1975). There are a large number of volatile compounds in butter, and as of 1996, 287 compounds had been detected. There is currently no chemical mixture that has been able to successfully replicate the aroma of butter (Peterson and Reineccius, 2003).

Alkanoic acids are present in triglycerides of fresh butter and upon heating they are converted via carboxylation to methyl ketones (Douglas, 2004, Kinsella, 1975). Methyl ketones are present in quantities below the flavor threshold value (FTV) in unheated butter and may not contribute significantly to flavor in fresh

butter. Methyl ketones contribute to the characteristic flavor of melted butter and may contribute desirable flavors in cooking and baking. These flavor compounds also contribute to the flavor of bleu cheeses (Douglas, 2004). The flavor threshold of methyl ketones with 6-10 carbons is 2-20 ppm in oil systems (Hammond, 1986).

Diacetyl (2,3-butanedione) is a specific ketone which provides the rich heated note of butter. It is important because it imparts the characteristic “butter smell” (Douglas, 2004, Kinsella, 1975). The importance of diacetyl in fresh butter has been the subject of some debate (Peterson and Reineccius, 2003). Schieberle et al. (1993) considered diacetyl to be less important in butters that were not fermented (i.e. sweet cream butter) since the concentration was considerably lower in these compared to the cultured butters. The result of the low concentration of diacetyl may be the reason for the overall mild and sweet odor in sweet cream butter. Peterson and Reineccius (2003) did find diacetyl to be above the flavor threshold in the headspace of sweet cream butter, suggesting it does significantly contribute to aroma.

The precursors of lactones are glyceride esters. These precursors are present in butter and form lactones from spontaneous hydrolysis and lactonization. Aside from spontaneous production, all glyceride esters are converted to lactones when butter is heated. Two forms of lactones exist in butter: the five-carbon-ring delta ( $\delta$ ) lactone and the four-carbon-ring gamma ( $\gamma$ ) lactone. The  $\delta$  lactone is present in higher quantities and has a greater impact on the flavor (Kinsella, 1975). Lactones account for some of the rich flavor notes from heating butter. The presence of these compounds contributes to the sweet, coconut, and fruity flavors associated with butter

(Douglas, 2004). Bovolide (the enol lactone of 2,3-dimethyl-4-keto-2-nonenoic acid) is an unsaturated lactone that is said to contribute to butter flavor (Hammond, 1986).

Short-chain fatty acids (less than 12 carbons), also contribute significantly to flavor. They are released upon heating, hydrolysis, and lipolysis. They concentrate in the aqueous phase of the butter because they are water soluble (Dairy Management Inc., 1996). Off-flavors due to these free fatty acids have been described as: rancid, butyric, goaty, soapy, bitter or unclean (Woo and Lindsey, 1983). One specific short-chain fatty acid that is a very important contribution to flavor is butyric acid (butanoic acid). It composes 3-4% of the glycerides in butter. The hydrolysis of this glyceride results in the foul odor identified in rancid butter (Douglas, 2004).

Lipid derived aldehydes are a factor in the flavor and odor profile of butter. When they accumulate to high enough levels, aldehydes contribute to the oxidized off-flavors associated with butter (Kinsella, 1975). The presence of these aldehydes is usually attributed to the autooxidation of unsaturated fatty acids (Hammond, 1986, Kinsella, 1975). Polyunsaturated fatty acid, specifically linoleic and linolenic acids, are the primary species that are oxidized, but there are other polyunsaturated fatty acids that are present in small quantities which can also be oxidized (Kinsella, 1975). The breakdown of isoleucine and leucine can produce aldehyde products (2-methyl and 3-methylbutanal) via the Strecker degradation (Peterson and Reineccius, 2003). Widder and Grosch (1997) determined that autooxidation of palmitoleic acid results in the formation of (E)-2-nonenal and (Z)-2-nonenal, which are responsible for a cardboardy off-flavor in butter oil.

The off-flavor of oxidized lipid is described as cardboardy, oxidized, or metallic. Extensive oxidation can result in a fishy or oily flavor. Controlling oxidized flavors is most practical in processing by ensuring that milk does not come in contact with copper surfaces (Hammond, 1986). Additionally, temperature fluctuations and long storage times lead to oxidative off-flavors; consistent storage conditions will prevent oxidative damage (Widder and Grosch, 1997, Woo and Lindsay, 1984). Aside from oxidation, aldehydes are also produced at low levels via fermentation (Kinsella, 1975). When aldehydes are present in trace quantities, they can impart desirable flavor characteristics. At the parts-per-billion level, cis-4-heptenal (creamy-buttery), trans-2-hexenal (green-grassy), 2,4-decadienal (deep-fried), and n-alkanals and 2-nonenal (nutty) contribute pleasing flavors (Kinsella, 1975).

Dimethyl sulfide (DMS) is the most significant of the sulfur compounds that is present in butter. Due to its extremely volatile nature, DMS only contributes to butter aroma for a small amount of time preceding its production (Peterson and Reineccius, 2003). It is able to smooth out some of the harsher flavor characteristics associated with diacetyl and other acid notes in butter when it is present at 30-50 ppb (Kinsella, 1975). Butter containing DMS is thought to have a “corn-like” flavor, and is typically graded higher because of this desirable flavor. Two other sulfur compounds have been found in butter. One of them, dimethyl sulphone, is thought to be an oxidation product of DMS. The other, dimethyl trisulphide, has been identified in cheeses and pasteurized milk, and is newly reported in butter (Peterson and Reineccius, 2003). A degradation product of tryptophan, skatole, has been found in

butter in numerous studies. The contribution of skatole to aroma and flavor has been confirmed in some studies, but not others (Peterson and Reineccius, 2003).

### **1.6.1 Instrumental Methods**

The analysis of flavor compounds can be somewhat difficult due to the vast variety of compounds that exist and the extremely small concentrations of these chemicals (often in the ppm or ppb range). In addition the complexity of the food matrix can make flavor and volatile isolation difficult, as well as the potential instability of some of the compounds (Parliment, 2002). Many methods have been developed to identify and quantify the molecules that contribute to flavor and aroma. Static and dynamic headspace sampling, solid-phase microextraction, and gas-chromatography which utilizes olfactometry or mass spectrometry are all techniques that are commonly used. No one type of analytical tool is ideal for all samples and there are many variations and methods associated with each of these techniques (Teranishi, 1998).

Headspace sampling can be done to analyze volatile compounds that maybe destroyed in a solvent-based extraction. This type of sampling also does not need to be carried out at high temperatures, which could result in the transformation of the compounds of interest. When using the static headspace sampling technique, a sample is placed in a sealed container and the volatiles are allowed to equilibrate (Povolo and Contarini, 2003, Reid, 2003). Heating can be used to increase the compounds present in the headspace if it does not result in the formation of unwanted substances or artifacts. After equilibration, some of the headspace is removed and injected into a gas chromatograph-mass spectrophotomer (GC-MS) or a gas

chromatograph-infrared spectroscophotomer (GC-IR) or other analytical instrument (Reid, 2003, Wampler, 2002). This technique allows for the analysis of low molecular weight compounds without the presence of solvent. Another advantage of static headspace sampling is that it is easily automated and has a simple sample preparation and a relatively low cost to analyze each sample (Wampler, 2002). Analysis of the static headspace may also be extremely important because it is possibly the best representation of the volatiles the nose receives when it smells a food product (Peterson and Reineccius, 2003). Static headspace analysis is disadvantageous for analytes present at low concentrations and those with high boiling points because most static headspace instruments can only heat to 150C (Wampler, 2002).

Dynamic headspace sampling or purge and trap sampling is more sensitive than the simpler static method. The sample sits in a container that contains a purge head with a trap. Inert gas is passed over the sample and exits through the trap (Povolo and Contarini, 2003, Reid, 2003). As the volatile compounds are flushed out, the equilibrium becomes unbalanced and more of the compounds are pulled from the sample (Wampler, 2002). The sample can be heated to increase the amount of the volatiles present in the headspace. Volatiles are removed from the trap by flushing a carrier gas through an adsorbant. Tenax (poly-2, 6-diphenyl-*p*-phenylene oxide) is a commonly used sorbant material because it can be used for a wide range of volatiles, especially aromatics, and it is stable to high heating temperatures (Wampler, 2002). The adsorbant is heated to release the volatiles that are trapped in it. The volatiles can also be extracted using a solvent. This is advantageous because the sample can

be analyzed more than once, but it is less sensitive since the concentration of volatiles is diluted in the solvent (Reid, 2003). Dynamic headspace analysis is more sensitive than static headspace analysis due to the trapping stage and offers the same advantages of the static headspace technique. The purge and trap has a greater chance to malfunction since the instrument is considerably more complicated than the simple static headspace sampler. The time required to extract one sample is also longer than in static headspace sampling (Wampler, 2002).

Another commonly used extraction technique is solid phase microextraction (SPME). A silica fiber coated in a polymer is suspended in the headspace of a solid sample or submerged in a liquid sample in a sealed container (Harmon, 2002, Reid, 2003). The make-up of the fiber and the conditions under which the sample is taken will affect the sensitivity of the test. Compounds are absorbed by the fiber and after equilibrium is reached it is placed into the injection port of a gas chromatograph. In the injection port, heat is used to desorb the compounds from the fiber, and then they are analyzed (Harmon, 2002). SPME is a rapid method that is some-what similar to static headspace analysis (Reid, 2003). An internal standard and carefully controlled extraction conditions will result in excellent quantitative results. SPME is also advantageous because it can be done without heating the sample and risking the formation of chemical biproducts (Harmon, 2002).

A recently developed technique utilizes high vacuum to distill the food directly or a solvent extract of the food. The high vacuum transfer (HVT) is possible because a large temperature gradient between two vessels connected by a glass tube allows the volatiles to be separated from the nonvolatile material. Either diethyl ether

or dichloromethane must be used as the solvent because other solvents may freeze at the low temperatures and clog up the joints. Another drawback to HVT is the possibility that compounds may condense before reaching the collection vessel. The equipment is also fragile, expensive, and the process is time consuming (Parliment, 2001).

An alternative piece of equipment that uses a similar technique of distillation of volatiles was developed by Engel et al. (1999). This technique is known as solvent assisted flavor evaporation (SAFE). The sample is dripped in and the flask is surrounded by liquid nitrogen, after the compounds volatilize, they go into a trap where the nonvolatile compounds are removed from the vaporous portion. Then the volatiles condense into another flask which is cooled by liquid nitrogen. The apparatus is kept at a constant temperature to prevent freezing within the joints. The SAFE method has been shown to give higher yields of volatiles than HVT (Engel et al., 1999). Furthermore, it is less time consuming, more compact, and economically efficient.

Gas chromatography-olfactometry (GC-O) can be used to analyze volatile food components. GC-O utilizes gas-chromatography in conjunction with a human nose to detect aroma-active compounds in a sample. As the sample eludes from the GC column, it is split. Part of the column effluent is sent to a traditional GC detector (such as an FID) hooked up to a computer. The other portion is mixed with air and water vapor and sent to a tube where a person can perceive the aroma (Reid, 2003). This technique is valuable because it allows compounds present in minute amounts that may be below instrumental detection limits to be recognized. Many of these

compounds are important to the aroma profile of the food because they have low odor thresholds (Blank, 2002).

Similar to the GC-O, the gas chromatograph-mass spectrometer (GC-MS) is another extremely powerful tool in the analysis of flavor and aroma compounds (Reid, 2003). This method can be used to create a unique profile for each test sample. After the sample is separated in the GC, the molecules go to a vacuum chamber where they are ionized. They are then quantified based on their mass to charge ratio and plotted versus time as a mass spectrogram (a histogram) (Holland and Gardner, 2002). A full scan can be run over a set mass range continually. If the compound of interest is known, selected ion monitoring (SIM) can be used to determine the quantity of the known compound that is present. This is often used in determining if a taint or specific off-flavor is present in a food (Reid, 2003). The profile of the sample can be compared with compound reference libraries and the components can be identified (Holland and Gardner, 2002).

### **1.6.1 Descriptive Sensory Analysis of Flavor**

Aside from instrumental quantification of flavor compounds, descriptive sensory analysis is often used to quantitatively assess the flavor profile of products. One common method of descriptive analysis is the Spectrum™ method. This method was developed to allow multiple attributes and products to be directly compared on a universal 15-point scale. Reference anchors at various intensities on the scale are used for panel and panelist calibration. Panelists have undergone 50-100 hours of training by a highly experienced leader and are able to use scales to evaluate characteristics from a standardized lexicon (Meilgaard et al., 1999). The Spectrum™

method allows a wide range of products to be compared due to its use of the universal scale approach and is extremely suitable to monitoring changes over time (Lawless and Heymann, 1998). The drawback to the Spectrum<sup>TM</sup> method is that it requires a large number of panel training hours and panel maintenance.

The Spectrum<sup>TM</sup> method of descriptive analysis has been widely used to characterize the flavors of many products including cheese, peanut butter, chocolate milk and many others (Young et al., 2004; McNeill et al., 2002; Thompson et al., 2004). Currently no published lexicon for butter flavors exists. Additionally, use of descriptive sensory analysis has been limited in previous studies of butter flavor. Kim et al. (2005) used descriptive analysis to compare butterfat-oil blends to pure butter. However, specific butter flavors were not characterized. Similarly, Tuorila et al. (1989) conducted limited descriptive sensory analysis on spreads with comparisons to butter. Specific butter flavors (milkfat, cooked, etc) were not evaluated. Michicich et al. (1999) also compared butter to three substitutes. Descriptive profiling was conducted on the butter and spreads. Broad butter and spread flavors were identified in the samples. Additional studies have focused on butter quality (storage, packaging, etc.). The studies have widely used grading as their chosen method of sensory flavor evaluation (Emmons et al., 1986; Tomlinson and Dixon, 1977; Jebson et al., 1974; MacBean, 1974).

## **1.7 Butter and Consumers**

Butter is a highly prized fat source for its rich flavor attributes. A wide array of butter is sold, varying widely in color, texture, and flavor. Most Americans are

familiar with sweet cream butter which is produced without the addition of starter culture, and may or may not have added salt.

Butter consumption is currently stagnant; consumption per capita in the United States increased from 4.5 to 4.6 lbs over the period of 2000-2004. Production during this time period has also remained relatively constant, down only 0.5% from the 1,256 million pounds produced in 2000 (International Dairy Foods Association, 2005). Health concerns have been often cited as the reason many consumers decreased their butter consumption and increased their intake of margarine (Crane, 1993). By definition, butter contains 80% fat (USDA, 1989). Over 60% of this fat is saturated (Riel, 1985). Since milkfat comes from an animal source, it is also a significant contributor of cholesterol.

Margarine and vegetable oil spreads, in contrast, are made from vegetable oil and contain no cholesterol. Margarine itself is not without controversy though, as it has come under fire recently for its high *trans* fat content. The hydrogenated oils that are used to make margarine contain *trans* fatty acids which may be linked to chronic heart disease and may contribute to increases in cholesterol levels (Stauffer, 1996b). The legal definition for margarine, similar to butter, is that it contains 80% fat (FDA, 2005). Many softer spreads contain less fat and by definition cannot be labeled as margarine. Per capita consumption of margarine in the United States declined between 2000 and 2004; from 7.5lbs per year to 5.3lbs per year (USDA-ERS, 2006).

To our knowledge, no studies have examined consumer attitudes toward the varying flavors exhibited among butters and compared them to margarine or vegetable oil spreads. This study was conducted to identify the sensory

characteristics (primarily specific flavors) that drive consumer acceptance of butter and margarine. A descriptive sensory language for butter and vegetable oil spreads was identified. This language was then used to document the sensory characteristics of butter and margarines. Consumer acceptance testing was applied followed by internal and external preference mapping. These results will help manufacturers understand what different segments of the market prefer and how to best accommodate these market segments.

### **1.8 Butter Storage**

Dairy manufacturers produce large amounts of butter in the winter months due to a surplus of milkfat. It is often necessary to store this butter for extended periods of time until there is a demand for it. During refrigerated and/or frozen storage, degradation of quality may occur and this is an important issue when companies develop specification sheets for butter suppliers or for butter suppliers to design storage regimes. Butter is commonly stored for extended periods in blocks (25 kg) which are subsequently re-worked into quarter-pound sticks. However, retail packages (sticks or quarters as they are referred to by industry) are often stored for extended periods.

To our knowledge, recent studies have not addressed butter storage stability. Further, stability of bulk and stick butter have not been compared. Our objective was to evaluate the flavor and texture stability of bulk and stick butter across frozen (-20C) and refrigerated (5C) storage. Descriptive sensory analysis, which has not been previously applied to butter flavor, was used to monitor flavor and texture. Instrumental methods were also used to evaluate chemical and texture changes.

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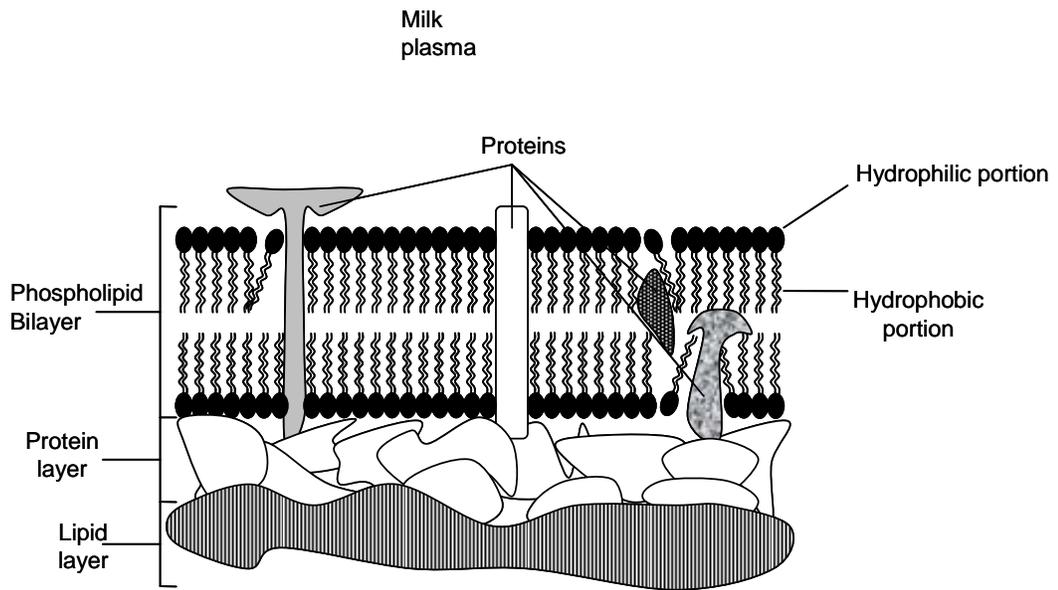
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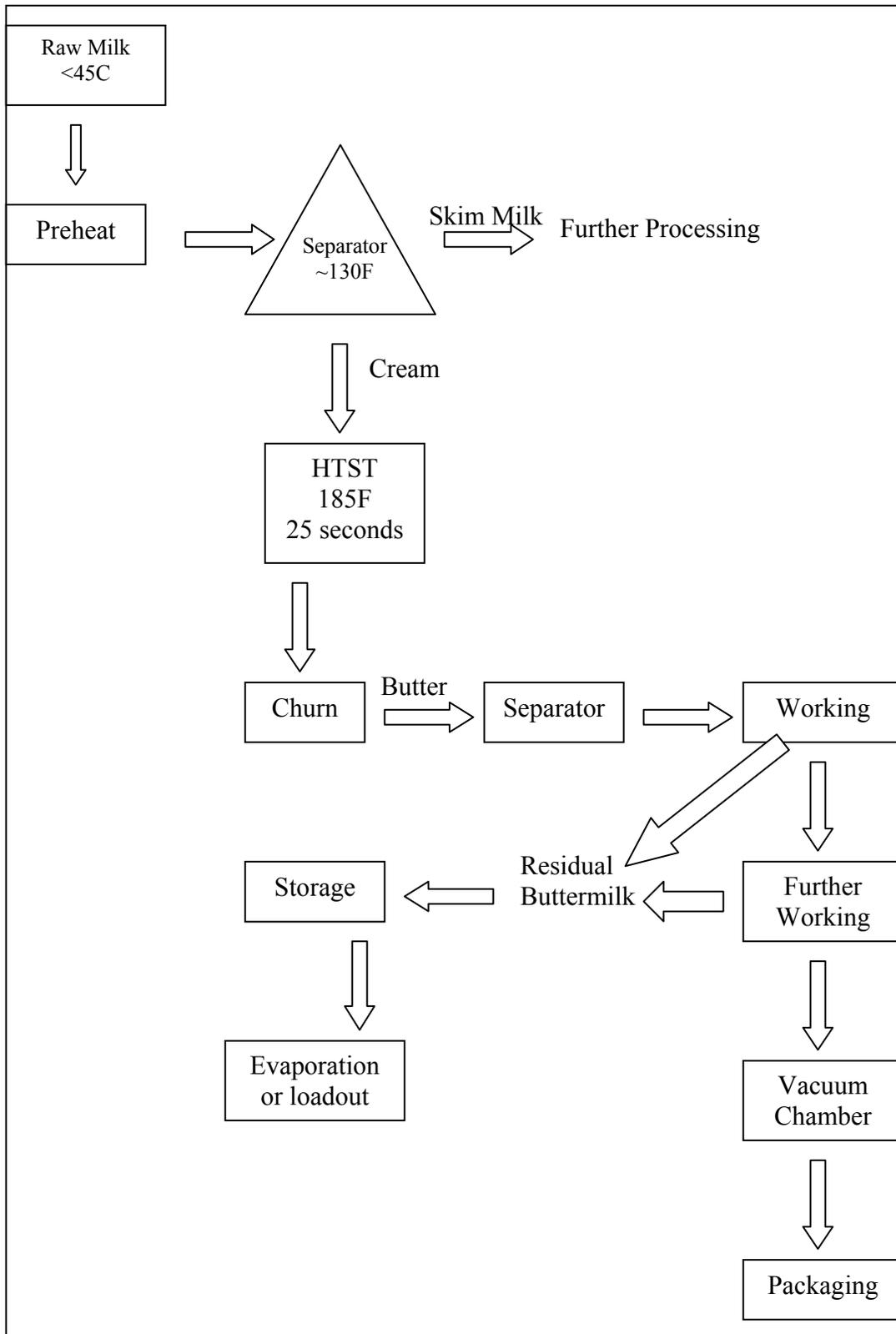
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**Figure 1.1 Schematic drawing of a MFGM. Adapted from Fox and McSweeney, 1998.**



**Figure 1.2 Fritz continuous method of buttermaking. Adapted from Kimenai (1986) and Thompson (2004).**

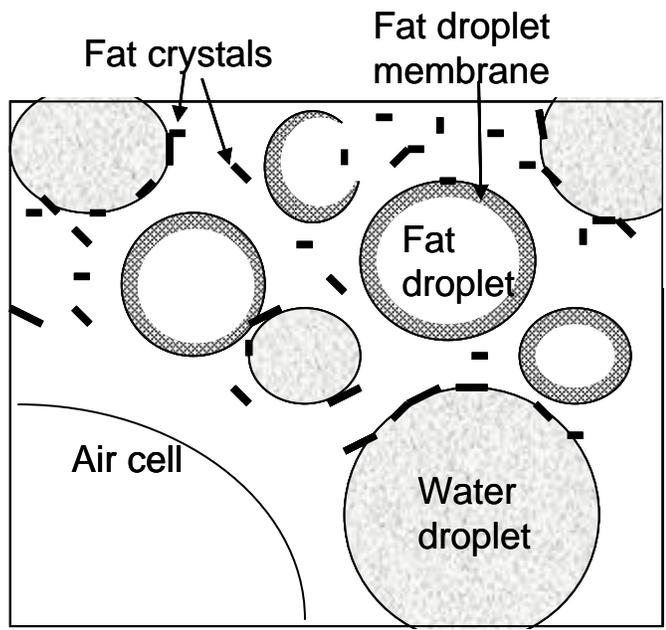
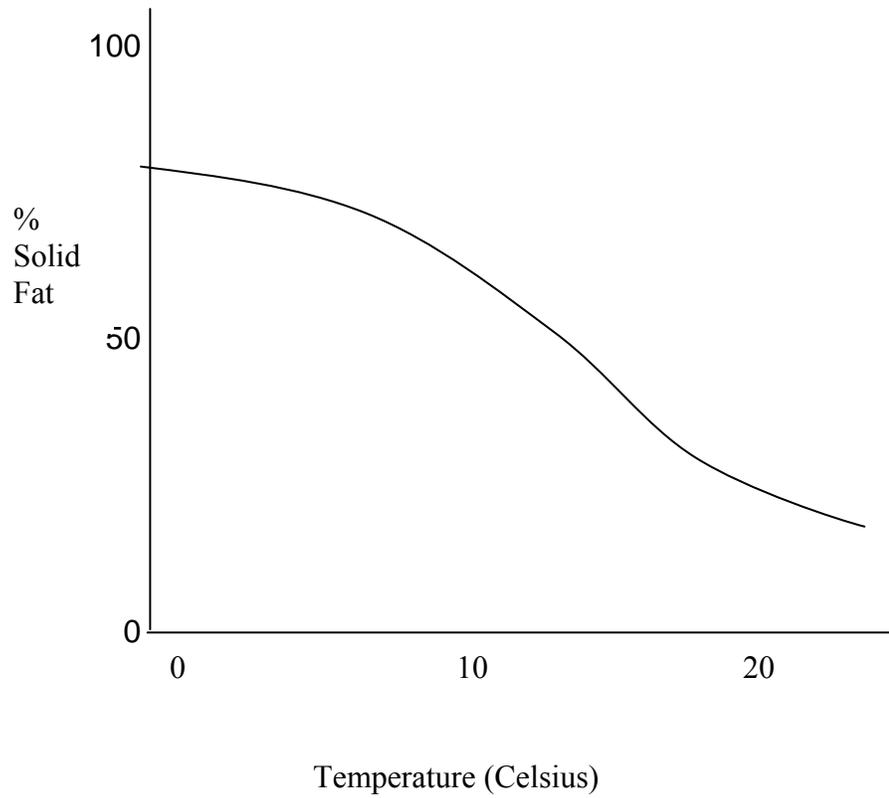
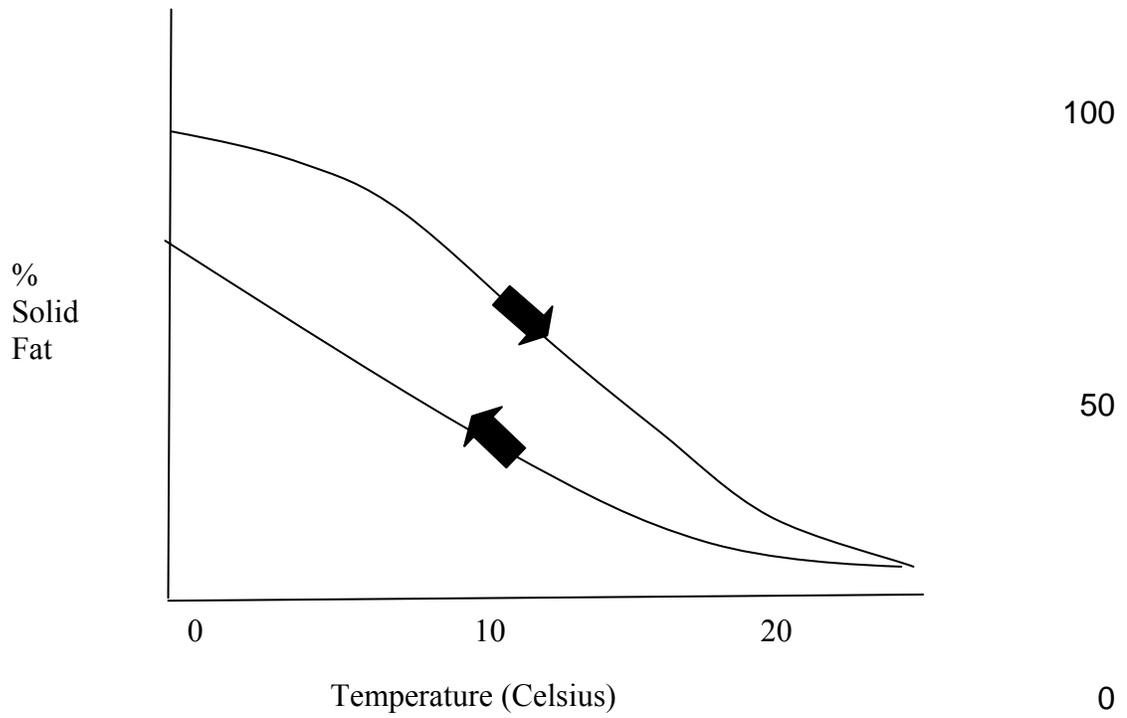


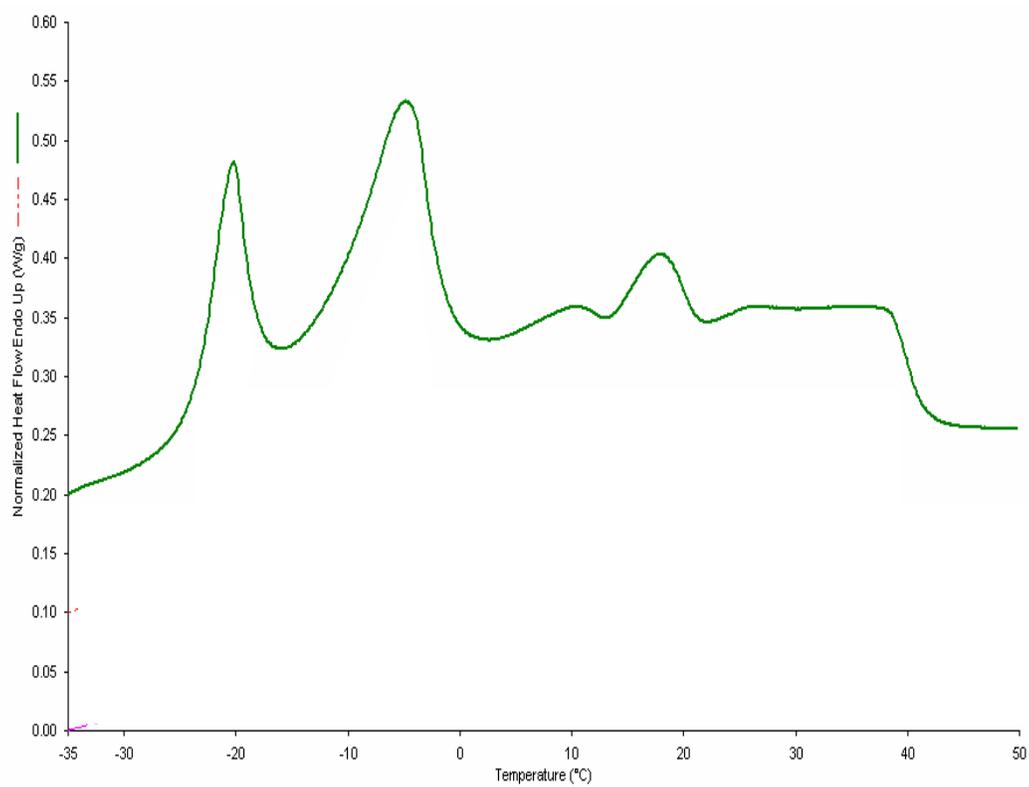
Figure 1.3 The microstructure of butter. Adapted from Walstra 1999.



**Figure 1.4 Amount of solid fat as temperature increases. Adapted from Prentice, 1992.**



**1.5 Hysteresis curve of solid fat of butter. Adapted from Prentice, 1992.**



**Figure 1.6 DSC scan of butter.**

**Table 1.1 MFGM components. Adapted from Fox, 1998.**

<b>Component</b>	<b>% (w/w) of Total Membrane</b>
Protein	41
Phospholipid	27
Crebrosides	3
Cholesterol	2
Neutral glycerides	14
Water	13
<b>Total</b>	<b>100</b>

**Table 1.2 Lipid composition of MFGM. Adapted from Keenan and Dylewski, 1983.**

<b>Component</b>	<b>Proportion of Total Lipids</b>
Triacylglycerols	62
Diacylglycerols	9
Monoacylglycerols	Trace
Sterols	0.2-2
Sterol Esters	0.1-0.3
Unesterified fatty acids	0.6-6
Hydrocarbons	1.2
Phospholipids	26-31

**Table 1.3 The most prevalent fatty acids in milkfat. Adapted from Riel, 1985.**

<b>Fatty Acid</b>	<b>Average Percent of Total Fatty Acids</b>	
<b><u>Saturated Fatty Acids</u></b>		
Butyric	4:0	3.4
Caproic	6:0	1.3
Caprylic	8:0	1.2
Capric	10:0	2.2
Lauric	12:0	3.9
Myristic	14:0	13.1
Palmitic	16:0	25.3
Stearic	18:0	10.6
Arachidic	20:0	1.3
Behenic	22:0	trace
		<b>62.3</b>
<b><u>Mono-unsaturated Fatty Acids</u></b>		
Caproleic	10:1	0.2
Lauroleic	12:1	0.3
Myristoleic	14:1	1.3
Palmioleic	16:1	3.7
Oleic	18:1	30.8
Caccenic, gadoleic acid	18:1	0.7
		<b>37.0</b>
<b><u>Polyunsaturated Fatty Acids</u></b>		
Linoleic	18:2	3.2
Arachidonic	20:4	1.1
		<b>4.3</b>
<b>TOTAL 100</b>		

**Table 1.4 Characteristic off-flavors in butter. Adapted from Bodyfelt 1988, USDA 1999.**

<b>Off-flavor</b>	<b>Characteristics</b>	<b>Cause</b>
Acid/Sour	Intense acid, sharp, sour taste on tip of the tongue, “buttermilk flavor,” no aftertaste	High-acid cream, overripened cream, excessive use of lactic culture, excessive retention of buttermilk
Aged	Lack of freshness, moderately persistent aftertaste (not to be confused with “storage” or “old cream” off-flavors in USDA grading)	Holding butter for extended period at relatively high temperatures
Bitter	Bitter taste, distinct, lingering aftertaste	Microorganisms, enzymes, specific feeds or weeds, late lactation milk, impurities in butter salt, inappropriate use of neutralizers
Briny/High Salt	Sharp salt, salty taste beyond range of ordinary acceptability	Too much salt, uneven salt and water distribution
Cheesy	Taste and aroma of Cheddar cheese, often accompanied by bitter aftertaste	Hydrolyzed protein by microorganisms, lightly salted/unsalted more susceptible
Coarse	Lacking sweet, pleasing, delicate flavor associated with fresh milkfat, harshness of flavor, lacks balance, reasonably good, but “just falls short”	Commingling some fresh high-quality cream with lower quality cream with slight acid development
Cooked	Smooth, nutty, custard-like, if not scorched-desirable flavor	Pasteurizing sweet cream at severely high temperatures, inadequate agitation, improper neutralizing
Feed	Aromatic, disappears quickly in aftertaste, may be bitter,	Alfalfa, grass-fed cows, too short amount of time between feeding and milking
Fishy	Flavor and aroma similar to codfish, cod-liver oil, or fish meal	High acid, high salt, overworking, elevated levels of metallic salts in cream
Flat	Lacks full, pleasing, “buttery” flavor	Lack of volatile acids, low content of diacetyl, low levels of volatile compounds, excessive washing, dilution of churning cream with water
Foreign	Odors or tastes of cleaning products, gasoline, sprays, paint, etc.	Residual cleaning products, sanitation chemicals, etc.
Garlic or Onion	Flavor or odor of garlic or onion	
Malty	Malted odor or “Grape Nuts” cereal, black walnuts	<i>Streptococcus lactis</i> var. <i>maltigenes</i> in improperly cooled milk or cream
Metallic	Astringent, puckery, iron, lingering aftertaste, may be bitter	Storing cream in direct contact with metals
Musty	Odor of poorly ventilated cellar, potatoes, swamp, musty or poorly cured hay	<i>Pseudomonas taetrolens</i> , storing cream in a dampy, musty space, improperly cleaned cream separators, cows fed musty smelling

**Table 1.4 Continued**

		feeds, slough grass, stagnant water
Neutralizer	Soda cracker-like, alkaline, bitter aftertaste, "limey," observed readily upon inhalation after expelling sample from mouth	Highly concentrated neutralizer, excessive amount of neutralizer
Old cream	Staleness, lack of freshness, unpleasant "background" odor, flavor lag, lingering off-flavor	Several days old, improperly washed, cans, utensils, processing equipment
Oxidized	Metal-induced, cardboardy, puckery mouthfeel,	Oxidation of unsaturated fatty acids
Rancid (lipase)	Soapy, delayed bitterness, darkened, decayed nut meats, volatile fatty acids, astringent, unclean aftertaste	Hydrolysis of milkfat by lipase to yield free short-chain fatty acids
Storage	Flavor deterioration, absence of freshness	Deterioration due to extended storage, stored next to odorous foods
Tallowy	Odor and taste of tallow, aftertaste, may just be on the surface	Extensive oxidation of unsaturated fatty acids, high storage temperatures in presence of light and metal contamination (Cu, Fe)
Unclean/Utensil	Unclean, dishrag, dirty socks, barny, cowy, smothered	Poor cream handling conditions, improper sanitation, spoilage bacteria
Weedy	Wild onion, wild garlic, unpleasant flavor	Churning cream that has absorbed by weed taint
Whey	Similar to coarse/acid flavor, aftertaste, moderate odor	Cream separated from cheese whey
Yeasty	Fruity, vinegary, yeasty, slightly fragrant, ethanol-like, yeast-raised bread, bitter	Summer butter production, poor handling, abused cream, by-products from yeast growth

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**Table 1.5. Textural defects of butter identified by the USDA. Adapted from Bodyfelt 1988, USDA 1999**

Textural Defect	Characteristics	Structural Reason	Processing Factors
<b>Crumbly</b>	Fat particles crumbly or brittle, lack of cohesion, some butter adheres to trier, rough appearance	Large fat crystals, too little liquid fat	Cottonseed meal and alfalfa hay fed to cows, temperature of cooling after processing, length of holding period, churning, wash water temperatures
<b>Greasy</b>	Extreme smoothness, immediate melting in mouth, greasy sensation	High proportion of lower melting triglycerides	Over working
<b>Gummy</b>	Gum-like, sticks to the roof of mouth	High percentage of high-melting triglycerides	Cottonseed products used for feed
<b>Leaky</b>	Beads or droplets of moisture on plug and back of trier	Large water droplets	Insufficient working
<b>Mealy/Grainy</b>	Grainy like cornmeal mush, not smooth and waxy	Not determined	Improper standardizing of high acid cream, melting frozen cream, allowing butter to oil off in pasteurizer, remelting butter scraps
<b>Ragged boring</b>	Full trier of butter can not be drawn up, butter rolls from trier rather than cutting a full plug	Not determined	Rate of cooling of cream after pasteurization, temperature before and during churning and of wash water
<b>Short</b>	Lacks plasticity and waxiness, plug breaks sharply with thumb pressure	High-melting point fats which contain relatively small fat globules	Part of milkfat is melted (normal granules not formed), rapid cooling of freshly made butter to a very low temperature
<b>Sticky</b>	Adheres to trier, dry	High-melting point triglycerides	Over-working, often concurrent with crumbly, alfalfa feed, churn working conditions, temperature treatment of cream
<b>Weak/Spongy</b>	Quick melt, exaggerated softness, imperfect plug	Incomplete milkfat crystallization, excess liquid milkfat, high proportion of low-melting triglycerides	Inadequate cooling of pasteurized cream,

**CHAPTER 2.**

**IDENTIFICATION OF FLAVOR CHARACTERISTICS THAT DRIVE  
CONSUMER LIKING OF BUTTER AND VEGETABLE OIL SPREADS**

**Submitted to *Food Quality and Preference* 7-18-06**

## **ABSTRACT**

This study identified the sensory characteristics that drive consumer liking of butter and vegetable oil spreads to aid marketing strategies for declining butter consumption. A trained descriptive panel evaluated 29 commercial butters and spreads using a defined sensory language. Two focus groups were conducted with butter consumers to gain an understanding of usage and consumption habits. Eight representative butters and spreads were selected for consumer acceptance testing. Both internal and external preference mapping techniques were applied to interpret consumer data. Key discriminating sensory characteristics of butter included color intensity, diacetyl/cultured, cooked/nutty, grassy/feed, milkfat flavors, and salty taste ( $p < 0.05$ ). From focus groups, key butter features were desirable flavor and natural image. Negative aspects included price and cholesterol. Five consumer clusters with distinct butter/spread likes and dislikes were identified. Butter acceptability varies among consumers and butters with specific sensory characteristics could be marketed to specific target market segments.

### **Key Words:**

Butter, spreads, consumer preference, segmentation, preference mapping, descriptive sensory analysis

## INTRODUCTION

Butter is a highly prized fat source for its rich flavor attributes. A wide array of butter is sold, varying widely in color, texture, and flavor. Butter flavor and texture is affected by type of animal the milk comes from (cow, goat, sheep, etc.), diet, stage of lactation, dietary supplementation, and season of the year which it is produced (Bobe, et al., 2003, Chen, et al., 2004, Hawke and Taylor, 1994, Nickerson, 1995, Ramaswamy, et al., 2001). Additionally, processing, storage conditions, addition of starter culture and salt contribute to the diverse flavor profile of butter. Most Americans are familiar with sweet cream butter which is produced without the addition of starter culture, and may or may not have added salt.

Butter consumption is currently stagnant; consumption per capita in the United States increased from 4.5 to 4.6 lbs over the period of 2000-2004. Production during this time period has also remained relatively constant, down only 0.5% from the 1,256 million pounds produced in 2000 (International Dairy Foods Association, 2005). Health concerns have been often been cited as the reason many consumers decreased their butter consumption and increased their intake of margarine (Crane, 1993). By definition, butter contains 80% fat (USDA, 1989). Over 60% of this fat is saturated (Riel, 1985). Since milkfat comes from an animal source, it is also a significant contributor of cholesterol.

Margarine and vegetable oil spreads, in contrast, are made from vegetable oil and contain no cholesterol. Margarine itself is not without controversy though, as it has come under fire recently for its high *trans* fat content. The hydrogenated oils that are used to make margarine contain *trans* fatty acids which may be linked to chronic

heart disease and may contribute to increases in cholesterol levels (Stauffer, 1996a). The legal definition for margarine, similar to butter, is that it contains 80% fat (FDA, 2005). Many softer spreads contain less fat and by definition cannot be labeled as margarine. Per capita consumption of margarine in the United States declined between 2000 and 2004; from 7.5lbs per year to 5.3lbs per year (USDA-ERS, 2006).

Numerous studies have evaluated consumer acceptance of margarine spreads and butter/oil blends (Avramis, et al., 2003, Chen, et al., 2004, Kim, et al., 2005, Michicich, et al., 1999). Kim et al. (2005) made cold spreadable butter-vegetable oil blends with structured lipids that could potentially reduce cholesterol. This blend was compared to regular canola oil-butter blends and butter. The flavor of the spread was not significantly different than that of traditional butter, but spreadability was different. Michicich et al. (1999) examined consumption of two new spreads, one made with a cholesterol-reduced lard and vegetable oil blend (Appetize® Lard), and a dairy spread made with reduced-cholesterol anhydrous milkfat, in comparison to butter and margarine. Butter consumers consumed significantly more spreads than margarine consumers and butter consumers even ate more of the spreads they liked less. There were no significant differences in acceptance between any of the spreads for margarine consumers. Crane (1993) used phone surveys to examine consumer understanding of health effects, price, taste, and usage of butter and margarine. They reported that over half of consumers surveyed thought that margarine contained less cholesterol, fat, and calories than butter. Only one-third of respondents indicated price was a factor in their decision to buy butter or margarine. Hellemann, Tuorila, Lampi, & Matuszewska (1990) examined acceptability of spreads. Brand name

ratings did not correlate well with consumer panel scores. The perception of spreadability of butter based on color was probed by Rohm, Strobl, & Jaros (1997). Under normal light, consumers reported that yellow butter was significantly easier to spread than its counterpart despite both samples having the same instrumental yield value. Under red-light conditions, the perception of spreadability was not significantly different.

To our knowledge, no studies have examined consumer attitudes toward the varying flavors exhibited among butters and compared them to margarine or vegetable oil spreads. This study was conducted to identify the sensory characteristics (primarily specific flavors) that drive consumer acceptance of butter and spreads. A descriptive sensory language for butter and vegetable oil spreads was identified. This language was then used to document the sensory characteristics of butter and margarines. Consumer acceptance testing was applied followed by internal and external preference mapping. These results will help manufacturers understand what different segments of the market prefer and how to best accommodate these market segments.

## **MATERIALS AND METHODS**

### ***Butter/margarine descriptive analysis***

A descriptive sensory language was first identified to characterize butter and margarine/spread flavor attributes. Fifty-six butters were screened and discussed by five sensory and dairy experts in three 2 h sessions. Samples included butters aged one week to two years, cultured, non-cultured, salted, unsalted, organic/pasture-fed, domestic, international, as well as goat and sheep milk butters. A sensory lexicon for

butter was created from the terms generated at these sessions. Ten commercial margarines and spreads were subsequently screened by these individuals and two terms were added to the lexicon (Table 1).

Twenty-seven representative commercial butters (consisting of international, domestic, fresh, cultured, organic/pasture-fed, salted and unsalted, and aged butters) and two vegetable oil spreads (one with buttermilk/cultured flavors, 60% fat, and a traditional margarine-type spread, 60% fat) (Table 2) were then evaluated by a trained descriptive panel using the defined sensory language. Commercial products were purchased and stored in the dark at 5C until analysis. The outer 0.3cm was trimmed to avoid flavors due to packaging or exposure. Testing was conducted in accordance with the NCSU Institutional Review Board for Human Subjects approval.

Nine panelists (seven women, two men) between 21 and 45 years of age, were selected based on time, availability, and previous experience in sensory analysis of dairy flavors. All panelists were experienced with the Spectrum<sup>TM</sup> method of descriptive analysis (> 60 h experience each) and participated in 40 hours of additional training on butter and margarine/spread flavor with the identified sensory language (Meilgaard, et al., 1999). During training, panelists evaluated and discussed samples in order to ensure panelist and panel consistency and understanding of the lexicon. Analysis of variance of data collected from the last part of training indicated that the panel and panelists could consistently use the attributes to differentiate the products.

Descriptive analysis of butters/margarines/spreads was conducted by each panelist in quadruplicate replications in a randomized balanced design. Panelists

individually evaluated 7g samples presented in 2oz plastic cups with plastic lids (Sweetheart Cup Co., Owings Mills, MD, U.S.A) in sensory booths. All sample cups were labeled with a three digit code and samples were tempered to 19C. This temperature was chosen since panelists could best detect subtle differences in flavor when samples were tempered to this temperature. Panelists evaluated six samples per session, and they were given room temperature deionized water and unsalted crackers to cleanse their palate between samples.

### ***Focus groups***

Two focus groups (8 females, ages 24-40 y and 8 females > 46 y) were conducted to gain a better understanding of butter/margarine/spread usage and consumption habits. A screener was filled out by the participants to obtain demographic information and background information on butter and non-butter spread usage. Focus groups lasted approximately 1.5 hours.

An experienced moderator asked the participants a series of pre-determined questions in a round-table format. Subjects were first asked about their consumption habits of butter and margarines/spreads (frequency, on what occasions, etc.) Attitudes on health and usage were probed and brand and type of butter used (salted/unsalted) were also discussed.

Towards the end of the discussion subjects were given tempered (19C) samples of stick margarine, spreadable (tub) margarine, stick butter, and spreadable butter to evaluate and discuss. All samples were identified by a three-digit code. Color, texture (spreadability, hardness, etc.), and flavor of the samples were discussed. Following this, the participants were given the same four samples with

labels that indicated type of sample and brand. Attitudes toward the brands were discussed. Focus groups were video-taped and tape-recorded for subsequent reference. Key points from focus groups were used to develop the consumer questionnaire and ballot.

### ***Consumer Testing***

Based on the descriptive sensory data means and examination of principle component plots, six representative butters were selected for consumer testing based on the attributes: salty taste, diacetyl/cultured, cooked/milky, yellow color, grassy/feed, and refrigerator/stale flavors. Two vegetable oil spreads (60% vegetable oil) were included; one represented a typical salted stick spread and one represented a “cultured, butter-flavored” vegetable-oil stick spread.

Samples (7g) were placed into 2 oz plastic cups with plastic lids (Sweetheart Cup Co., Owings Mills, MD, U.S.A), numbered with a three-digit code, and tempered to 19C. Samples were evaluated individually in sensory booths and were presented in a randomized balanced order. Consumers were recruited via e-mail, classified advertisements, and flyers. All participants were screened for allergies to dairy products. Testing was conducted in accordance with the NCSU Institutional Review Board for Human Subjects guidelines. Subjects were given ambient temperature de-ionized water to cleanse their palates between samples. Compusense® version 5.0 (Compusense, Guelph, Canada) was used for data collection. Demographic information as well as butter and spread usage information were collected. Additionally, panelists were asked about the occasions and how often they used butter and/or margarine/spreads, what factors affected their purchases, whether they viewed

butter and margarine/spreads as natural, and whether they viewed butter as healthier than margarine/spreads. For the last two question categories, consumers were provided with the statement “Butter (or margarine) is a natural product” and “Butter is healthier than margarine.” Consumers were then asked to indicate how they felt about each statement (agree strongly, agree, neither agree nor disagree, disagree, disagree strongly).

During the sample evaluation, panelists were instructed to spread the sample on a piece of white pita bread (previously screened and determined to be bland by the trained panel, Neomonde Bakery, Raleigh, NC) or to consume it without the bread if they desired. Participants were asked to evaluate overall acceptance, appearance liking, color intensity, color liking, salty taste intensity, salty taste liking, freshness intensity, texture liking, and flavor liking. All attributes were evaluated using a 9-point hedonic scale where “high intensity/like extremely” (score=9) and “low intensity/dislike extremely” (score=1). Subjects received food treats and a gift certificate for their participation.

### ***Statistical Analysis***

Univariate and multivariate techniques were used to analyze the data. Statistical analysis of descriptive data was conducted using SAS (version 9.2, Cary, NC). Analysis of variance with means separation and principle component analysis (PCA) of descriptive data were performed to identify characteristic differences between the samples. Analysis of variance with means separation as well as internal preference mapping (PCA of consumer liking data) was conducted on consumer data (SAS, version 9.2). Frequency counts were tabulated for consumer demographic,

habit, and attitude information. The Pearson Chi-square test was used to identify significant associations and trends between demographic, habit, and attribute and identified consumer segments. These analyses were performed using SPSS® version 12.0 (Chicago, IL, USA).

Partial Least Square regression 2 (PLS2) was used to construct external preference maps. PLS2 focuses on explaining the variation in Y-variables (Consumer-liking matrix; 8 products, 161 consumers) by using the descriptive sensory results (Biasioli, 2006; Martens and Martens, 2001). Important descriptive attributes were identified using the Jack-knife method. PLS2 and Jack-knife optimization methods were performed using the Unscrambler® version 9.2 (CAMO, Oslo, Norway). Two attributes (vegetable oil/fatty and sweet aromatic (not dairy)) which were exclusively associated with margarine/spreads were excluded from the analysis because the attributes did not contribute any variation in the data set.

Two-Step Cluster analysis (TCA) was performed on individual consumer coordinates within the preference space generated by PLS2. TCA was performed using Log-likelihood as a distance measure and number of segments was automatically determined using the combination of changes in Akaike's Information Criterion (AIC) and greatest changes in the distance when clusters were divided sequentially (Anonymous, 2001; Chiu, Fang, Cheng, Wang, & Jeris, 2001; Zhang, Ramakrishnon, & Livny, 1996; Banfield and Raftery, 1993). Discriminant analysis (DA) with cross-validation was used to confirm and determine final segmentation from TCA with at least 95% correct allocation. TCA and DA were performed using SPSS® version 12.0 (Chicago, IL, USA).

After segmentation, the liking profiles for each segment were generated. Means of liking attributes were estimated using 2-way ANOVA without an interaction term (consumer = random effect and sample = fixed effect). Tukey-HSD multiple comparison was performed on the significant sample effect for each liking attribute at the 95% confidence level. PCA was performed to study correlations between overall liking and other attribute likings in order to gauge important aspects of butter and margarine. The analysis was performed on the whole data set and within each segment using Maximum Likelihood extraction and Varimax rotation. PCA and ANOVA were performed using SPSS® version 12.0 (Chicago, IL, USA).

## **RESULTS**

### ***Descriptive Analysis***

The identified sensory language differentiated the butters and spreads (Table 3). Principle component plots are shown in Figures 1 and 2. PC1 and PC2 accounted for 24 and 18% of the variability, respectively. PC3 and PC4 accounted for 16% and 11% of the variability, respectively. Two terms, painty and prickly, were identified in butters used for language generation but were not identified in the samples used in this study. Other attributes were identified in a few samples. Waxy/animal was only identified in butters made from goat or sheep's milk. Methyl ketone, free fatty acid, and fecal/mothball were also documented in a few of the international samples. Other terms were associated with butter types. Sour taste and diacetyl were associated with cultured butters. The two non-butter spreads had very distinct sensory profiles which differentiated them from the other butters. They were characterized by distinct

intensities of vegetable oil/fatty and non-dairy sweet aromatic. These flavors were not detected in butters.

### ***Focus Groups***

Participants indicated they generally used butter several times a week or sparingly. Those that used it sparingly, consumed margarine/spreads on a regular basis and used butter only for special occasions. There was one person in each group that only consumed margarine. For both groups, it was noted that consumers appeared to primarily fall into two groups: butter only consumers and margarine/butter consumers. The latter group was very diverse. Individuals who consumed margarine on a regular basis quite often used butter for special occasions or for baking.

Many of the older participants indicated that the negative health aspects of butter (high fat, cholesterol, and calories) were a deciding factor in their purchase and consumption of butter. The younger group was generally not concerned with the health aspects of butter, and butter was viewed as about as healthy as margarine. The consensus among these consumers was that butter and margarine were best consumed in moderation. Many in the younger group viewed butter as a natural product. Across both focus groups, most women indicated they preferred butter for baking uses, as it was prized for its flavor. Most purchased butter in quarter pound sticks, salted or unsalted depending on desired use and personal preference. Most participants indicated that they decided which brand to buy based on prices and sales.

Most participants indicated that margarine had a distinctive odor and a deeper yellow color than butter. Light yellow color was deemed desirable for butter. Butter

was more difficult for the subjects to spread, and some preferred the texture of the whipped butter. The majority of subjects preferred the taste of butter over margarine. Many people could identify which samples were butter and which were margarine. None were surprised by which brand went with which sample.

### ***Consumer Results***

Consistent with focus group results, consumers primarily fell into two groups: butter-only (n=52) and butter/margarine (n=107) consumers. Two of the 161 consumers polled were margarine-only consumers. Their results were excluded from analysis. User category did influence consumer usage and perception for certain things (Table 4). Presence/absence of salt and spreadability influenced purchase decision differently for the two user groups ( $p<0.05$ ). Presence/absence of salt played a larger role in the purchase decision for butter-only users while not surprisingly, spreadability played a larger role for butter/margarine consumers. Butter only users were generally more in agreement that “Butter is healthier than margarine” compared to butter/margarine users. Both consumer groups generally agreed that “Butter is a natural product” while more butter/margarine users than butter-only users were more positive or neutral with the statement “Margarine is a natural product” ( $p<0.05$ ).

Overall, consumers indicated distinct differences and likings for butters and margarines (Table 5, Figure 3). Product 23 was the best-liked product. This product was a domestic sweet cream butter that is nationally marketed. Perhaps not surprisingly it was best-liked when averaged across all consumers. Products 21, 27, and 28 scored the lowest overall acceptance scores. Product 28 was a vegetable oil

spread and 21 and 27 were unsalted and salted butters, respectively. Examination of descriptive data did not reveal any common attributes among these three products.

### ***External Preference Mapping***

External preference mapping was applied to further explore consumer likes and dislikes for the butters and vegetable oil spreads. The PLS sample liking scores are presented in Figure 4 and the loadings of the descriptive sensory attributes are presented in Figure 5. In Figure 4, the x axis, which explains the most variability, primarily separates salted and unsalted butters. The y axis separates the two vegetable oil spreads from the butters and is comprised of yellow color and cooked flavor. The subsequent set of PLS sample loadings (Figures 6 and 7, 19 and 12% of the variability, respectively) differentiated products based on presence/absence of methyl ketone flavor and yellow color intensity. Overall, consumers clearly perceived differences between vegetable oil spreads and butters and unsalted and salted products. Consumers did not perceive a lot of differences between butters 23, 25, and 27 although low intensities of refrigerator/stale flavors were detected in butter 25 by trained panelists. The fresh butter (high cooked/nutty flavor, butter 24) and the international butter (yellow color, methyl ketone, grassy feed flavor, butter 16) were moderately differentiated.

Two-step cluster analysis was applied to PLS2 results to more clearly identify specific consumer groups. This analysis yielded five distinct preference segments of butter and margarine consumers (Figure 8). Segment 1 (n=42) contained traditional butter lovers. This is where the largest number of butter only consumers was found (n=21). These consumers liked traditional butter flavors: cooked/milky, milkfat

flavor and even refrigerator/stale flavors were acceptable. The presence/absence of salt did not matter. Margarines, yellow color and unusual butter flavors such as grassy/feed were not desirable. The top 2 butters for these consumers were P23 and P25.

Segment 2 (n=34) contained primarily margarine lovers. The largest number of margarine/butter users were found in this segment (n=29). The traditional vegetable oil spread (P29) was their favorite. Their next choice was essentially a tie between traditional butter (P23) and "butter-style" margarine (P28). They liked the unsalted butter least by a wide margin. Consumers in segment 3 (n=21) preferred butters with unusual flavors. Salt was not a driver. Butters with high yellow color, feed/grassy, methyl ketone, or stale flavors were their preferred butters. Their favorite butters were P16 and P21. The picky/discerning butter consumers comprised segment 4 (n=30). Samples P23 and P24 were their top picks (low yellow, milkfat, cooked/milky). Salty taste was preferred over unsalted. Stale flavored butters were least preferred by these consumers, below vegetable oil spreads. Consumers in segment 5 (n=31) like salted butter. They generally liked all butter flavors: cooked/milky, milkfat, grassy/feed, methyl ketone, stale, as well as salty taste. Vegetable oil spreads were not well-liked, and unsalted butter was liked less than vegetable oil spreads.

## **DISCUSSION**

All butters were differentiated by descriptive analysis. A wide range of flavors and flavor intensities were documented. Clearly butters and spreads represent a wide range of sensory characteristics. Similar descriptive techniques have been

used to differentiate other products including cheese, peanut butter, chocolate milk and many others (Young, Drake, Lopetcharat, & McDaniel, 2004; McNeill, Sanders, & Civile, 2002; Thompson, Drake, Lopetchararat, & Yates, 2004).

Most participants in the two focus groups in this study indicated that butter was preferred over margarine. However, many didn't consume butter often due for economic or health reasons. Similarly, participants in focus groups carried out by Wright (1991) preferred the taste of butter but were concerned about health issues associated with it. Likewise, Crane (1993) found that 69% of consumers surveyed over the telephone agreed that they preferred the taste of butter to margarine. Crane also found that 93% of participants said that concern over health influenced their butter/margarine purchase decision. While consumers surveyed by Crane agreed (86%) that butter was more expensive than margarine, only 36% said that price influenced their purchase decision. In this study, many focus group subjects considered price as a factor in their purchase decision and a majority of the participants in our quantitative consumer test did as well (60% of butter-only consumers and 72% of butter/margarine consumers).

Wright (1991) found that many participants indicated they used butter for everyday uses and margarine in baking applications. Of participants interviewed by Crane (1993), 78% used margarine for cooking. In this study, many focus group participants indicated the opposite, that they used butter for some applications (baking, special occasions) and margarine/spreads for others (every day use). Participants in the consumer test who were classified as butter/margarine users tended to use more butter than margarine for cooking/sautéing and baking (75% and 74% use

butter and 64% and 46% use margarine for cooking/sautéing and baking, respectively). Butter/margarine consumers tended to use butter on special occasions more than margarine (56% and 22%, respectively). Over half of butter/margarine users (53%) consumed butter less than four times per month.

Crane (1993) found that consumers were not well informed about the differences between butter and margarine including hydrogenation and *trans* fat content. Our focus group participants were generally informed on the nutritional differences between butter and margarine. In quantitative consumer tests, there was a difference in opinion between butter-only users and butter/margarine users on whether butter is healthier than margarine. Of the butter-only users, 58% strongly agreed or agreed, while only 28% of the butter/margarine users believed this statement to be true. The shift in knowledge of *trans* and saturated fat content may be due to recently implemented regulations by the Food and Drug Administration requiring labeling of *trans* fats (FDA, 2003) and more attention on saturated and *trans* fat and their negative health consequences.

The “natural” image of butter was emphasized by our focus group participants as a positive aspect of butter. Results from the consumer test confirmed this; the majority of both butter-only and butter/margarine users strongly agreed or agreed that butter is a natural product (88% and 84% respectively). This natural image of butter is an attribute consumers value and that manufacturers should emphasize. The market for less processed and natural foods is among the fastest growing market segments according to a report by the Agricultural and Marketing and Research Center (Norwood, 2004).

Overall, butter was preferred over the vegetable oil spreads across all preference segments. This finding is consistent with the findings of Hellemann et al. (1990). Crane (1993) also found that the majority of consumers surveyed agreed that they like the taste of butter better than margarine. Nearly all of the participants in the consumer test, with the exception of two, consumed butter at least occasionally. This suggests that butter is a product that most people enjoy, even those consumers who also eat margarine or only those who only consume butter occasionally. There is an opportunity for butter manufacturers to reach out to the majority of margarine/spread consumers who enjoy the flavor of butter equally or more than margarine/spreads but do not consume/purchase it for other reasons such as price, health, and spreadability.

Spreadability had a larger influence on purchase decision for butter/margarine users than for butter-only consumers ( $p < 0.05$ ) (41% and 17%, respectively). Across all segments, texture liking trends generally reflected overall liking results (results not shown). Consumers in segment 2 (“margarine lovers”) rated both vegetable-oil spreads (P28 and P29) as their favorite in terms of texture. While texture does appear to be an issue of importance to many consumers, products in this study were tempered to more clearly distinguish flavor differences and thus some of the textures of the samples may not accurately depict texture directly from refrigerated storage or how the consumer would temper them prior to use.

Consumption trends suggested that butter and margarine usage separates consumers into three categories, the majority—those who use both butter and margarines and spreads, a smaller portion who use only butter, and a smaller segment who exclusively use margarine/spreads. Butter and butter/margarine consumers can

further be divided into five segments. The largest amount of butter-only users fell into the “traditional” butter-lover category (segment 1). A large number of butter-only consumers desired flavors traditionally associated with sweet-cream butter and they do not accept any unusual flavors. The butter/margarine consumers were distributed throughout the five segments, with the largest percentage (27%) falling into segment 2, the “margarine-lover” category. This group liked “traditional” vegetable oil spread the best, and “cultured-type” vegetable oil spread and a nationally marketed brand of butter about the same. This leaves over 70% of butter/margarine consumers in the other categories, indicating that although they consume margarine/spreads, they like butter better.

Moskowitz (2001) examined drivers of margarine liking and was able to segment participants into two groups based on attributes that were desirable for margarine consumption. The first group was identified as liking lighter color, intermediate flavor strength, and intermediate softness in texture; Moskowitz calls this group the “low impact” seekers. In this study, the “low impact” seekers may be similar to individuals in segment 1 who tended to like “traditional” butter flavors, lighter colors, and rejected extremely soft textures. This group found the butter with the highest yellow intensity (P16) to be unacceptable in terms of color, texture, and overall acceptance. They also found the texture of the vegetable oil spreads to be less acceptable than the majority of butters. The second segment of margarine consumers designated by Moskowitz (2000) was comprised of consumers who liked darker colors, stronger flavors, and a very soft texture. This second group may be somewhat analogous to segments 3 and 5 in this study, who are accepting of more unusual

flavors and colors. The “margarine lovers” (segment 2) also fit into the second group from the study done by Moskowitz (2000). Segment 2 rated the softest textures and darkest colors (vegetable oil spreads and P16) as the most desirable. Segment 4 in this study showed a combination of desirable attributes.

## **CONCLUSIONS**

The market for butter and margarine/spreads contains a vast variety of consumers who purchase these products for many different uses. Factors that influence purchase decision are different for butter-only users and butter/margarine consumers. Acceptance of butter and spreads differs across consumer segments and is based on many characteristics with specific segments preferring specific butter and spread flavor profiles.

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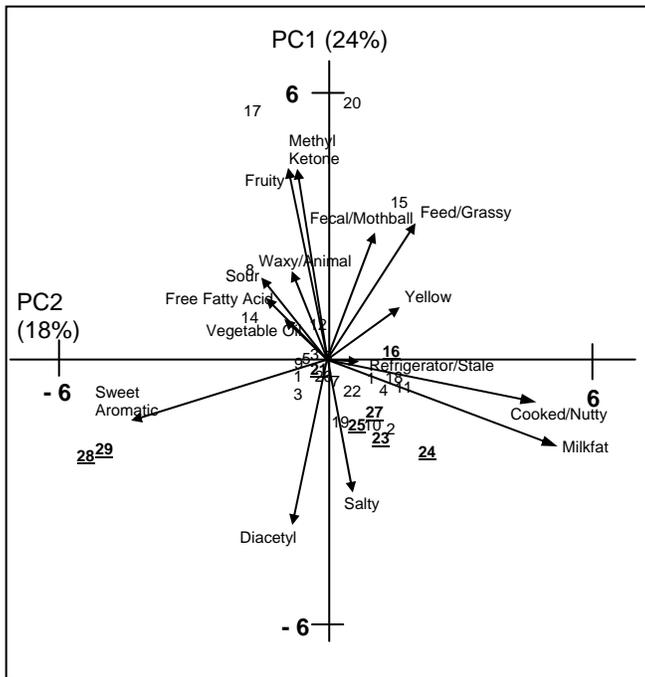
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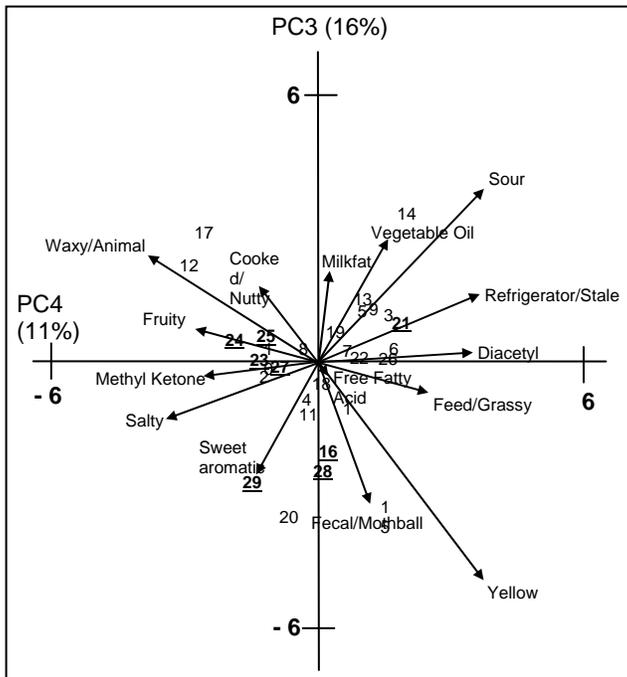
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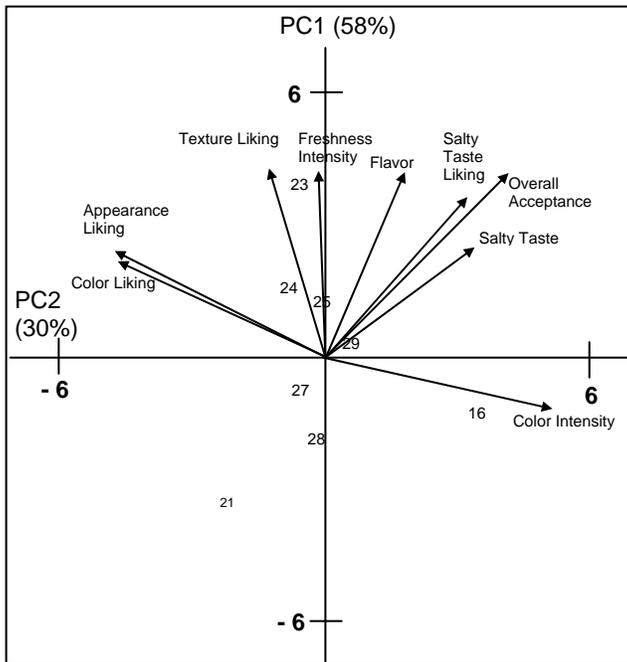
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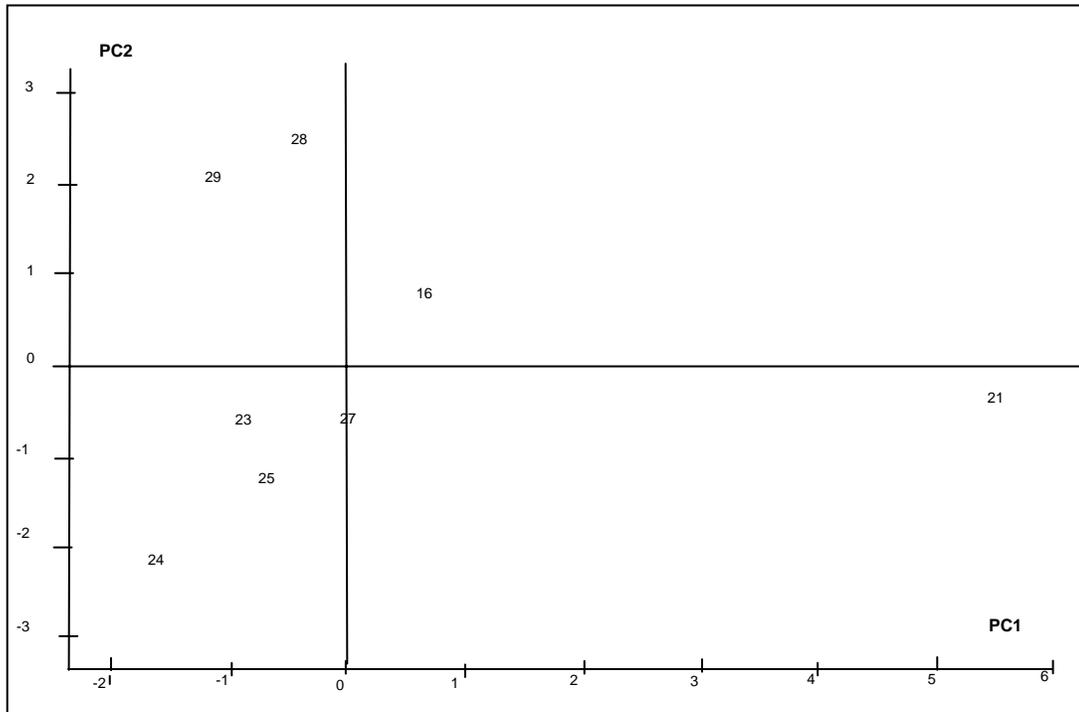
**Figure 2.1 Principle component biplot of descriptive sensory analysis of commercial butters and vegetable oil spreads. Numbers represent samples (Table 2). Underlined numbers represent those chosen for consumer testing. PC1 = principle component 1; PC2 = principle component 2.**



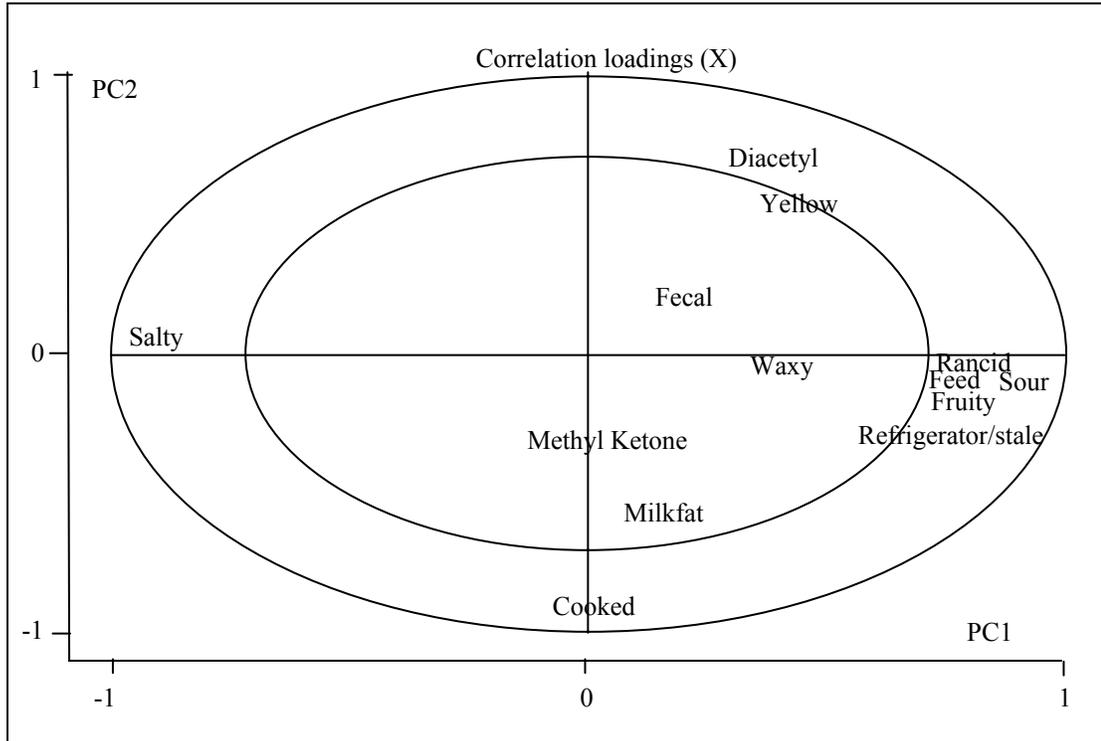
**Figure 2.2 Principle component biplot of descriptive sensory analysis of commercial butters and vegetable oil spreads. Numbers represent samples (Table 2). Underlined numbers represent those chosen for consumer testing. PC3 = principle component 3; PC4 = principle component 4.**



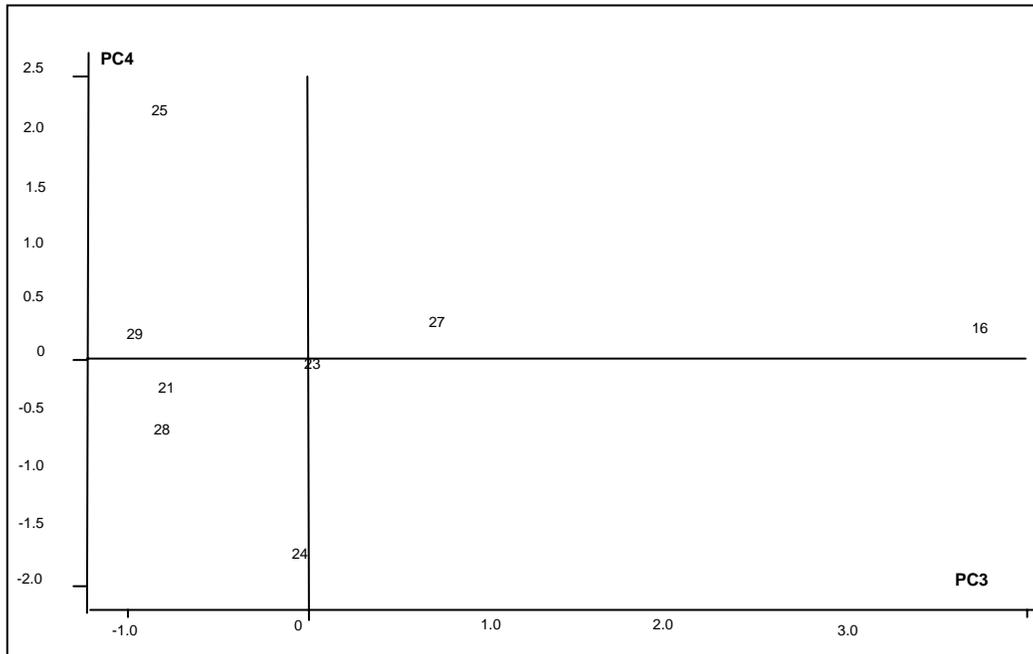
**Figure 2.3 Internal preference map of consumer results. Numbers represent samples (Table 2). PC1 = principle component 2; PC2 = principle component 2.**



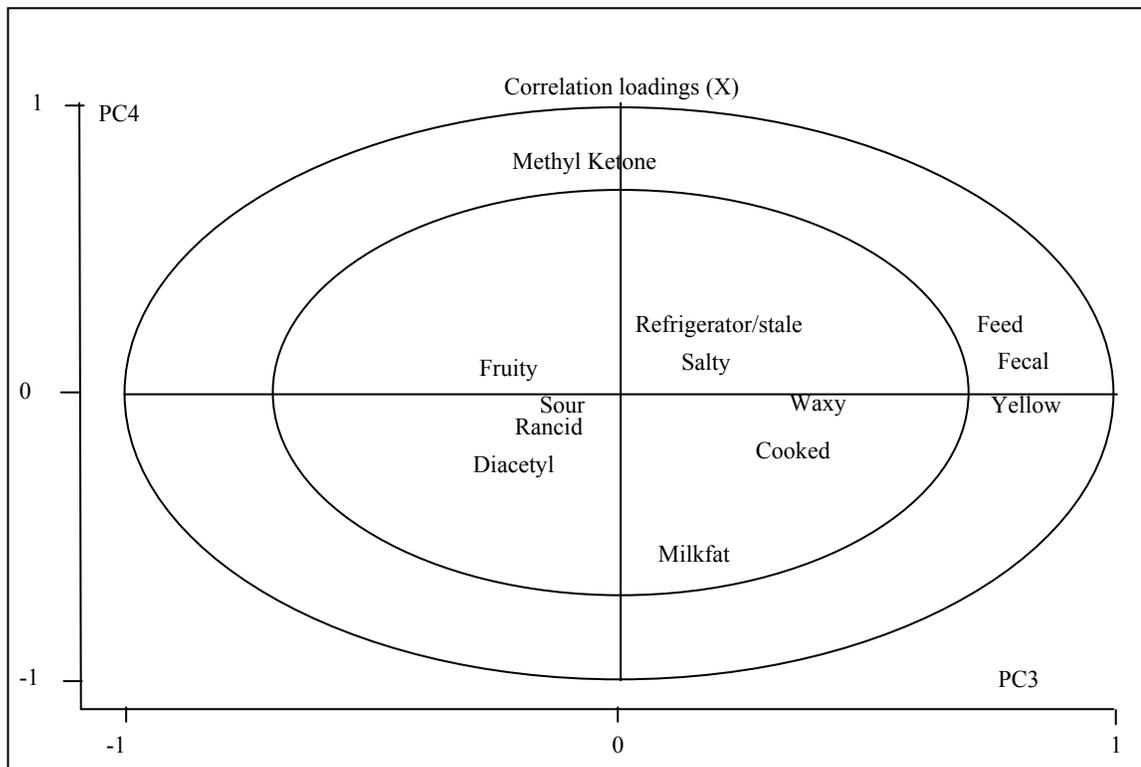
**Figure 2.4** Partial least squares model of consumer scores. Loading plot of PC1 versus PC2. PC1 explains 40%; PC2 explains 20%. Numbers indicate samples (Table 2).



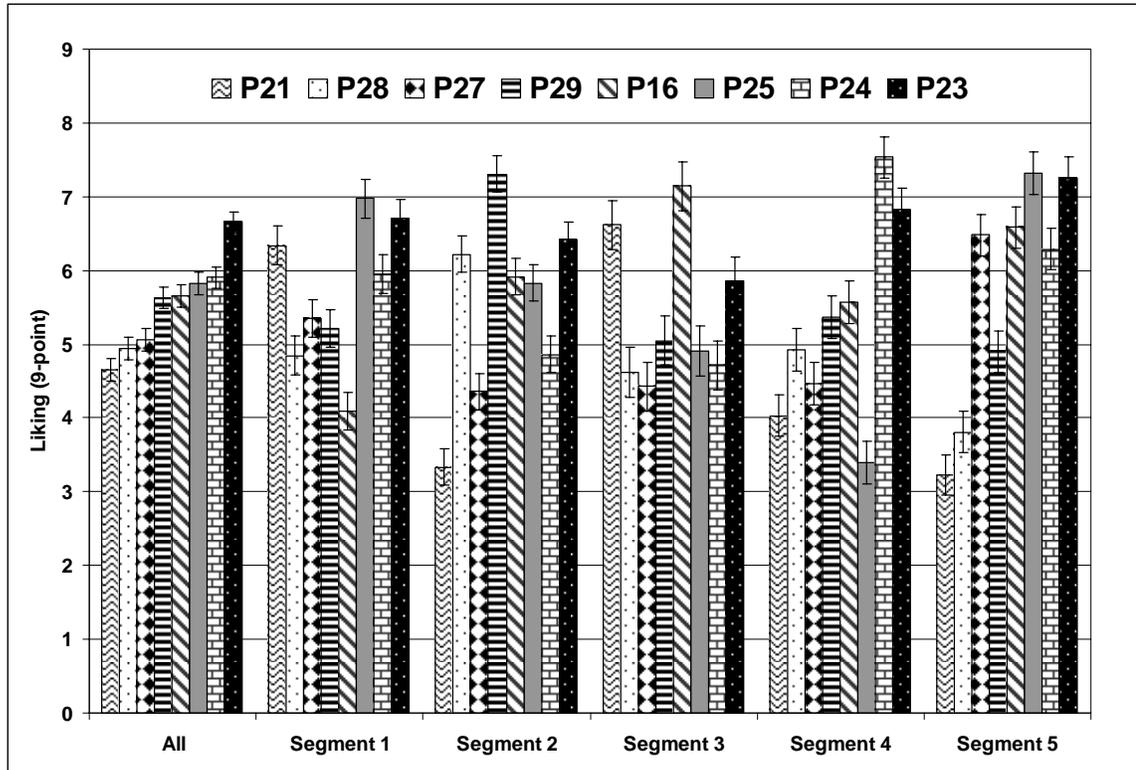
**Figure 2.5** Correlation loadings for PC1 and PC2 using the Partial Least Squares model of descriptive attributes. PC1 explains 40%; PC2 explains 20%. Numbers indicate samples (Table 2).



**Figure 2.6 Sample Partial least squares model of consumer scores. Loading plot of PC3 versus PC4. PC3 explains 19%; PC4 explains 12%. Numbers indicate samples (Table 2).**



**Figure 2.7** Correlation loadings for PC3 and PC4 using the Partial Least Squares model of descriptive attributes. PC3 explains 19%; PC4 explains 12%. Numbers indicate samples (Table 2).



**Figure 2.8 Overall acceptability scores for butter and spreads within different consumer segments. Acceptability was scored based on a 9-point hedonic scale where 1 = dislike extremely and 9 = like extremely**

**Table 2.1 Sensory language used for the descriptive sensory analysis of butter and margarines/spreads**

<b>Term</b>	<b>Definition</b>	<b>Reference</b>
Diacetyl/Cultured <sup>a</sup>	Sweet aromatic characteristic of cultured dairy products, of which diacetyl is a primary source	Diacetyl, 20ppm
Milkfat/Lactone	Aromatic characteristic of milkfat, lactones, and coconut	Heavy cream
Cooked/Nutty	Aromatic associated with cooked milk and canned corn	1% fat milk heated in a microwave for 8 min
Refrigerator/Stale	Stale aromatic characteristic of refrigerator with old food left in it	Butter quarters (sticks) stored in a refrigerator for 18 mo
Feed/Grassy	Aromatics associated with grasses and feeds	Grass clippings; hexanal 20 ppm
Dairy Sour	Sour aromatics associated with sour cream or plain yogurt	Plain yogurt, sour cream
Painty	Aromatics associated with wall paint and oxidized fats	Linseed oil
Fecal/Mothball <sup>a</sup>	Aromatics associated with mothballs; associated with complex protein degradation	Mothballs; indole, skatole, 20 ppm
Waxy/Animal <sup>a</sup>	Waxy/crayon like aromatic, commonly associated with goat's or sheep's milk products; aromatics associated with medium chain fatty acids	4-methyl octanoic acid 143 ppb; 4-ethyl octanoic acid 187 ppb
Vegetable oil/Fatty*	Aromatics associated with vegetable oil	Crisco soybean oil
Sweet aromatic (not milkfat or diacetyl)*	Sweet aromatic, not dairy in nature	Freshly purchased Parkay margarine
Free Fatty Acid <sup>a</sup>	Aromatics associated with short chain fatty acids	Butyric acid, 20 ppm
Methyl Ketone <sup>a</sup>	Aromatics associated with blue-veined cheeses	2-octanone, 40 ppm
Fruity <sup>a</sup>	Aromatics associated with different fruits	Fresh pineapple; ethyl hexanoate 20 ppm
Prickle <sup>a</sup>	Chemical feeling factor of which the sensation of carbonation on the tongue is typical	Soda water
Salty <sup>c</sup>	Taste elicited by NaCl	Sodium chloride solutions; 0.5%, 0.7%, 0.9%
Yellow <sup>d</sup>	Intensity of yellow color	Yellow color scale

<sup>a</sup> Reference taken from Drake, McIngvale, Gerard, Cadwallader, & Civille, 2001

<sup>b</sup> Reference taken from Carunchia Whetstine, Karagul Yuceer, Avsar, & Drake, 2003

<sup>c</sup> Reference taken from Meilgaard, Civille, & Carr, 1999

<sup>d</sup> Reference taken from Kornerup & Wanscher, 1978

\* Only detected in margarines/spreads

**Table 2.2 Butters and spreads used for descriptive analysis<sup>a</sup>**

<b>Treatment</b>	<b>Type<sup>b</sup></b>	<b>Salt Content<sup>b</sup></b>	<b>Country<sup>b</sup></b>	<b>Age</b>
1	Cultured	Salted	France	Within package date
2	Sweet Cream	Salted	USA	Within package date
3	Cultured	Unsalted	Italy	Within package date
4	Sweet Cream	Salted	USA	Within package date
5	Cultured	Unsalted	USA	Within package date
6	Sweet Cream	Unsalted	USA	Within package date
7	Cultured	Unsalted	Denmark	Within package date
8	Cultured	Salted	USA-Southeast	Within package date
9	Sweet Cream	Unsalted	USA	Within package date
10	Sweet Cream	Salted	USA	Within package date
11	Sweet Cream	Salted	England	Within package date
12	Goat's milk	Salted	France	Within package date
13	Cultured	Unsalted	Spain	Within package date
14	Cultured	Salted	USA-Southeast	Within package date
15	Sweet Cream	Unsalted	Australia	Within package date
<b>16</b>	<b>Sweet Cream</b>	<b>Salted</b>	<b>Ireland</b>	<b>Within package date</b>
17	Sheep's milk	Unsalted	Greece	Within package date
18	Sweet Cream	Salted	USA-West Coast	Within package date
19	Sweet Cream	Salted	USA	24 mo. at 5C
20	Cultured	Unsalted	New Zealand	Within package date
<b>21</b>	<b>Cultured</b>	<b>Unsalted</b>	<b>USA-Northeast</b>	<b>Within package date</b>
22	Sweet Cream	Salted	France	Within package date
<b>23</b>	<b>Sweet Cream</b>	<b>Salted</b>	<b>USA-Midwest</b>	<b>Within package date</b>
<b>24</b>	<b>Sweet Cream</b>	<b>Salted</b>	<b>USA-West Coast</b>	<b>Within 48 hrs of production</b>
<b>25</b>	<b>Sweet Cream</b>	<b>Salted</b>	<b>USA-West Coast</b>	<b>15 mo. at 5C</b>
26	Cultured	Salted	USA-Northeast	Within package date
<b>27</b>	<b>Sweet Cream</b>	<b>Salted</b>	<b>USA</b>	<b>Within package date</b>
<b>28</b>	<b>70% vegetable oil</b>	<b>Salted</b>	<b>USA</b>	<b>Within package date</b>
<b>29</b>	<b>70% vegetable oil</b>	<b>Salted</b>	<b>USA</b>	<b>Within package date</b>

<sup>a</sup> Products in bold were used for consumer testing

<sup>b</sup> Determined from product label

**Table 2.3 Means separation for descriptive analysis of commercial butters and spreads.**

Attribute/ Treatment	Diacetyl	Cook ed/ Nutty	Milkfat	Salty	Refrigerator/ Stale	Feed/ Grassy	Sour	Fecal/ Mothball	Waxy/ Animal	Fruity	Methyl Ketone	Free Fatty Acid	Yellow	Vegeta ble Oil	Sweet Aromatic (not milkfat)
<b>1</b>	1.6	3.3	3.57	9.13	ND	2.2	ND	ND	ND	ND	ND	ND	3.62	ND	ND
<b>2</b>	ND	3.87	4.04	10.22	ND	ND	ND	ND	ND	ND	ND	ND	2.68	ND	ND
<b>3</b>	ND	2.72	3.32	ND	1.31	ND	1.6	ND	ND	ND	ND	ND	2.03	ND	ND
<b>4</b>	ND	3.35	3.73	9.05	ND	1.91	ND	ND	ND	ND	ND	ND	2.96	ND	ND
<b>5</b>	ND	2.8	3.36	ND	1.54	1.72	1.57	ND	ND	ND	ND	ND	1.73	ND	ND
<b>6</b>	1.96	2.96	3.6	ND	1.51	1.8	1.65	ND	ND	ND	ND	ND	2.65	ND	ND
<b>7</b>	1.57	3.09	3.67	ND	ND	1.57	ND	ND	ND	ND	ND	ND	2.35	ND	ND
<b>8</b>	ND	2.52	2.62	4.07	ND	ND	1.77	ND	ND	ND	1.65	1.30	2.34	ND	ND
<b>9</b>	1.77	2.79	3.36	ND	1.86	1.53	1.5	ND	ND	ND	ND	ND	1.94	ND	ND
<b>10</b>	ND	3.66	3.84	9.45	ND	ND	ND	ND	ND	ND	ND	ND	2.01	ND	ND
<b>11</b>	ND	3.38	3.61	9.82	ND	1.12	ND	ND	ND	ND	ND	ND	3.38	ND	ND
<b>12</b>	ND	3.42	3.6	8.29	ND	1.65	ND	ND	2.36	ND	ND	ND	1.16	ND	ND
<b>13</b>	1.69	2.91	3.63	ND	ND	ND	1.9	ND	ND	ND	ND	ND	1.68	ND	ND
<b>14</b>	1.44	3.26	3.14	5.4	ND	0.74	2.36	ND	ND	ND	ND	ND	1.52	ND	ND
<b>15</b>	ND	2.97	3.36	ND	ND	2.25	ND	1.7	ND	ND	ND	ND	3.72	ND	ND
<b>16</b>	ND	3.27	3.56	8.35	ND	1.86	ND	1.5	ND	ND	ND	ND	3.41	ND	ND
<b>17</b>	ND	2.46	3.02	ND	ND	2.03	1.68	ND	1.63	1.74	2.07	ND	1.42	ND	ND
<b>18</b>	ND	2.39	3.89	7.77	ND	0.93	ND	ND	ND	ND	ND	ND	3.05	ND	ND
<b>19</b>	ND	2.94	3.38	9.42	2.54	ND	ND	ND	ND	ND	ND	ND	1.94	ND	ND
<b>20</b>	ND	2.61	3.05	ND	ND	2.10	ND	1.4	ND	1.56	2.38	ND	4.23	ND	ND
<b>21</b>	2.17	2.97	3.54	ND	1.57	1.61	1.85	ND	ND	ND	ND	ND	2.38	ND	ND
<b>22</b>	1.59	3.19	3.42	9.51	1.76	2.03	ND	ND	ND	ND	ND	ND	2.50	ND	ND
<b>23</b>	ND	3.9	3.9	9.72	ND	ND	ND	ND	ND	ND	ND	ND	1.91	ND	ND
<b>24</b>	ND	5.66	4.09	10	ND	ND	ND	ND	ND	ND	ND	ND	1.85	ND	ND
<b>25</b>	ND	3.48	3.73	10.1	1.2	ND	ND	ND	ND	ND	ND	ND	1.80	ND	ND
<b>26</b>	2.34	3.16	3.49	6.65	ND	2.00	1.80	ND	ND	ND	ND	ND	3.09	ND	ND
<b>27</b>	ND	3.6	3.73	9.55	ND	ND	ND	ND	ND	ND	ND	ND	2.30	ND	ND
<b>28</b>	3.75	ND	ND	10.5	ND	ND	ND	ND	ND	ND	ND	ND	2.60	2.50	1.5
<b>29</b>	1.5	ND	ND	9	ND	ND	ND	ND	ND	ND	ND	ND	2.20	3.50	2.31
<b>LSD</b>	<b>0.24</b>	<b>0.23</b>	<b>0.2</b>	<b>0.78</b>	<b>0.86</b>	<b>0.31</b>	<b>0.22</b>	<b>0.14</b>	<b>0.14</b>	<b>0.13</b>	<b>0.16</b>	<b>0.09</b>	<b>0.18</b>	<b>0.12</b>	<b>0.02</b>

ND = not detected

LSD = least significant difference. Means in a row that differ by more than LSD are different (p<0.05)

Underlined products were used for consumer testing

Painty and prickle were not detected in any of the samples

Attributes were evaluated by trained panelists using the 15 point universal Spectrum™ intensity scale where 1 = very low intensity and 15 = highest possible intensity (Meilgaard et al., 1999)

**Table 2.4 Gender, age, and butter and spread consumption characteristics of consumers**

	<b>Butter only consumers (N=52)</b>	<b>Butter and margarine consumers (N=107)</b>
Gender (% male/female)	42% Male 58% Female	36% Male 64% Female
Age group	22% 18 to 24 41% 25 to 35 37% >36 y	36% 18 to 24 34% 25 to 35 30% >36 y
Shop for household	86% Shop for household 14% Do not shop for household	94% Shop for household 6% Do not shop for household
Butter usage*	16% Less than once a month 16% 2-4 times a month 51% More than once a week 17% Everyday	20% Less than once a month 33% 2-4 times a month 41% More than once a week 6% Everyday
Non-butter spreads (margarine) usage	N/A	23% Less than once a month 34% 2-4 times a month 37% More than once a week 6% Everyday
Type of butter usage	83 % Cooking/sautéing 79 % Baking 62 % On vegetables 77 % On Bread 35 % On popcorn 52 % Special occasions	75 % Cooking/sautéing 74 % Baking 49 % On vegetables 73 % On Bread 36 % On popcorn 56 % Special occasions
Type of margarine usage	N/A	64 % Cooking/sautéing 46 % Baking 52 % On vegetables 72 % On Bread 21 % On popcorn 22 % Special occasions
Factors that influence purchase decision	60 % Price 52 % Salted/unsalted* 17 % Availability 29 % Brand 4 % Organic 39 % Health 17 % Spreadability* 48 % Flavor 8 % Package	72 % Price 34 % Salted/unsalted* 26 % Availability 36 % Brand 8 % Organic 47 % Health 41 % Spreadability* 64 % Flavor 8 % Package
Agreement with the statement: "Butter is healthier than margarine"*	25 % Agree strongly 33 % Agree 27 % Neither agree nor disagree 15% Disagree 0 % Disagree strongly	5 % Agree strongly 23 % Agree 43 % Neither agree nor disagree 27 % Disagree 3 % Disagree strongly
Agreement with the statement: "Butter is a natural product."	31 % Agree strongly 58 % Agree 10 % Neither agree nor disagree 2 % Disagree 0 % Disagree strongly	25 % Agree strongly 59 % Agree 14 % Neither agree nor disagree 2 % Disagree 0 % Disagree strongly
Agreement with the statement: "Margarine is a natural product."*	2 % Agree strongly 4 % Agree 10 % Neither agree nor disagree 62 % Disagree 23 % Disagree strongly	0 % Agree strongly 10 % Agree 22 % Neither agree nor disagree 55 % Disagree 13 % Disagree strongly

\*Indicates differences between the two groups (p<0.05)

**Table 2.5 Consumer acceptance scores for commercial butters and spreads**

<b>Attribute / Treatment</b>	<b>Overall Acceptance</b>	<b>Appearance liking</b>	<b>Color Intensity</b>	<b>Color Liking</b>	<b>Salty Intensity</b>	<b>Salty Taste Liking</b>	<b>Freshness Intensity</b>	<b>Texture Liking</b>	<b>Flavor Liking</b>
<b>16</b>	5.65	5.34	7.43	5.18	5.33	5.71	5.52	5.75	5.57
<b>21</b>	4.66	6.21	4.91	6.05	3.04	4.16	5.13	5.66	4.34
<b>23</b>	6.67	6.68	5.13	6.46	5.30	6.08	6.52	6.73	6.50
<b>24</b>	5.87	6.34	4.65	6.15	4.96	5.52	6.08	6.47	5.75
<b>25</b>	5.86	6.21	4.52	5.92	5.69	5.58	5.98	6.25	5.74
<b>27</b>	5.10	6.35	5.63	6.07	5.34	5.20	5.37	6.07	4.84
<b>28</b>	4.98	6.04	5.44	5.95	4.99	5.17	5.24	5.65	4.68
<b>29</b>	5.64	6.23	6.36	6.19	6.02	5.49	5.53	6.00	5.49
<b>LSD</b>	<b>0.41</b>	<b>0.36</b>	<b>0.29</b>	<b>0.38</b>	<b>0.36</b>	<b>0.40</b>	<b>0.36</b>	<b>0.39</b>	<b>0.44</b>

Products were scored using a 9-point scale where 1 = low intensity/dislike extremely and 9 = high intensity/like extremely

LSD-least significant difference

Means in a column that differ by more than the LSD are significantly different (P<0.05)

## **CHAPTER 3**

### **THE EFFECT OF REFRIGERATED AND FROZEN STORAGE ON BUTTER FLAVOR AND TEXTURE**

## **INTERPRETIVE SUMMARY**

Shortened title: Storage effects on butter flavor and texture

KRAUSE

Butter is often stored for several months in refrigerated or frozen storage.

Deterioration of flavor and texture may occur during this time. This study examined the effect of refrigeration (5C) and frozen (-20C) storage on the physical properties and flavor of butter. Changes were monitored through descriptive sensory analysis of flavor, texture, and color by a trained panel using a defined sensory language.

Additionally, physical changes were monitored through oxidative stability index, peroxide value, free fatty acid value, vane, instrumental color, and oil turbidity.

Currently no specifications exist for butter storage. This study will help to establish guidelines for the industry so they can better market and distribute stored butter.

## **ABSTRACT**

Butter is often stored for extended periods of time; therefore, it is important for manufacturers to know the refrigerated and frozen shelf-life. The objectives of this study were to characterize the effect of refrigerated and frozen storage on the sensory and physical characteristics of butter. Fresh butter was obtained on two occasions from two facilities in quarter pound sticks and nine pound bulk blocks (2 facilities, 2 package forms). Butters were placed into both frozen (-20°C) and refrigerated storage (5°C). Frozen butters were sampled after 0, 6, 12, and 15 months; refrigerated butters were sampled after 0, 3, 6, 9, 12, and 15 months. After 6 and 12 month frozen storage, butters were also removed and placed in refrigerated storage for 3 and 6 months. Every 3 months, oxidative stability index (OSI) and descriptive sensory analysis (texture, flavor, and color) were conducted. Every 6 months, peroxide value (PV), free fatty acid value (FFA), fatty acid profiling, differential scanning calorimetry (DSC), vane, instrumental color, and oil turbidity were examined. Analysis of variance was conducted to characterize the effects of storage time, temperature, and package type. Storage time, temperature, and package type impacted butter flavor, OSI, PV, and FFA ( $p < 0.05$ ). Refrigerated butter quarters exhibited refrigerator/stale off-flavors concurrent with increased levels of oxidation (lower oxidative stability and higher PV and FFA) within 6 months. Off-flavors and oxidation were not evident in bulk refrigerated butter until 9 months. Frozen butter quarters after 6 months of refrigeration showed no differences from fresh butter, but low levels of off flavors were evident following 12 month storage. Bulk butter showed no sensory changes through 15 mo frozen storage. Since butter is such a

highly prized fat source in terms of its flavor and textural properties, it is important that manufacturers understand how long their product can be stored before negative attributes develop. These off-flavors could potentially carry-through to applications and negatively impact consumer perception.

**KEY WORDS**

Butter, storage temperature, butter quality, descriptive sensory analysis, vane, oxidative stability

## INTRODUCTION

Dairy manufacturers produce large amounts of butter in the winter months. It is often necessary to store this butter for extended periods of time until there is a demand for it. During refrigerated and/or frozen storage, degradation of quality may occur and this is an important issue when companies develop specification sheets for butter suppliers or for butter suppliers to design storage regimes. Butter is commonly stored for extended periods in blocks (25 kg) which are subsequently re-worked into quarter-pound sticks. However, retail packages (sticks or quarters as they are referred to by industry) are also often stored for extended periods.

Previous studies have been conducted to examine the effect of storage time on butter flavor. Emmons et al. (1986) examined the effect of different wrapping types on one-pound butter prints stored at -18C and found no deterioration over a 12-month period. Butter stored for 14 weeks at 5C was found to have some deterioration in flavor. Butter flavor was analyzed by trained graders so statistical analysis of results, relative intensities, and the exact nature of the flavor degradation were not characterized. Jebson et al. (1974) examined storage of 25kg blocks of butter packaged in parchment paper inside a fiber-board box at -18C, -10C, -4C, and 4C for eight months. Grading and peroxide value were used to evaluate butter quality. The coldest storage temperature (-18C), did not yield significantly higher quality butter than the -10C storage over the eight month time period. From these results, it was concluded that -10C was the best storage temperature in terms of convenience and butter quality.

A variety of studies have examined butter wrapping materials. Parchment paper was found to be a source of pro-oxidants (copper, iron, and sulphuric acid) by Pont (1961). Downey and Murphy (1968) found that off-flavors in butter were related to the amount of light that the wrapper was able to transmit. MacBean and Chandler (1974) compared cellophanes, low and high density polyethylenes, and polypropylenes to vegetable parchment. Samples were stored under accelerated conditions (62 days at 5C, with three four-day periods where the butter was stored at ambient temperature) to simulate one year of frozen storage. Grading, peroxide value, and mold count were performed to measure degree of degradation. High density polyethylene was most similar to vegetable parchment in terms of quality maintenance though it was concluded that cost would preclude its usage. Tomlinson and Dixon (1977) confirmed the findings of MacBean and Chandler (1974) by evaluating similar butter wrapping materials over 34 weeks at sub-zero storage temperatures. Polyethylene films provided the best protection against surface oxidation and provided the best freeze-thaw stability.

Few studies have evaluated butter texture. Daubert et al. (1998) used a rheological test, the vane method, to measure yield stress and yield strain of commercial items including margarine spreads at 19C and 7C. From these measurements, a map of spreadability was created. Mortensen and Danmark (1982) determined that yield stress was an ample measure of spreadability in butter as instrumental results were correlated to a trained sensory panel. Fearon and Johnston (1989) used probe penetration, instrumental texture profile analysis, and cone penetrometry to examine spreadability of mechanically worked butter at 5 and 15C.

Several studies have correlated sensory analysis of butter texture to analytical measurements. Trained panel results were correlated to apparent yield value, extrusion force, penetration force and sectility by Rohm and Ulberth (1989). Apparent yield value was best correlated to spreadability, and sectility correlated most closely to firmness. The sliding pin consistometer was correlated to untrained sensory analysis of spreadability and hardness by Davey and Jones (1985). Kawanari et al. (1981) compared instrumental butter texture data to that of a trained texture panel. Three tests: shear, uniaxial compression, and penetrometry, were correlated to trained descriptive sensory results. Rousseau and Marangoni (1999) also found that cone penetrometry was highly correlated to trained sensory spreadability profiles.

To our knowledge, recent studies have not addressed butter storage stability. Further, stability of bulk and stick butter have not been compared. Our objective was to evaluate the flavor and texture stability of bulk and stick butter across frozen (-20C) and refrigerated (5C) storage. Descriptive sensory analysis, which has not been previously applied to butter flavor, was used to monitor flavor and texture. Instrumental methods were also used to evaluate chemical and texture changes.

## **MATERIALS AND METHODS**

### ***Production and Sampling***

Two butter production facilities (CA, USA) were sampled on two different days. On each of the two days, bulk butter (25 kg blocks split into 4 kg blocks and packaged individually in polyethylene bags, Grade AA, salted; 122.5 kg from each facility on each day) and stick butter (113g wax paper-wrapped sticks, in packages of four, Grade AA, salted, 49 kg each day from each facility) were obtained by

overnight shipment on ice packs. A total of 735 kg of butter was received at the beginning of February 2005 (245 kg as sticks and 490 kg as bulk blocks). Fat, moisture, and salt content were analyzed upon receipt. Products were assigned to refrigerated (5°C) and frozen (-20°C) storage conditions and stored in the dark. Sample information and temperature conditions are listed in Table 3.1.

Every three months, samples were pulled from refrigerated storage and every six months, samples were taken from frozen storage. At each frozen storage time point, samples were removed and placed into a refrigerator (5°C). At three month time points, oxidative stability index (OSI) was evaluated and sensory analysis was performed. At six months all tests were performed (color, peroxide value, oxidative stability index, free fatty acid value, vane, sensory analysis, turbidity). Fatty acid profiling was conducted initially and after 6 mo. Tests were performed within two weeks of the sampling date. The outer 1 cm of bulk samples was trimmed before evaluations to prevent flavors due to packaging. The outer 2 mm of butter quarters was trimmed to remove any discoloring.

### ***Descriptive Sensory Analysis***

Color, flavor, and texture attributes (Table 3.2) were evaluated across storage. Eight panelists (7 females, 1 male) were selected based on availability and previous experience (>75 h each) with descriptive sensory analysis of dairy products using the Spectrum<sup>TM</sup> method (Meilgaard et al., 1999). Panelists received an additional 25 h of training to focus on identification and scaling of butter flavor and texture attributes. During training, panelists discussed and evaluated an array of commercial butters. The Spectrum<sup>TM</sup> universal scale was used to scale the intensity of flavor attributes

(Meilgaard et al., 1999) using the language described by Krause et al. (2006) (Table 2). Color intensity/hue was evaluated using the scale adapted by Krause et al. (2006) (Table 3.2). A 10-point product specific scale was used to score butter spreadability and firmness (Table 3.2). Prior to testing, analysis of variance of panel and panelist performance on selected butters was used to determine that panelists could consistently identify and scale butter color, flavor and texture attributes. Two weeks prior to each testing timepoint, panelists received an additional 3 h of refresher training and calibration, and panel and panelist performance on butter sensory attributes were once again confirmed to be consistent.

For sensory analysis, samples (7g) were prepared with the overhead lights turned off to prevent light-induced flavor changes, and placed in 2 oz soufflé cups (Sweetheart Cup Company Inc, Owings, MD) and stored at 5°C in the dark. One and a half hours before the panel session they were tempered to 19°C. This temperature was chosen for sensory analysis since panel training sessions indicated that panelists could best identify subtle variations in butter flavor at this temperature and this was also the temperature used for vane texture analysis. Panelists individually evaluated samples under white lights using paper ballots or computerized data entry (Compusense 5 v4.6, Compusense, Guelph, Canada) in individual booths in a positive air pressure room dedicated to sensory analysis. Each treatment was evaluated in duplicate by each panelist. For flavor evaluations, two warm-up samples, butters that had previously been profiled by the panel, were provided with their consensus flavor profiles along with salty taste solution references (Table 2). For texture evaluation, panelists were provided with firmness and spreadability references (Table 2). To

evaluate color, panelists were instructed to compare the sample to a provided color scale. Flavor/color and texture were evaluated in separate sessions. Panelists were given de-ionized water and unsalted saltine crackers between samples for palate cleansing. To prevent temporal cues from unduly influencing panelists, at each timepoint, a fresh butter (less than 72 h old) was included.

### ***Solid Butter Color***

Two 10g samples were taken and pressed into separate 60 x 15mm polystyrene petri dishes (Falcon® 1007, Becton Dickerson, Franklin Lakes, NJ). Five measurements of L\* a\* b\* were taken at random places on each petri dish with a Minolta Colorimeter (Konica Minolta, Tokyo, Japan). The samples were evaluated at 19°C.

### ***Centrifuged Oil Color***

Two samples (45g each) were placed into 50mL conical centrifuge tube (Falcon®, Becton Dickinson Labware, Lincoln Park, NJ). The tubes were wrapped in foil and placed in a 50°C water bath for 8 minutes to melt the butter followed by centrifugation (Model 225, Fisher Scientific, Fairlawn, NJ) for 12 minutes at 3400xG in a 50°C oven (Despatch Industries, Minneapolis, MN). The top oil layer was pipetted off and combined. An aliquot of oil (5g) was placed into each of two Petri dishes (60mm x 15mm polystyrene, Falcon® 1007, Becton Dickerson Labware, Franklin Lakes, NJ). Five measurements of L\* a\* b\* were taken at random places on each petri dish with a Minolta Colorimeter (Konica Minolata). The oil samples were evaluated at 50°C.

### ***Oxidative Stability Index (OSI)***

Two samples (45g each) of butter were placed into 50mL conical centrifuge tube (Falcon®, Becton Dickinson Labware, Lincoln Park, NJ) and prepared as described for oil color measurements. An aliquot of oil (5g) was placed into each of three 100mL glass disposable OSI tubes (Omnion Inc., Rockland, MA). The OSI tubes were placed in the Oxidative Stability Instrument (Omnion, Inc., Rockland, MA) and a conductivity meter was inserted. Air at 5psi was connected. The temperature was set at 110°C. Testing was conducted in duplicate until a peak in conductivity was recorded by the computer.

### ***Peroxide Value***

Peroxide value (PV) was modified from AOCS Official Method 965.33. Briefly, two samples (45g each) of butter were placed into a 50mL conical centrifuge tube (Falcon®, Becton Dickinson Labware, Lincoln Park, NJ), wrapped in foil and placed in a 50°C water bath for 8 minutes. Following this, they were centrifuged (Model 225, Fisher Scientific, Fairlawn, NJ) for 12 minutes at 3400xG in a 50°C oven (Despatch Industries, Minneapolis, MN). The top oil layer was pipetted off and combined. An aliquot of oil (5g) was placed into each of three 250mL Erlenmeyer flasks. Thirty mL of 3:2 acetic acid/chloroform (both Fisher Scientific, Fairlawn, NJ) and 0.5mL saturated potassium iodide (Fisher Scientific, Fairlawn, NJ) solution was added. After one minute, 30mL of deionized water was added. The flasks were titrated with 0.01M sodium thiosulfate (Fisher Scientific, Fairlawn, NJ) until disappearance of yellow color. Starch solution (1%, J.T. Baker Chemical Co., Phillipsburg, NJ) was added to the flask (0.5mL). The titration was continued until the blue color disappeared. PV (milliequivalent peroxide/kg oil) was calculated as:

(mL sodium thiosulfate) x (molarity of sodium thiosulfate) x 1000/(sample weight in grams).

### ***Free Fatty Acid Value***

Two samples (45g each) of butter were placed into 50mL conical centrifuge tube (Falcon®, Becton Dickinson Labware, Lincoln Park, NJ) and prepared as described for oil color measurements. An aliquot of oil (7.05g) was placed into each of three 250mL Erlenmeyer flasks. Fifty mL neutralized isopropyl alcohol (99% neutralized with NaOH to a faint pink color, both Fisher Scientific, Fairlawn, NJ) and 1 mL of indicator (phenolphthalein 1% (w/v) in 95% ethanol, both Fisher Scientific, Fairlawn, NJ) was added to the flask. The solution was titrated with .025N NaOH until a faint pink color was maintained for a minute. Free fatty acid value was calculated as follows: (mL NaOH x Normality NaOH x 40)/g oil, as measured in mg NaOH/g oil.

### ***Vane rheometry***

The vane test was used as an instrumental texture analysis. The vane test has been used successfully to evaluate products such as ice cream (Briggs, et al., 1996), peanut butter, margarine spreads, sour cream, whipped topping (Daubert et al., 1998) and cream cheese (Breidinger and Steffe, 2001). Yield stress and apparent yield strain have together been used to create texture maps indicative of spreadability. The yield stress was calculated from the amount of torque necessary to rotate the vane through the butter. The apparent yield strain was calculated from the time to maximum yield stress. A Haake VT550 (Thermo Electron Corporation, Waltham, MA) was used. Butter samples were tempered in a 19°C incubator until they reached

an internal temperature of 18°C. Sample was forced into a metal box (3.5cm x 3.5cm x 7.5cm (inside diameter) mounted on a 17.6cm x 4cm metal plate). The metal box was clamped to a stand. When the internal temperature of the butter reached 19°C, the vane was inserted (1.0 cm diameter, 2.5cm height). The instrument rotated at .02rpm. The peak torque was used to calculate the yield stress by single-point method (Steffe, 1996):

$$\sigma = 2M_0/\pi d^3 (h/d + 1/(m+6))^{-1}$$

where:

$\sigma$  = yield stress, Pa

$M_0$  = maximum torque in N m

$d$  = diameter of the vane (.01m)

$h$  = height of the inserted vane (.025m)

$m$  = constant, dimensionless

Studies have shown the assumption that  $m = 0$  to be valid (Daubert et al., 1998, Yoo et al., 1995). Thus, yield stress was calculated as follows (Dzuy and Boger, 1983):

$$\sigma = 2M_0/\pi d^3 (h/d + 1/6)^{-1}$$

where:

$\sigma$  = yield stress in Pascals

$M_0$  = maximum torque in Newton meters

$d$  = diameter of the vane in meters (.01m)

$h$  = height of the inserted vane in meters (.025m)

The strain exhibited at the yield stress, termed the apparent yield strain, is calculated to be radians (distance) that the vane rotates up to the point where the maximum yield stress is reached. It was calculated as follows (Daubert et al., 1998, Breidinger and Steffe, 2001):

$$\gamma_0 = t \Omega / 2\pi$$

where:

$\gamma_0$  = apparent yield strain in radians

$t$  = time to reach yield stress in seconds

$\Omega$  = rotational speed of vane in revolutions/second

Although this is not a true yield strain, it is still useful because it is proportional to the yield strain and can be used to create a texture map of spreadability.

### ***Fatty Acid Profiling***

Fatty acid methyl ester esters were prepared with methods adapted from Bannon et al. (1982). Each sample was analyzed in triplicate. One drop of oil from melted butter (20-30 mg) was weighed into a screw capped tube, 1 mL 0.5M methanolic potassium hydroxide (Fisher Scientific, Fairlawn, NJ) was added. The tube was capped and placed into an 80°C. After the tube was cooled, 1 mL boron-trifluoride (Sigma-Aldrich, St. Louis, MO) was added. The capped tube was heated for 5 minutes in the 80°C water bath. After the tube was cooled slightly, 1 mL of deionized water and 1 mL of hexane were added (Optima grade, Fischer Scientific, Fairlawn, NJ). The tube was vortexed for 30 seconds and the contents were allowed to settle. The top hexane layer was removed and placed into a tube containing 1g sodium sulfate (Sigma-Aldrich, St. Louis, MO) to remove any small amount of water that might be present. The hexane phase containing the fatty acid methyl esters was then transferred to a vial for analysis.

A PerkinElmer Autosampler XL (PerkinElmer, Boston, MA) was used with a Restek, RT-2560 column (Restek, Bellefonte, PA) equipped with a flame ionization detector (detector temperature 220°C, injector temperature 220°C). The initial temperature was 100°C, which was held for 2 minutes then the temperature was increased at 3°C/min up to 250°C, which was held for 2 minutes. The total run time was 54 minutes. Helium at 40psi was used for the carrier gas. Hexane was used as a

blank. GLC-21A (Nu-Chek Prep Inc., Elysian, MN), Kel-Fim FAME-7 Standard (Matreya, Pleasant Gap, PA), and Restek #35078 (Restek, Bellefonte, PA) were used as standards.

### ***Turbidity***

Melted butter clarity or turbidity was evaluated across storage as this is an important functional characteristic for the restaurant industry. One stick (or 110 g bulk butter) was melted on low heat (setting “2” on hot plate) for 30 minutes. The melted butter was cooled at 21C for 15 minutes, and any floating foam/particles were spooned off the top. The oil was decanted and filtered twice through four-ply 100% cotton, fine mesh Pyrex® cheese cloth (Robinson Knife Company, Buffalo NY). The turbidity was measured at 50C on an Orbeco-Helligae 964-10A Digital Direct-Reading Turbidimeter (Orbeco Analytical Systems Inc., Farmingdale, NY).

### ***Differential Scanning Calorimetry (DSC)***

Differential scanning calorimetry (DSC) was performed on the butter to determine if any differences in melting characteristics existed. A PerkinElmer DSC7 with Pyris software with and Intracooler II refrigeration unit and dry box (Perkin Elmer Corp, Norwalk, CT) were used to perform the analysis. Two replicates of each butter sample were run. The DSC was calibrated with mercury, water, and indium. All equipment was turned on 1 hour before use. Nitrogen was flowing through the sample holder at 24psi and helium gas purge (250 psi) was used. The sample was weighed into an aluminum DSC pan. The sample and an empty pan were placed into the dry box and the program was started. The pans were heated from 20°C to 50°C at a rate of 200°C/min. They were held at 50°C for 1 minute, then cooled to -40°C at

5°C/min. The samples were then held at -40°C for 20 minutes. Lastly, they were heated from -40°C to 50°C at 5°C/min.

### ***Statistical Analysis***

Statistical analysis was conducted to determine the impact of package (bulk, stick), storage temperature, and time. Replications (production facility and day) were not significantly different from one another, so these data were averaged. The Proc Mixed command (SAS software, version 9.1, Cary, NC, U.S.A.) was used to evaluate main effects, packaging and time/temperature interactions, and secondary interactions between packaging and time/temperature variables. The SAS slice command was used to clarify effects in the event of significant secondary interactions. Fishers least significant difference (lsd) was used for means separation.

## **RESULTS & DISCUSSION**

### ***Proximate Analysis***

Proximate analysis of fat, moisture, and salt content was conducted at the time of manufacturing. Stick butter averaged  $80.6\% \pm 0.15\%$  fat and bulk butter was  $80.37\% \pm 0.04\%$ . Bulk butter contained slightly more moisture and salt ( $16.44 \pm 0.06\%$  and  $1.77 \pm 0.04\%$ , respectively) than butter quarters ( $16.31 \pm 0.13\%$  and  $1.63 \pm 0.06\%$ , respectively).

### ***Descriptive Sensory Analysis***

A decline in cooked/nutty flavor (a flavor which is prevalent in freshly churned butter) occurred over time ( $p < 0.05$ ) (Table 3.3). An interaction between packaging and time/temperature was also evident as flavor changes were different for different package types and storage temperatures. At refrigeration temperature, a

significant decrease ( $p < 0.05$ ) in cooked/nutty flavor occurred with storage time across all packaging types. This decrease was most pronounced and rapid in the wax-paper wrapped sticks compared to bulk butter. A perceived decrease in this fresh flavor was observed by six months refrigeration in both bulk and stick butter. A much slower reduction was observed in the frozen product, and the same trend in packaging interactions was noted (wax-wrapped sticks had the fastest decline and bulk butter had the slowest). Levels of cooked/nutty flavor detected after 12 months in the frozen product were not significantly different than levels observed at 3 months in the refrigerated product. Butters that were frozen and then refrigerated maintained cooked/nutty flavor longer than refrigerated storage alone.

Development of a refrigerator/stale flavor was observed in butters over storage time. Differences in the development of this storage flavor between package and time/temperature were also observed, similar to cooked/nutty flavor ( $p < 0.05$ , Table 3.4). Refrigerator/stale flavor developed more quickly in refrigerated butter compared to frozen butter. After 6 months, refrigerator/stale flavor was above sensory threshold in refrigerated wax-paper wrapped sticks and at 9 months it was present above sensory threshold in refrigerated bulk butter. Bulk butter developed refrigerator/stale flavor more slowly than stick butter. At frozen storage temperatures, refrigerator/stale flavor was detected at 12 months in wax-paper wrapped sticks and was not detected after 15 months in bulk butters. Samples that were frozen for 6 months and then refrigerated had slower refrigerator/stale flavor development than samples at refrigeration alone. Detectable levels of refrigerator/stale flavor were noted at 9 months (frozen 6 months, refrigerated 3

months), higher levels were observed at 12 months (frozen 6 months, refrigerated 6 months) of storage time. Levels seen in the two types of packaging during mixed frozen and refrigerated storage were consistent with that of trends in refrigerated and frozen storage.

Milkfat flavor intensity (another flavor prevalent in freshly churned butter) changed over time based on storage temperature ( $p < 0.05$ ) (Table 3.5). A decline in milkfat flavor was detected in both refrigerated and frozen samples, but the decline occurred more rapidly in refrigerated samples. The treatment that was frozen for 6 months and then refrigerated for 3 months had the same level of milkfat flavor as samples refrigerated for six months. Bulk butter maintained significantly more milkfat flavor than wax-paper wrapped sticks over the course of storage, having an average intensity over 12 months of  $2.75 \pm 0.02$  while stick butter had an average of  $2.55 \pm 0.02$  (LSD = 0.05).

Some changes in texture were observed by the trained panel. A decrease in the ease of spreadability was observed over time by the trained panel in both refrigerated and frozen butter ( $p < 0.05$ , Table 3.6). Several sensory attributes were not affected by temperature or time. Salty taste was consistent over time and storage conditions ( $p > 0.05$ ). Bulk butter had a higher salt intensity ( $10.08 \pm 0.15$ ) than butter sticks ( $9.10 \pm 0.06$ , LSD = 0.2,  $p < 0.05$ ), in agreement with its higher salt content. Color and firmness showed no significant interactions and did not change with storage time or storage conditions ( $p > 0.05$ , results not shown).

To our knowledge, our study is the first to use a trained panel to evaluate texture changes in butter over storage time and the first to use descriptive sensory

analysis to document butter flavor and butter flavor changes across storage. Our sensory results are consistent with previous studies that used qualitative sensory analysis techniques (grading). Emmons et al. (1986) found that butter frozen at -18C for 12 months had undergone less flavor changes (increases in oxidized, paper/cardboard, and other sensory defects determined by graders) than butter 454g prints stored at 5C for 14 weeks. Furthermore, all but one of the paper-based wrappers had more grading deductions than prints stored in polyethylene-based wrappers. Jebson et al. (1974) found butter stored in 25 kg blocks in boxes lined with parchment at -18C and 4C incurred storage flavors after 8 months. Butter stored at 4C exhibited a higher level of storage flavor and in one case, fishy flavor, than that of butter stored at -18C. Tomlinson and Dixon (1977) found that bulk butter stored in 25 kg blocks at -15C did not have any off-flavors after 34 weeks, consistent with the results observed in this study. Kristensen et al. (2000) found no perceivable sensory differences by trained panelists in color of a sweet cream spread stored at -18C and 5C for a 10 week period which is consistent with the first timepoint results of this study.

### ***Oxidative Stability Index (OSI)***

The OSI value for a butter was determined as the length of time prior to the onset of a rapid increase in the rate of oxidation induced by high temperatures and air sparged into the sample. This lag time was measured in hours. Higher values indicate greater oxidative stability, and a decrease in OSI with storage time is indicative of oxidation. Time/temperature and packaging interactions were observed ( $p < 0.05$ , Table 3.7). Refrigerated butters showed the largest decline in oxidative

stability over the 15 month time period. Frozen samples exhibited a constant OSI from 6 to 15 months. Butter that was frozen for 12 months and then placed into refrigerated storage for 3 months had an OSI equivalent to that of butter refrigerated for 6 months. Butters that were frozen for 6 months and refrigerated for 6 months showed a significantly lower OSI than butter refrigerated for 6 months alone. Unlike sensory changes observed in butter flavor, there was no clear trend as to whether bulk or quarters had a slower rate of oxidation.

OSI has not been widely applied to storage of butters and spreads. In a 10-week study done by Kristensen et al. (2000), no oxidative degradation was detected when a sweet cream spread was stored at -18C and 5C using a similar oxidative stability method. This is comparable to the 3-month results observed in this study. The sensory results of this study clearly showed that the development of refrigerator/stale flavor was more rapid in butter quarters than butter stored in bulk. This is a possible indication that lipid oxidation is not entirely responsible for the evolution of refrigerator/stale flavor since differences were not observed between bulk and stick butter by the OSI.

### ***Peroxide Value***

Peroxide value is a measure of the initial products of lipid oxidation. Differences in peroxide value between temperature treatments over storage time were significant ( $p < 0.05$ , Table 3.8). Additionally, secondary packaging interactions with temperature treatment and time were observed ( $p < 0.10$ , Table 3.9). A large increase in PV was seen over the course of the first 6 months in both refrigerated and frozen storage, although it was greater in the refrigerated butter. Six months later, the PV of

frozen samples increased significantly while the refrigerated butter did not show a significant increase. Butter that was frozen for 6 months and then refrigerated for 6 months was not significantly different from butter that was refrigerated for 6 months only, nor was it statistically different from the 12 month refrigerated butter.

Emmons et al. (1986) found that peroxide value was not elevated in the butters that exhibited storage grading defects suggesting a different cause for stale off-flavors in butter. Similar to this study, Jebson et al (1974) found an increase in peroxide value occurred in bulk samples stored at 4C after 4 and 8 months. In contrast to this study, they found butter stored at -18C yielded only a slight increase in peroxide value after 8 months. Okturk (2001) found that butter stored at 5C for 3 months had an almost three-fold increase in peroxide value over this time period. This result is consistent with the increase seen in this study between fresh butter and that stored 6 months at 5C. Downey et al. (1980) found that when peroxide value was greater than 2 meq O<sub>2</sub>/kg fat, off-flavors were detected in butter by trained graders. This is not consistent with the findings of this study. Bulk butter was stored for 12 months at -20C without any detection of off-flavors by the trained panel, despite the elevated peroxide value. Abdel-Mageed and Fadel (1995) isolated volatile components from butter stored for 7.5 months at -18C and found that carbonyls caused by peroxidation of fatty acids were at their highest levels after 4.5 months and decreased there-after. This is concurrent with the PV decreases in this study after 6 months of storage.

### ***Free Fatty Acid***

Free fatty acids are a product of hydrolytic rancidity. The free fatty acid value in the butters increased slightly over 12 months of storage. FFA value was higher in refrigerated samples than their frozen counterparts ( $p < 0.05$ , Table 3.10). The treatment that received combined frozen and refrigerated storage (6 months at each) had the same FFA value as that of the frozen samples. Although the difference in FFA value between bulk ( $0.21 \pm 0.002$ ) and stick butter ( $0.22 \pm 0.003$ ,  $LSD = 0.009$ ) was statistically significant ( $p < 0.05$ ), the actual difference was very small and is likely not of practical value or significance.

O'Connell et al. (1975) found that FFA levels did not increase over 56 weeks of storage at  $-18^{\circ}\text{C}$  of both butter prints and bulk butter (56 lb blocks). Similarly in this study, levels of FFA in butters were consistent through 6 months of storage before they exhibited a slight increase. Conversely, Okturk et al. (2001) found that levels of free fatty acids increased significantly over 90 days of storage at  $5^{\circ}\text{C}$ .

In this study, the decrease in OSI was the best predictor of the sensory perception of off-flavors for bulk stored butter. Butter that was stored as sticks did not have a lower OSI despite exhibiting off-flavors by sensory analysis at some timepoints. Despite the elevated levels of PV in some of the butters, off-flavors were not observed by the trained panel.

### ***Vane rheometry***

Differences in yield stress (Pa) were significant between temperature treatments and between packaging types ( $p < 0.05$ , Table 3.11). Significant changes occurred in butters between 0 and 6 months in both the refrigerated and frozen storage conditions. At 12 months, neither treatment was significantly different from

the 6 month value. When comparing packaging types, bulk butter had a significantly lower yield stress than butter in wax-wrapped sticks. Bulk butter had a yield stress of  $5787 \text{ Pa} \pm 114$  over time, while stick butter had an average yield stress of  $6506 \pm 104$  over 12 months (LSD = 246 Pa,  $p < 0.05$ ).

A spreadability map is shown in Figure 1. As refrigeration time was increased, the yield stress increased and yield strain decreased meaning that the refrigerated butter took less rotation to fracture but more force. Daubert et al. (1998) found this direction of change to be a decrease in the spreadability of the product in the direction of a more brittle product. This decrease in spreadability of refrigerated butters is in agreement with the sensory results from this study. Kawanari et al. (1981) found that butter stored for 12 weeks at 5C was harder than butter stored at -30C for the same time period. De Man and Wood (1958) found butter stored at -20 for 5 weeks did not have an increase in hardness as butter samples stored at 5C did. They also found that butter which was first stored at -20C and removed to the 5C condition became increasingly hard up to the same level of butter that had been stored at 5C for the entire time. De Man and Wood (1958) attribute the softer texture of the frozen samples to a delay in crystallization that occurs after the continuous butter making process. The setting of these crystals is interrupted by freezing and does not resume until the temperature is above freezing. Tverdokhleb and Auvakum (1978) ascribe temperature storage changes in temperature to supplementary crystallization of the glycerides. Kulkarni and Ramamurthy (1985) also explain temperature/storage effects on butter texture as attributable to the changes in the solidification of triglycerides during storage.

### ***Fatty Acid Profiling***

Comparison of the levels of saturated, monounsaturated, and polyunsaturated fatty acids in the samples were conducted upon receipt and on both refrigerated and frozen samples after 6 months. Butter produced in the summer months (August) was also collected fresh from the same facilities to determine if the fatty acid content was different with season. Statistical analysis revealed no significant differences between the fresh butter and 6 month refrigerated and frozen butter. Additionally, there were no significant differences between the butter produced in the summer and the butter collected in February and used for the storage study (results not shown).

Abdel-Mageed and Fadel (1995) found frozen storage (-18C) had an effect on the fatty acid composition of sweet cream butter after 6 months. They found decreased levels of linolenic acid after 6 months. This was not consistent with results seen in this study. The butter used by Abdel-Mageed and Fadel (1995) contained more linolenic acid initially than butter used in this study.

### ***Instrumental Color-Whole Butter***

The Hunter L\* a\* b\* was instrumentally measured to examine any color changes that occurred over storage time. Differences in the L\* value (where 100 = white and 0 = black) between bulk and stick butter were significant ( $p < 0.05$ ). Stick butter was significantly lighter in color ( $L = 78.5 \pm 0.13$ ) than bulk butter ( $L^* = 77.7 \pm 0.10$ ,  $LSD = 0.3$ ,  $p < 0.05$ ). Time and storage conditions also had a significant effect on L\* value (Table 3.12). Over time, storage tended to lighten the color of butter stored under both refrigerated and frozen storage conditions.

The green/red hue of the butter color was measured as the  $a^*$  value (+ values are red hues and – values are green hues). The  $a^*$  values had differences based on packaging, storage condition, and time ( $p < 0.05$ , Table 3.13). Bulk butter tended to be more greenish (more negative  $a^*$  value) than stick butter ( $p < 0.05$ ). While refrigerated and frozen storage were not significantly different from one another, the largest change in green color from the fresh samples was between 0 and 6 months of storage.

The  $b^*$  value indicates the degree of yellow or blue hue (+ values are yellow hues and – values are blue hues). While differences were present in the samples ( $p < 0.05$ ), the trends were not clear. Interactions between time and temperature and packaging are shown in Table 3.14. At 6 months, the color became less yellow in both samples, but at 12 months it was higher in both samples. The frozen samples were consistently more yellow than the refrigerated samples after 12 months of storage. There is no clear trend as to which packaging type had a higher  $b^*$  value. Despite these instrumental differences, it is important to emphasize that there were no sensory perceived visual differences between any of the samples at any of time-points.

A study of sweet cream dairy spreads by Kristensen et al. (2000) found that very high storage temperature (20C) yielded darker butter (lower  $L^*$  values) after 10 weeks of storage, and few differences were observed between samples kept at -18C and 5C for the same time period. They also found that samples stored at the highest temperature were more green (lower  $a^*$  values) and had higher  $b^*$  values (were more yellow). This trend was less pronounced in butter stored at -18C and 5C. Tomlinson and Dixon (1977) found that butter stored in polyethylene wrapping was found to

have less surface darkening after 34 weeks at -15C than butter wrapped in parchment. This result was mirrored in this study by the bulk butter, stored in polyethylene which exhibited less darkening than the stick butter wrapped in wax-paper.

### ***Instrumental Color-Clarified Oil***

Oil color was also evaluated using the Hunter L\* a\* b\* scale instrumentally. The degree of lightness of the oil, L\*, decreased significantly after 12 months of storage. The L\* value exhibited interactions based on storage conditions, packaging, and time ( $p < 0.05$ , Table 3.15). Decreasing levels for L\* were observed in all samples across all packaging types, temperatures, and time.

The green hue of the oil (a\* value) had significant differences based on storage temperature and time (Table 3.16). Slight differences were observed. Most notably the reduction in green color of the refrigerated samples after 12 months was observed and a similar trend was noted in the samples that were frozen for 6 months and then refrigerated for 6 months.

The yellowness of the oils (b\*) changed with storage conditions and time (Table 17). A slight dip in yellowness at 6 months occurred at both storage conditions. This is consistent with what was seen in the solid butter color. Frozen butter had a higher level of yellow color than refrigerated butter.

While studies have examined butter oil and ghee stability over time and storage conditions (Kehagias and Rademema, 1973), they have not examined color changes to our knowledge. Additionally, no studies have been conducted that examine changes in butter oil produced from stored butter.

### ***Turbidity***

Particulates suspended in the oil were measured by means of oil turbidity. Turbidity was measured in Nephelometric Turbidity Units (NTU's), which are a measure of light scattering from particles present in the sample. Increasing ranges of turbidity were seen with increasing time (Figure 3.2). At the 12 months sampling point, bulk samples had a higher level of turbidity than butter stored as sticks across all temperature storage conditions. At initial and 6 month evaluations, all samples displayed approximately the same turbidity range. Turbidity of butter oil has not previously been evaluated throughout the course of butter storage. These results indicate that butter oil clarity is negatively influenced by storage time.

#### ***Differential Scanning Calorimetry***

Melting characteristics were examined through the use of the DSC. No differences were seen between bulk and stick samples though 9 months (results not shown).

### **CONCLUSIONS**

There were no significant differences between bulk and stick butter in OSI and PV. While FFA values showed a significant difference between the different temperature conditions, the difference was so small that there was likely no practical value. There was a significant difference between bulk and stick butter in development of refrigerator/stale flavor and the reduction of cooked/nutty flavor. Observed changes in lipid oxidation with storage were not in entire agreement with sensory perceived changes. Since development of stale flavors was not entirely consistent with instrumental measurements of lipid oxidation, this is an indication that off-flavor development in stored butter is coming at least in part from a source other

than lipid oxidation. This is not the first study to come to the conclusion that off-flavors in stored butter were not solely a result of oxidation (Emmons et al., 1986; MacBean, 1974; Pont, 1961). These studies theorized that compounds in product packaging may also be contributing to these storage-associated off-flavors.

Butter that was frozen and then refrigerated developed off-flavors faster after freezing than butter that was not frozen prior to refrigerated storage. Mixed frozen and refrigerated storage, while allowing a longer shelf-life than refrigeration alone, was not entirely additive of frozen and refrigerated storage effects. Refrigerated butter quarters showed the fastest decline in quality. To this end, for optimum quality, butter quarters should be refrigerated for less than 6 months. When frozen at -20C, sticks can be stored for up to 12 months. It is still advantageous for manufacturers to continue to store butter in large blocks. While it may not completely stop lipid oxidation, it will maintain the flavors of freshly produced butter (milkfat, cooked/nutty flavors) longer. For bulk butter in refrigerated conditions, flavor quality is maintained for at least 9 months. In frozen storage, bulk butter can be stored in excess of 15 months without flavor detriment. Our estimate of shelf-life of bulk butter is conservative since 4 kg blocks were used for the study for convenience and cost, and butter is often stored in 25 kg block form.

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**Table 3.1 Sample conditions.**

Facility/Day/Bulk or Stick	Temperature Treatment	
Facility 1, Day 1, Bulk	-20C	5C
Facility 1, Day 2, Bulk	-20C	5C
Facility 2, Day 1, Bulk	-20C	5C
Facility 2, Day 2, Bulk	-20C	5C
Facility 1, Day 1, Stick (wax-paper wrapped)	-20C	5C
Facility 1, Day 2, Stick (wax-paper wrapped)	-20C	5C
Facility 2, Day 1, Stick (wax-paper wrapped)	-20C	5C
Facility 2, Day 2, Stick (wax-paper wrapped)	-20C	5C

**Table 3.2 Sensory language used for the descriptive sensory analysis of butter**

<b>Term</b>	<b>Definition</b>	<b>Reference</b>
Diacetyl/Cultured <sup>a</sup>	Sweet aromatic characteristic of cultured dairy products, of which diacetyl is a primary source	Diacetyl, 20ppm
Milkfat/Lactone <sup>a</sup>	Aromatic characteristic of milkfat, lactones, and coconut	Heavy cream
Cooked/Nutty	Aromatic associated with cooked milk and canned corn	1% fat milk heated in a microwave for 8 min
Refrigerator/Stale	Stale aromatic characteristic of refrigerator with old food left in it	Butter quarters (sticks) stored in a refrigerator for 18 mo
Painty	Aromatics associated with wall paint and oxidized fats	Linseed oil
Salty <sup>b</sup>	Taste elicited by NaCl	Sodium chloride solutions; 0.5% (5), 0.7% (8), 0.9% (11.5)
Yellow <sup>c</sup> Spreadability	Intensity of yellow color Force necessary to spread three strokes, backward, forward, backward on unsalted saltine cracker (1/2" tip of the knife); 0=not spreadable, 10=very spreadable	Yellow color scale 3A Land O Lakes spreadable butter (9) Crisco vegetable oil sticks (8.5) Challenge butter (5); tempered for 1.5 hours at 19C
Firmness	Force required to deform sample against roof of mouth; 0=not at all firm, 10=very firm	Crisco vegetable oil sticks (5); Land O Lakes spreadable butter (1); tempered for 1.5 hours at 19C

Language adapted from Krause et al. (2006)

<sup>a</sup> Reference taken from Drake et al., 2001

<sup>b</sup> Reference taken from Meilgaard and others 1999

<sup>c</sup> Reference taken from Kornerup and Wanscher, 1978

**Table 3.3 Trained panel perception of cooked/nutty flavor intensity separated by packaging type, time, and temperature treatment.**

Temperature	Packaging	Time					
		0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	Bulk	3.7	3.2	2.3	2.4	1.9	1.7
	Sticks	3.9	2.7	2.0	2.1	1.4	1.6
Frozen	Bulk	.	.	3.1	.	3.3	2.8
	Sticks	.	.	3.0	.	2.6	2.7
Frozen 6 mo/ Refrigerated 3 mo	Bulk	.	.	.	2.5	.	.
	Sticks	.	.	.	2.3	.	.
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	.	.	2.0	.
	Sticks	.	.	.	.	1.4	.
Frozen 12 mo/ Refrigerated 3mo	Bulk	.	.	.	.	.	2.5
	Sticks	.	.	.	.	.	2.0

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Cooked/nutty flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Table 3.4 Mean refrigerated/stale flavor intensity evaluated by a trained panel separated by packaging type, time, and temperature treatment.**

Temperature	Packaging	Time					
		0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	Bulk	ND	ND	ND	0.60	1.0	1.8
	Sticks	ND	ND	0.98	1.1	1.8	2.2
Frozen	Bulk	.	.	ND	.	ND	ND
	Sticks	.	.	ND	.	0.50	0.50
Frozen 6 mo/Refrigerated 3 mo	Bulk	.	.	.	0.50	.	.
	Sticks	.	.	.	0.60	.	.
Frozen 6 mo/Refrigerated 6 mo	Bulk	.	.	.	.	0.60	.
	Sticks	.	.	.	.	1.6	.
Frozen 12 mo/Refrigerated 3 mo	Bulk	.	.	.	.	.	0.60
	Sticks	.	.	.	.	.	1.2

LSD – least significant difference.

ND – not detected

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Refrigerator/stale flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Table 3.5 Trained panel perception of milkfat flavor intensity separated by temperature treatment and time.**

LSD=0.12 Temperature	Time					
	0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	3.1	3.0	2.6	2.5	2.5	2.0
Frozen	.	.	3.0	.	3.0	2.7
Frozen 6 mo/Refrigerated 3 mo	.	.	.	2.6	.	.
Frozen 6 mo/Refrigerated 6mo	.	.	.	.	2.5	.
Frozen 12 mo/Refrigerated 3mo						2.5

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Milkfat flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Table 3.6 Mean spreadability scores evaluated by a trained panel separated by packaging type, time, and temperature treatment.**

LSD=0.22 Temperature	Time					
	0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	6.0	6.2	5.9	5.8	5.7	5.2
Frozen	.	.	5.8	.	5.9	5.3
Frozen 6 mo/Refrigerated 3 mo	.	.	.	5.9	.	.
Frozen 6 mo/Refrigerated 6mo	.	.	.	.	5.6	.
Frozen 12 mo/Refrigerated 3mo	.	.	.	.	.	5.8

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Spreadability was scored on a product-specific scale where 0 = not at all spreadable and 10 = extremely spreadable

**Table 3.7 OSI (hours) over 15 months separated by time, temperature treatment and packaging.**

LSD=0.86 Temperature	Packaging	Time					
		0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	Bulk	10.9	11	9.1	5.5	6.3	5.7
	Sticks	12.3	11	9.7	6.5	6.5	5.2
Frozen	Bulk	.	.	8.5	.	8.3	8.6
	Sticks	.	.	9.3	.	8.8	8.1
Frozen 6 mo/Refrigerated 3 mo	Bulk	.	.	.	7.2	.	.
	Sticks	.	.	.	10.9	.	.
Frozen 6 mo/Refrigerated 6mo	Bulk	.	.	.	.	7.5	.
	Sticks	.	.	.	.	7.8	.
Frozen 12 mo/Refrigerated 3mo	Bulk	.	.	.	.	.	10.6
	Sticks	.	.	.	.	.	8.7

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Table 3.8 Peroxide value measured by AOCS method 965.33 separated by temperature treatment and time. Results are given in meq peroxide/kg oil.**

LSD=0.33 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	0.18	2.9	2.5
Frozen	.	1.6	2.4
Frozen 6 mo/Refrigerated 6 mo	.	.	2.6

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Table 3.9 Peroxide value measured by AOCS method 965.33 separated by temperature treatment, package, and time. Results are given in meq O<sub>2</sub>/kg oil.**

Temperature	Packaging	Time		
		0 mo	6mo	12mo
Refrigerated	Bulk	0.18	2.8	2.3
	Sticks	0.18	3.1	2.8
Frozen	Bulk	.	1.2	2.2
	Sticks	.	1.9	2.6
Frozen 6 mo/Refrigerated 6mo	Bulk	.	.	3.0
	Sticks	.	.	2.2

LSD – least significant difference.

Means that differ by more than the LSD are different (p<0.10).

**Table 3.10 FFA measured as mg NaOH/g butter oil separated by temperature treatment and time.**

LSD=0.01 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	0.21	0.21	0.25
Frozen	.	0.20	0.22
Frozen 6 mo/Refrigerated 6 mo	.	.	0.22

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Table 3.11 Yield stress (Pa) measured using the vane method separated by temperature treatment and time.**

LSD=557 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	5639	6640	6917
Frozen	.	6322	5759
Frozen 6 mo/Refrigerated 6mo	.	.	6220

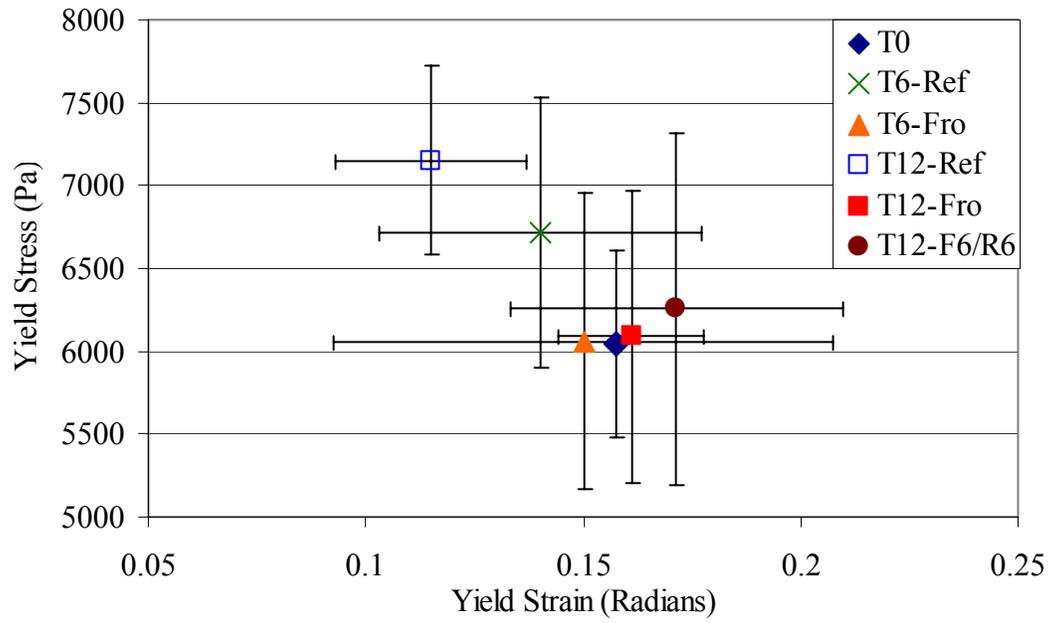
LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Yield stress was measured at 19°C

Vane had a diameter of 1.0 cm and a height of 2.5cm

Rotational speed of vane was 0.02 rpm.



**Figure 3.1. Spreadability map of Yield Stress (Pa) vs. Yield Strain (Radians)**

**Table 3.12 L\* values for solid butter color, separated based on storage conditions and time.**

LSD=0.42 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	77.80	76.70	79.30
Frozen	.	76.60	79.70
Frozen 6 mo/Refrigerated 6mo	.	.	78.70

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

L\* value on Hunter Scale, where 100=white and 0=black

**Table 3.13. a\* values for solid butter color, separated based on storage conditions and time.**

LSD=0.11		Time		
Temperature	Packaging	0 mo	6mo	12mo
Refrigerated	Bulk	-3.24	-3.6	-3.57
	Sticks	-3.02	-3.5	-3.46
Frozen	Bulk	.	-3.5	-3.69
	Sticks	.	-3.46	-3.41
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	-3.43
	Sticks	.	.	-3.52

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

a\* value on Hunter Scale, where + values are red hues and – values are green hues

**Table 3.14 b\* values for solid butter color, separated based on storage conditions, packaging, and time.**

LSD=0.31		Time		
Temperature	Packaging	0 mo	6mo	12mo
Refrigerated	Bulk	13.57	12.3	12.53
	Sticks	13.28	12.38	13.71
Frozen	Bulk	.	12.11	14.26
	Sticks	.	12.17	13.97
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	12.16
	Sticks	.	.	12.29

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

b\* value on Hunter Scale, where + values are yellow hues and – values are blue hues

**Table 3.15 L\* values for oil color, separated based on storage conditions, packaging, and time.**

LSD=0.31 Temperature	Packaging	Time		
		0 mo	6mo	12mo
Refrigerated	Bulk	33.56	32.96	28.54
	Sticks	33.59	33.72	28.48
Frozen	Bulk	.	33.59	28.39
	Sticks	.	33.66	28.45
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	28.39
	Sticks	.	.	28.4

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

L\* value on Hunter Scale, where 100=white and 0=black

**Table 3.16 a\* values for oil color, separated based on storage conditions and time.**

LSD=0.04 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	-1.03	-1.05	-0.91
Frozen	.	-1.07	-1.01
Frozen 6 mo/ Refrigerated 6mo	.	.	-0.90

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

a\* value on Hunter Scale, where + values are red hues and – values are green hues

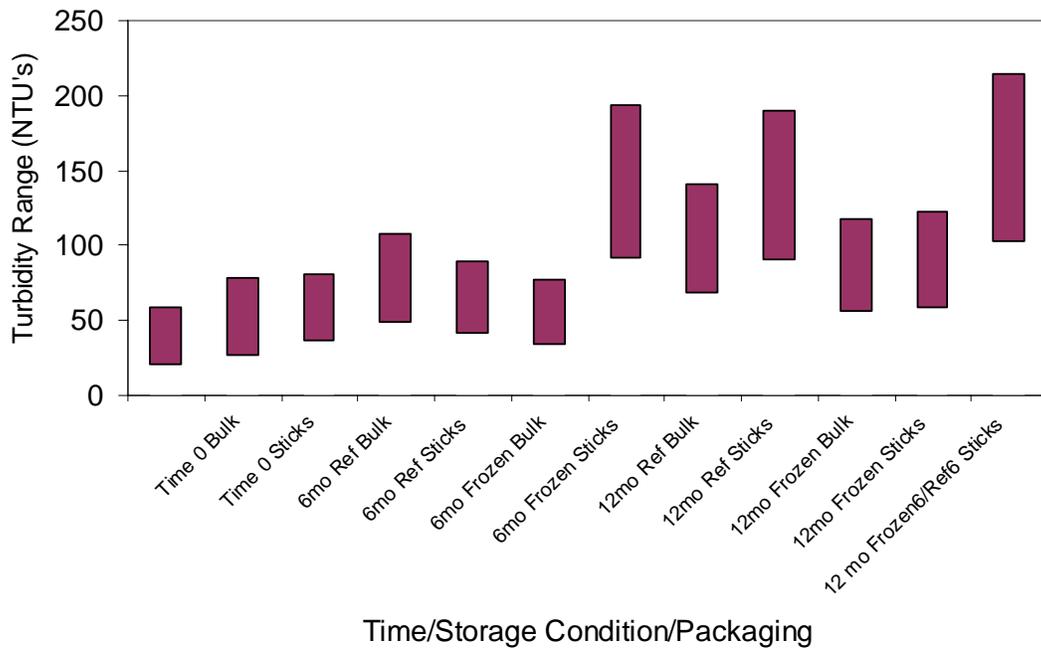
**Table 3.17 b\* values for oil color, separated based on storage conditions and time.**

LSD=0.17 Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	3.25	2.84	3.05
Frozen	.	2.80	3.56
Frozen 6 mo/ Refrigerated 6mo	.	.	3.18

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

b\* value on Hunter Scale, where + values are yellow hues and – values are blue hues



**Figure 3.2 Turbidity Range vs. Time, Storage Condition, and Packaging.**

**CHAPTER 4**  
**CONCLUSIONS**

## CONCLUSIONS

Butter is a very important commodity in the American dairy industry. A better understanding of it from both a consumer and an analytical standpoint is important for future marketing and expanded distribution of the product. The appeal of butter appears to be widening with the increasing demand for “natural” products and the focus on moderation of high-fat foods in a balanced diet as opposed to eliminating them completely. Though butter has been largely studied, few studies have utilized the powerful tool of descriptive sensory analysis. The two studies conducted in this thesis demonstrate how sensory techniques can be used in diverse ways to study very different objectives. This research will serve to compliment previously published studies on butter and will add to current knowledge of consumer acceptance of butter and proper storage of butter to maximize quality.

Consumers value the flavor and the natural image of butter. This was displayed through focus groups and consumer test questionnaires. While a large portion of consumers like equally or prefer butter to spreads, many also use margarine/spreads for reasons such as: cost, spreadability, and health. Factors that influence purchase decision are different for different sections of the consumer population. Acceptance of butter and spreads differs across consumer segments with specific segments preferring specific butter and spread flavor profiles. Through the use of descriptive sensory analysis differentiation of various types of butter was achieved and separation of the various market groups was possible.

On the production side of butter, distributors and manufacturers often store surplus butter for extended periods of time until there is a demand for it. During

refrigerated and/or frozen storage, degradation of flavor occurs. This research will help companies to develop specification sheets for butter suppliers and for butter suppliers to design storage regimes.

Storage at -20C drastically slowed the evolution of off-flavors. Mixed frozen and refrigerated storage, while allowing a longer shelf-life than refrigeration alone, was not entirely additive of the effects of frozen and refrigerated storage. It is most advantageous for manufacturers who are storing product for longer than 6 months to use frozen storage. Another factor that extended storage time was the packaging/size of the stored butter; it is beneficial for manufacturers to store butter in large blocks. While it may not completely stop lipid oxidation, it will maintain the flavors of freshly produced butter (milkfat, cooked/nutty flavors) longer.

Development of refrigerator/stale flavor occurred more quickly in stick butter than in bulk butter, while observed changes in lipid oxidation with storage were not in entire agreement with these sensory perceived changes. Since development of stale flavors was not entirely consistent with instrumental measurements of lipid oxidation, this is an indication that off-flavor development in stored butter is coming at least in part from a source other than lipid oxidation. This is not the first study to come to the conclusion that off-flavors in stored butter were not solely a result of oxidation. Previous studies (Emmons et al., 1986; MacBean, 1974; Pont, 1961), have theorized that compounds in product packaging may also be contributing to these storage-associated off-flavors.

Future research may explore off-flavor carry-through in ingredient applications with stored butter and the nature of the consumer response to storage

flavor in butter. This aspect was touched on in the descriptive study. When compared to a range of butters, including butter made fresh within 48 hours, a 12 month old butter with stale flavors was rated the third highest in overall acceptability by consumers. This is a possible indication that consumers have come to accept refrigerator/stale flavors in the product they purchase and store in their homes. Further research would be necessary to completely understand attitudes towards these flavors.

## **APPENDICES**

## Appendix 1 Butter/spreads descriptive ballot

NAME \_\_\_\_\_ Date \_\_\_\_\_

### Butter Ballot-Flavor

Please taste the warm up (WU) sample and note the flavor profile before starting your analysis and please taste the samples in the order given below. Be sure to use your salty references and take a break as needed. Please evaluate color after you have tasted the sample.

Tasting order \_\_\_\_\_

	WU	WU2	024	501	892	233	906	531
<b>Diacetyl/cultured</b>	0	0						
<b>Cooked/nutty</b>	2.5-3.0	2.5						
<b>Milkfat</b>	3	2.5						
<b>Salty</b>	9-10	9.5						
<b>Refrigerator/stale</b>	--	--						
<b>Grassy/feed</b>	--	1.5						
<b>Sour</b>	--	--						
<b>Other</b>	--							
<b>COLOR</b>								

## Appendix 2 Focus group questionnaire

### FOCUS GROUP QUESTIONNAIRE

Please check the appropriate answer for the following demographic information:

1. Sex  male  female
2. Age group  
 18 years or younger  
 19 – 24 years  
 25 – 30 years  
 31 – 35 years  
 36 – 45 years  
 46 – 55 years  
 55 years or older

*Please answer the following questions. There are no right or wrong answers. We want to know about you and what you think. Please ask if you have any questions!*

3. Do you shop for your household, even if it is you alone?  yes  no
4. How often do you use butter?
  - a.  I never use butter
  - b.  Less than once a month
  - c.  2-4 times a month
  - d.  More than once a week
  - e.  Every day
5. How often do you use non-butter spreads?
  - a.  I never use spreads
  - b.  Less than once a month
  - c.  2-4 times a month
  - d.  More than once a week
  - e.  Every day
6. What do you use butter for? (check all that apply)  
 Cooking/baking  
 On vegetables  
 On bread  
 Other (please specify) \_\_\_\_\_
7. What do you use non-butter spreads for? (check all that apply)  
 Cooking/baking  
 On vegetables  
 On bread  
 Other (please specify) \_\_\_\_\_

### Appendix 3 Questions for focus group

#### Questions for Butter Focus Group

- How often do you consume butter?
- On what type of occasions? (lunch, snacks, etc)
- Why do you choose to use butter instead of other types of fat?
- Why do you not use butter more frequently?
- Compared to other fats, what do you particularly like about butter?
- Is butter good for you?
- What do you not like about butter?
- What brands do you purchase? Why?
- Do you buy unsalted or salted butter? Why?
  
- Now we are going to look at some samples (numerically labeled)...

  - Look at them first  
What do you think of their appearance? Comments?
  
  - Taste them  
Texture?  
Flavor?  
What do you think they are?

  
- Here are 4 more samples (this time w/ labels)

  - Look at them first
  - What do you think of their appearance? Comments?
  
  - Taste them  
Texture?  
Flavor?

## Appendix 4 Consumer butter and spreads questionnaire

### CONSUMER QUESTIONNAIRE

Please check the appropriate answer for the following demographic information:

1. Sex  male  female
2. Age group
  - 18 years or younger
  - 19 – 24 years
  - 25 – 30 years
  - 31 – 35 years
  - 36 – 45 years
  - 46 – 55 years
  - 55 years or older

Please answer the following questions. There are no right or wrong answers. We want to know about you and what you think. Please ask if you have any questions!

3. Do you shop for your household, even if it is you alone?  yes  no
4. How often do you use **butter**?
  - a.  I never use butter
  - b.  Less than once a month (occasionally)
  - c.  2-4 times a month
  - d.  More than once a week
  - e.  Every day
5. How often do you use **non-butter spreads (margarine)**?
  - a.  I never use spreads
  - b.  Less than once a month (occasionally)
  - c.  2-4 times a month
  - d.  More than once a week
  - e.  Every day
6. Please check the statement that best describes you:
  - a.  I only use butter
  - b.  I use butter and margarine
  - c.  I only use margarine
7. What do you use **butter** for? (check all that apply)
  - a.  Cooking/sautéing
  - b.  Baking
  - c.  On vegetables
  - d.  On bread
  - e.  On popcorn
  - f.  Special occasions/holidays
  - g.  Other (please specify) \_\_\_\_\_
  - h.  Do not use butter
8. What do you use **non-butter spreads (margarine)** for? (check all that apply)
  - a.  Cooking/sautéing
  - b.  Baking
  - c.  On vegetables
  - d.  On bread
  - e.  On popcorn
  - f.  Special occasions/holidays
  - g.  Other (please specify) \_\_\_\_\_
  - h.  Do not use non-butter spreads

## Appendix 4 Consumer butter and spreads questionnaire (continued)

9. What factors influence your purchase of butters and non-butter spreads? (check all that apply)

- |                                          |                                                       |
|------------------------------------------|-------------------------------------------------------|
| <input type="checkbox"/> Price           | <input type="checkbox"/> Health                       |
| <input type="checkbox"/> Salted/unsalted | <input type="checkbox"/> Spreadability                |
| <input type="checkbox"/> Availability    | <input type="checkbox"/> Flavor                       |
| <input type="checkbox"/> Brand           | <input type="checkbox"/> Package                      |
| <input type="checkbox"/> Organic         | <input type="checkbox"/> Other (please specify) _____ |

10. Please indicate how you feel about the following statement “**Butter is healthier than margarine.**”

- agree strongly
- agree
- neither agree nor disagree
- disagree
- disagree strongly

11. Please indicate how you feel about the following statement “**Butter is a natural product.**”

- agree strongly
- agree
- neither agree nor disagree
- disagree
- disagree strongly

12. Please indicate how you feel about the following statement “**Margarine is a natural product.**”

- agree strongly
- agree
- neither agree nor disagree
- disagree
- disagree strongly

## Appendix 5 Consumer butter and spreads ballot

No. \_\_\_\_\_

Please spread the sample that is indicated below onto the bread provided or use a spoon to taste the product. After you have tasted the product, please circle your response for the questions below. PLEASE ANSWER ALL of the questions!

Sample \_\_\_\_\_

<b>Overall Acceptance:</b>								
1	2	3	4	5	6	7	8	9
Dislike		Neither Like			Like			
Extremely		nor dislike			Extremely			

Please describe what you **LIKE** about the sample.

---



---



---



---

Please describe what you **DISLIKE** about the sample.

---



---



---



---

<p><b>Overall Appearance Liking:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Dislike            Neither Like            Like</p> <p>Extremely            nor dislike            Extremely</p>	<p><b>Color Intensity:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Low                    Moderate                    High</p>
<p><b>Salty Taste Intensity:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Low                    Moderate                    High</p>	<p><b>Color Liking:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Dislike            Neither Like            Like</p> <p>Extremely            nor dislike            Extremely</p>
<p><b>Freshness Intensity:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Low                    Moderate                    High</p>	<p><b>Salty Taste Liking:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Dislike            Neither Like            Like</p> <p>Extremely            nor dislike            Extremely</p>
<p><b>Overall Flavor Liking:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Dislike            Neither Like            Like</p> <p>Extremely            nor dislike            Extremely</p>	<p><b>Overall Texture Liking:</b></p> <p>1 2 3 4 5 6 7 8 9</p> <p>Dislike            Neither Like            Like</p> <p>Extremely            nor dislike            Extremely</p>

## Appendix 6 Butter descriptive flavor ballot

NAME \_\_\_\_\_ Date 5-1-06 (1)

### Butter Ballot-Flavor

Please taste the warm up (WU) sample and note the flavor profile before starting your analysis and please taste the samples in the order given below. Be sure to use your salty references and take a break as needed. Please evaluate color after you have tasted the sample.

PLEASE TAKE YOUR TIME--REST BETWEEN SAMPLES!!

Tasting order \_\_\_\_\_

	WU	WU2	531	534	107	283	722	300	628	023	952
<b>Diacetyl cultured</b>	--	--									
<b>Cooked/ nutty</b>	3-3.5	2.5									
<b>Milkfat</b>	3-3.5	2.5									
<b>Salty</b>	9	9									
<b>Refrigerator/stale</b>	--	1.5									
<b>Other</b>	--	--									
<b>COLOR</b>		1.8-2									

## Appendix 7 Descriptive panel texture ballot-spreadability

### Butter Ballot-Spreadability

NAME \_\_\_\_\_ Date 5-9-06 \_\_\_\_\_(1)

Please spread the reference samples and note the values before starting your analysis. Please evaluate the samples in the order given below. Be sure to take a break as needed

ORDER

	Ref. 1	Ref 2	Ref 3	228	796	019	511	300	881	444	581	942
Spreadability	9	8.5	5									

**Spreadability**-force it takes to spread three strokes, backward, forward, backward on cracker (1/2" tip of the knife);

0=not spreadable, 10=very spreadable

Reference 1=9

Reference 2=8.5

Reference 3=5

## Appendix 8 Descriptive panel texture ballot-firmness

### Butter Ballot-Firmness

NAME \_\_\_\_\_ Date \_\_\_\_\_ (1)

Please evaluate firmness for the reference samples and note the values before starting your analysis. Please evaluate the samples in the order given below. Be sure to take a break as needed.

ORDER

	Ref. 1	Ref. 2	228	796	019	511	300	881	444	581	942
<b>Firmness</b>	1 Whipped Butter	5 Crisco									

**Firmness**-place butter in mouth, press to roof of mouth, evaluate force required to deform sample against roof of mouth; 0=not at all firm, 10=very firm  
 Reference 1=1.0  
 Reference 2=5.0

## Appendix 9 Fatty acid profiles of stored butter and summer butter

<b>Storage Conditions</b>	<b>C4:0</b>	<b>C6:0</b>	<b>C8:0</b>	<b>C10:0</b>	<b>C12:0</b>	<b>C14:0</b>	<b>C14:1, trans</b>	<b>C14:1, cis</b>	<b>C15:0</b>	<b>C15:1, cis</b>	<b>C16:0</b>	<b>C16:1, trans</b>	<b>C16:1, cis</b>
Time 0	0.865	1.140	0.976	2.473	2.676	9.354	0.386	0.727	0.970	0.073	28.328	0.247	1.748
6mo-refrigerated	0.652	1.166	0.796	1.951	2.283	9.258	0.514	0.698	1.002	0.158	27.990	0.571	1.550
6mo-frozen	0.704	1.456	0.772	1.932	2.365	8.700	0.490	0.613	0.926	0.123	28.085	0.538	1.558
Summer-Facility 1	0.769	1.046	0.811	1.990	2.361	8.631	0.833	1.405	0.956	0.000	27.210	0.611	1.516
Summer-Facility 2	0.613	0.891	0.738	1.751	2.054	7.855	0.376	0.491	0.861	0.000	27.689	0.576	1.469

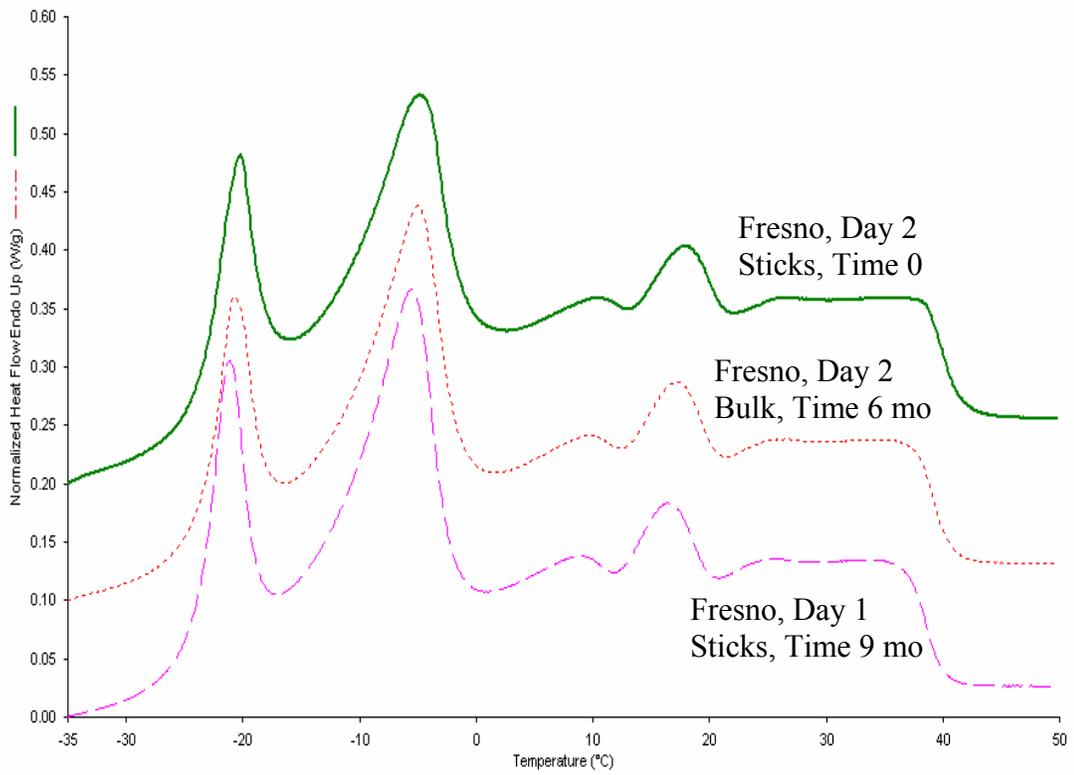
  

<b>Storage Conditions</b>	<b>C17:0</b>	<b>C17:1, cis</b>	<b>C18:0</b>	<b>C18:1 trans</b>	<b>C18:1 cis</b>	<b>C18:2 trans</b>	<b>C18:2 cis</b>	<b>C20:0</b>	<b>C20:1, cis</b>	<b>C21:0</b>	<b>C18:3, cis</b>	<b>C22:0</b>	<b>C24:0</b>
Time 0	0.617	0.044	15.684	2.983	25.583	0.616	3.241	0.095	0.010	0.486	0.392	0.000	0.192
6mo-refrigerated	0.662	0.185	14.894	3.129	26.968	0.920	3.288	0.196	0.109	0.623	0.381	0.041	0.026
6mo-frozen	0.664	0.187	14.885	3.151	27.292	0.860	3.295	0.199	0.109	0.634	0.400	0.040	0.020
Summer-Facility 1	0.665	0.148	15.182	3.061	27.002	0.958	3.315	0.230	0.076	0.609	0.422	0.111	0.071
Summer-Facility 2	0.691	0.209	17.365	2.959	27.709	0.760	3.407	0.232	0.105	0.539	0.400	0.121	0.140

<b>Storage Conditions</b>	<b>Saturated</b>	<b>Monounsaturated</b>	<b>Polyunsaturated</b>
Time 0	61.852	34.030	4.243
6mo-refrigerated	62.327	33.085	4.557
6mo-frozen	61.181	34.204	4.576
Summer-Facility 1	60.972	34.454	4.445
Summer-Facility 2	61.624	33.578	4.609

## Appendix 10 DSC profile of butter at 0, 6, and 9 months of storage



## Appendix 11 Sample conditions for storage study including foil-wrapped sticks

Facility/Day/Bulk or Stick	Temperature Treatment	
Facility 1, Day 1, Bulk	-20C	5C
Facility 1, Day 2, Bulk	-20C	5C
Facility 2, Day 1, Bulk	-20C	5C
Facility 2, Day 2, Bulk	-20C	5C
Facility 1, Day 1, Stick (wax-paper wrapped)	-20C	5C
Facility 1, Day 2, Stick (wax-paper wrapped)	-20C	5C
Facility 2, Day 1, Stick (wax-paper wrapped)	-20C	5C
Facility 2, Day 2, Stick (wax-paper wrapped)	-20C	5C
Facility 2, Day 2, Stick (foil-wrapped)	-20C	5C

**Appendix 12 Trained panel perception of cooked/nutty flavor intensity separated by packaging type, time, and temperature treatment including foil-wrapped treatment**

LSD=0.20		Time					
		0 mo	3 mo	6mo	9 mo	12mo	15mo
Temperature	Packaging						
Refrigerated	Bulk	3.7	3.2	2.3	2.4	1.9	1.7
	Wax-wrapped Sticks	3.9	2.7	2	2.1	1.5	1.6
	Foil-wrapped Sticks	3.4	2.8	2.2	2.4	1.7	1.8
Frozen	Bulk	.	.	3.1	.	3.3	2.8
	Wax-wrapped Sticks	.	.	3	.	2.6	2.7
	Foil-wrapped Sticks	.	.	3.2	.	3.1	3.0
Frozen 6 mo/ Refrigerated 3 mo	Bulk	.	.	.	2.5	.	.
	Wax-wrapped Sticks	.	.	.	2.3	.	.
	Foil-wrapped Sticks	.	.	.	2.6	.	.
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	.	.	2.0	.
	Wax-wrapped Sticks	.	.	.	.	1.4	.
	Foil-wrapped Sticks	.	.	.	.	1.9	.
Frozen 12 mo/ Refrigerated 3mo	Bulk	.	.	.	.	.	2.5
	Wax-wrapped Sticks	.	.	.	.	.	2.0
	Foil-wrapped Sticks	.	.	.	.	.	2.0

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Cooked/nutty flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Appendix 13 Mean refrigerated/stale flavor intensity evaluated by a trained panel separated by packaging type, time, and temperature treatment.**

LSD=0.25		Time					
Temperature	Packaging	0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	Bulk	ND	ND	ND	0.60	1.0	1.8
	Wax-wrapped Sticks	ND	ND	1.0	1.1	1.8	2.2
	Foil-wrapped Sticks	ND	ND	ND	0.60	1.6	1.7
Frozen	Bulk	.	.	ND	.	ND	ND
	Wax-wrapped Sticks	.	.	ND	.	0.50	0.50
	Foil-wrapped Sticks	.	.	ND	.	ND	ND
Frozen 6 mo/ Refrigerated 3 mo	Bulk	.	.	.	0.50	.	.
	Wax-wrapped Sticks	.	.	.	0.60	.	.
	Foil-wrapped Sticks	.	.	.	ND	.	.
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	.	.	0.60	.
	Wax-wrapped Sticks	.	.	.	.	1.6	.
	Foil-wrapped Sticks	.	.	.	.	0.70	.
Frozen 12 mo/ Refrigerated 3mo	Bulk	.	.	.	.	.	0.60
	Wax-wrapped Sticks	.	.	.	.	.	1.2
	Foil-wrapped Sticks	.	.	.	.	.	0.90

LSD – least significant difference.

ND – not detected

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Refrigerator/stale flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Appendix 14. Trained panel perception of milkfat flavor intensity separated by temperature treatment and time.**

LSD=0.1	Time					
	0 mo	3 mo	6mo	9 mo	12mo	15 mo
Refrigerated	3.1	3.0	2.6	2.5	2.5	2.0
Frozen	.	.	3.0	.	3.0	2.7
Frozen 6 mo/ Refrigerated 3mo	.	.	.	2.6	.	.
Frozen 12 mo/ Refrigerated 3mo	.	.	.	.	2.5	2.5

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Milkfat flavor was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Appendix 15. Salty taste intensity evaluated by the trained panel separated based on packaging type.**

<b>Packaging</b>	<b>Salty Taste Intensity</b>
Bulk	10.0
Stick	9.0
Foil	9.4
<b>LSD</b>	<b>0.30</b>

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Salty taste was scored on a 15 point universal intensity scale where 0 = absence of attribute and 15 = highest possible intensity of attribute in any product.

**Appendix 16. OSI (hours) over 15 months separated by time, temperature treatment and packaging.**

LSD=1.20		Time					
Temperature	Packaging	0 mo	3 mo	6mo	9 mo	12mo	15mo
Refrigerated	Bulk	10.9	10.7	9.1	5.5	6.3	5.7
	Wax-wrapped Sticks	12.3	10.7	9.7	6.5	6.5	5.2
	Foil-wrapped Sticks	11.4	10.4	6.9	5.9	7.7	4.8
Frozen	Bulk	.	.	8.5	.	8.3	8.6
	Wax-wrapped Sticks	.	.	9.3	.	8.8	8.1
	Foil-wrapped Sticks	.	.	9.3	.	9.0	8.2
Frozen 6 mo/ Refrigerated 3 mo	Bulk	.	.	.	7.2	.	.
	Wax-wrapped Sticks	.	.	.	10.7	.	.
	Foil-wrapped Sticks	.	.	.	8.8	.	.
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	.	.	7.5	.
	Wax-wrapped Sticks	.	.	.	.	7.8	.
	Foil-wrapped Sticks	.	.	.	.	5.5	.
Frozen 12 mo/ Refrigerated 3mo	Bulk	.	.	.	.	.	10.6
	Wax-wrapped Sticks	.	.	.	.	.	8.7
	Foil-wrapped Sticks	.	.	.	.	.	9.8

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Appendix 17. Peroxide value measured by AOCS method 965.33 separated by temperature treatment and time. Results are given in meq peroxide/g oil.**

LSD=0.67		Time		
		0 mo	6mo	12mo
Temperature	Packaging			
Refrigerated	Bulk	0.18	2.8	2.3
	Wax-wrapped Sticks	0.18	3.1	2.8
	Foil-wrapped Sticks	0.21	3.0	2.9
Frozen	Bulk	.	1.2	2.2
	Wax-wrapped Sticks	.	1.9	2.6
	Foil-wrapped Sticks	.	2.2	2.0
Frozen 6 mo/ Refrigerated 6mo	Bulk	.	.	3.0
	Wax-wrapped Sticks	.	.	2.2
	Foil-wrapped Sticks	.	.	3.1

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Appendix 18. FFA measured as % oleic acid equivalents separated by temperature treatment and time.**

**LSD=0.01**

Temperature	Time		
	0 mo	6mo	12mo
Refrigerated	0.21	0.21	0.25
Frozen	.	0.2	0.22
Frozen 6 mo/ Refrigerated 6mo	.	.	0.22

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

**Appendix 19. Yield stress (Pa) measured using the vane method separated by temperature treatment and time.**

**LSD=527**

	Time		
	0 mo	6mo	12mo
Refrigerated	5663	6692	6917
Frozen	.	6335	5876
F3/R6	.	.	.
F6/R6	.	.	6241

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

Yield stress was measured at 19°C

Vane had a diameter of 1.0 cm and a height of 2.5cm

Rotational speed of vane was 0.02 rpm.

**Appendix 20. L\* values for solid butter color, separated based on storage conditions and time.**

**LSD= 0.39**

	Time		
	0 mo	6mo	12mo
Refrigerated	77.8	76.7	79.5
Frozen	.	76.6	79.8
F3/R6	.	.	.
F6/R6	.	.	78.8

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

L\* value on Hunter Scale, where 100=white and 0=black

**Appendix 21. a\* values for solid butter color, separated based on storage conditions and time.**

**LSD= 0.075**

	0 mo	6mo	12mo
Refrigerated	-3.13	-3.54	-3.51
Frozen	.	-3.47	-3.53
F3/R6	.	.	.
F6/R6	.	.	3.45

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

a\* value on Hunter Scale, where + values are red hues and – values are green hues

**Appendix 22. b\* values for solid butter color, separated based on storage conditions, packaging, and time.**

**LSD=0.22**

	Time		
	0 mo	6mo	12mo
Refrigerated	13.41	12.24	13.22
Frozen	.	12.15	-3.53
F3/R6	.	.	.
F6/R6	.	.	12.15

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

b\* value on Hunter Scale, where + values are yellow hues and – values are blue hues

**Appendix 23. L\* values for oil color, separated based on storage conditions, packaging, and time.**

**LSD= 0.14**

	Time		
	0 mo	6mo	12mo
Refrigerated	33.6	33.4	28.5
Frozen	.	33.6	28.5
F3/R6	.	.	.
F6/R6	.	.	28.4

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

L\* value on Hunter Scale, where 100=white and 0=black

**Appendix 24. a\* values for oil color, separated based on storage conditions and time.**

**LSD= 0.04**

	Time		
	0 mo	6mo	12mo
Refrigerated	-1.02	-1.06	-0.92
Frozen	.	-1.06	-1.01
F3/R6	.	.	.
F6/R6	.	.	-0.92

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

a\* value on Hunter Scale, where + values are red hues and – values are green hues

**Appendix 25. b\* values for oil color, separated based on storage conditions and time.**

**LSD=0.16**

	Time		
	0 mo	6mo	12mo
Refrigerated	3.23	2.87	3.09
Frozen	.	2.78	3.57
F3/R6	.	.	.
F6/R6	.	.	3.22

LSD – least significant difference.

Means that differ by more than the LSD are different ( $p < 0.05$ ).

b\* value on Hunter Scale, where + values are yellow hues and – values are blue hues