CHAPTER 4. THE EFFECT OF MILKFAT FRACTION MELTING PROPERTIES ON PHYSICAL PROPERTIES OF 20% REFORMULATED CREAM

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(ABSTRACT)

Low-melt and medium-melt fractionated butteroils with different melting ranges were recombined into dairy systems to manufacture 20% milkfat creams. The butteroils were emulsified using either skim or sweet buttermilk component obtained at two different separation temperatures (49°C and 55°C) and butter-derived aqueous phase from a commercial processor was used with sweet buttermilk. Creaming stability, viscosity, feathering, microbiological analyses, and sensory quality of reformulated creams stored at 3.3°C were evaluated over a 13 day period. Reformulated creams were compared to natural creams processed from skim and cream obtained at 49°C and 55°C separations. Formulation, separation temperature, or melting range characteristics did not significantly (p > 0.01) affect creaming stability of reformulated and control creams. The day within storage period was a significant factor (p < 0.01) in determining creaming stability. All creams displayed characteristic non-Newtownian behavior at 7°C, demonstrated by hysteresis curves in which viscosity decreased as shear rate steadily increased. Formulation and separation temperature used to obtain components did not have a significant (p > 0.01) effect on viscosity; however, melting range characteristics of butteroil used in formulating creams was significant (p ≤ 0.01) at shear rate 692.48 s⁻¹ for creams formulated with sweet buttermilk and butter-derived aqueous phase and at 692.48s⁻¹, 1384.96s⁻¹, and 2769 s⁻¹ for creams formulated with skim component. All creams, except natural creams, had consistent apparent viscosity during the two week storage period. Regardless of formulation, separation temperature, and melting range characteristics of butteroils, all creams feathered in a pH range of 4.70-5.09. Reformulated and natural creams were relatively low in microbiological counts. Reformulated and natural creams met sensory quality specifications as determined by the In/Out Method of Specification, except for creams formulated with skim milk and low-melt fraction butteroil. Creams formulated with buttermilk and butter-derived aqueous phase components had similar creaming stability and sensory quality as natural creams. In considering apparent viscosity, creams consisting of sweet buttermilk obtained at 49°C and butter-derived aqueous phase components, and low-melt fraction butteroil were very comparable to natural creams manufactured from skim and cream obtained at 49°C separation.
Introduction. Milkfat can be separated to yield fractions which are unique in chemical, physical, and organoleptic attributes. These fractions can be used in food applications to achieve optimum milkfat functionality. Khalifa and Mansour (1988) concluded that differences in oxidative stability, texture, and flavor existed among a milkfat liquid fraction at 20°C and milkfat solid fractions at 35, 25, and 20°C obtained by fractionation. Kaylegian and Lindsay (1992) were able to produce cold-spreadable butter from fractionated milkfat having different melting characteristics. The functionality of fractionated milkfat, however, in reformulated dairy products has not been thoroughly investigated. Thus, the question of how reformulated dairy products with different milkfat fractions compare to natural dairy products remains unanswered.

Good emulsion stability is achieved by using components abundant in surface active molecules and efficient processing procedures. Components such as skim milk, sweet buttermilk, butter-derived aqueous phase, whey proteins, casein dispersions, phospholipids, and purified milkfat globule membrane suspensions have proven to successfully emulsify butteroil (Smith and Dairiki, 1975; Oortwijn et al., 1977; Oortwijn and Walstra, 1979; Kanno, 1989; Kanno et al., 1991; Rosenberg and Lee, 1993; Oehlmann et al., 1994; Tomas et al., 1994; Elling et al., 1996). Skim milk is a rich source of whey and casein proteins whereas sweet buttermilk and butter-derived aqueous phase are also abundant in phospholipids which remain after the churning process (Elling et al., 1996). Elling et al. (1996) found that the membrane surface material surrounding the surface of lipid globules in 20% reformulated creams was dependent on the components used in formulation. Elling and Duncan (1996) also found that emulsion stability and physical properties of 20% reformulated creams varied with formulation.

Processing parameters such as heating and homogenization enhance emulsion stability. Over an extended storage period, dairy emulsions will exhibit instability by separating into two distinct layers. Less dense in nature, lipid globules rise to the top of the emulsion, forming an undesirable cream plug. Since the skim phase is denser, it remains at the bottom of the emulsion. Over a 12 month storage period, Yamauchi et al. (1982) noticed a gradual decrease in creaming stability of ultra-heat-treated milk with formation of a cream plug within the first month of storage. Elling and Duncan (1996) found that reformulated and control creams homogenized at 13.6/3.4 MPa had better creaming stability values than those homogenized at 10.2/3.4 MPa.

Other indications of creaming instability are flocculation and coalescence. Flocculation results when milkfat globules join together, forming a collective unit, which contributes to a rise in creaming rate (Mulder and Walstra, 1974). Three potential forms of flocculation are floccules, clumps, and homogenization clusters. Homogenization clusters commonly occur in high fat dairy products such as cream. In a homogenization cluster, at least two fat globules are bonded by a casein micelle, which is simultaneously a component of each fat globules’ surface layer (Walstra, 1983). Monitoring homogenization pressure and fat content can control the formation of homogenization
clusters. If fat content is too high, the surface area of the homogenized fat globules becomes too large for sufficient coverage by surface active molecules. When homogenization pressure is too high, fat globules leave the homogenizer insufficiently covered by proteins from the skim phase. Coalescence is caused by the merging of two lipid globules, resulting in a disruption of the film of continuous phase between two drops that are closely associated with each other (Walstra, 1983). Coalescence contributes to rapid creaming, increased viscosity, and transport of components of the milkfat globule membrane to the plasma phase due to reduction in surface area of lipid globules (Walstra, 1983).

The viscosity of cream is affected by fat content, temperature, storage, and processing conditions (Phipps, 1982; Langley, 1984; Prentice, 1992; Elling and Duncan, 1996). An increase in fat content is accompanied by an increase in apparent viscosity of cream (Prentice, 1972). As the fat content increases, the distance between lipid globules decreases, resulting in an increase in the hindering effect among lipid globules. Lowering temperature promotes cold agglutination of lipid globules, causing a rise in apparent viscosity. Cold agglutination is caused by interactions between complexes of immunoglobulins, particularly IgM, and fat globules (Walstra and Jenness, 1984). Upon cooling, cryoglobulins attach to fat globules, resulting in subsequent flocculation. As floccules grow in size, the creaming rate rises. Processes such as heat treatment and homogenization have an impact on the viscosity of cream. As temperatures rise, apparent viscosity decreases (Prentice, 1992). Unlike heat treatment, homogenization results in an increase in apparent viscosity. The increase in apparent viscosity due to homogenization can be attributed to protein adsorption to lipid globules and formation of homogenization clusters.

Creams display non-Newtonian flow at high fat contents and/or storage temperatures below 40°C (Fox and McSweeney, 1998). Non-Newtonian behavior is characterized by an inverse relationship between apparent viscosity and shear rate. Non-Newtonian fluids demonstrate hysteresis in which the coefficient of viscosity depends on increases and decreases in shear rate (Sherbon, 1988). Cream is considered a Non-Newtonian fluid because it is a colloidal dispersion, consisting of aggregate particles which contribute to a large effective volume due to irregularities in shape (Fox and McSweeney, 1998). An increase in shear rate is accompanied by an increase in shear stress applied to aggregates which results in dispersal of smaller, round forms of the aggregate particles. This process decreases the interstitial space between fat globules, causing a lower total volume fraction of the fat phase and apparent viscosity (Fox and McSweeney, 1998).

Feathering is a quality defect of cream which is characterized by the formation of undesirable particulate precipitate upon the addition of cream to coffee. Homogenized creams are more susceptible to feathering than unhomogenized creams because newly adsorbed protein from the homogenization process destabilize and flocculate with fat globules. Destabilization and flocculation result from the heat and acidity associated with coffee. Resistance to feathering can be achieved by two-stage homogenization,
addition of calcium sequestering agents, and use of additives such as sodium phosphate or sodium citrate (Towler, 1982).

Research related to emulsion stability, physical properties, and organoleptic characteristics of creams formulated with butteroils and milk-derived components is scarce in scientific literature (Smith and Dairiki, 1975; Oortwijn and Walstra, 1982; Kanno, 1989; Melsen and Walstra, 1989; Kanno et al., 1991; Oehlmann et al., 1994; Elling and Duncan, 1996). Melsen and Walstra (1989) found that creams formulated with skim milk and anhydrous milkfat (5-30%) were more stable to coalescence and clumping than natural creams, thus exhibiting superior emulsion stability. Elling and Duncan (1996) found that creams formulated with cholesterol reduced butteroil and emulsified with skim, sweet buttermilk, or sweet buttermilk and butter-derived aqueous phase components and homogenized at 13.6/3.4 MPa had significantly better creaming stability than similar creams homogenized at 10.2/3.4 MPa. For all reformulated and control creams, decreases in creaming stability were evident by day 7 of storage (Elling and Duncan, 1996).

Kanno et al. (1991) observed that apparent viscosity of reconstituted milkfat emulsions increased with an increase in emulsifying time (0-6 min) and an increase in milkfat globule membrane concentration (20, 40, 60, and 80 mg/g fat). Elling and Duncan (1996) also found that reformulated creams had higher apparent viscosities than control creams due to the formation of homogenization clusters. A significant difference in viscosity did not exist between creams homogenized at 10.2/3.4 MPa and 13.6/3.4 MPa (Elling and Duncan, 1996). Elling and Duncan (1996) found that reformulated and control creams feathered from pH 4.86-5.09 regardless of formulation, homogenization pressure, and length of storage period.

The objective of this part of the research was to manufacture a 20% milkfat cream using skim milk or sweet buttermilk and butter-derived aqueous phase components to re-emulsify low-melting and medium-melting modified butteroils. The creams formulated from fractionated butteroils and milk-derived components were analyzed for emulsion stability and physical properties as compared to natural creams. Over a two week storage period, creaming stability, viscosity, and feathering of reformulated creams were analyzed and compared to natural creams. Another objective of this part of the research was to determine the implications of separation temperature in obtaining components and melting point characteristics of butteroil on the physical properties of formulated creams.
Materials and Methods. Separation of Cream and Skim. Separation of Cream and Skim at two different temperatures (49 and 55°C) was completed as described in Chapter III.

Preparation of Components (Skim Milk, Buttermilk, and Butter-Derived Aqueous Phase). Components used in formulating 20% creams were prepared as described in Chapter III.

Cream Reformulation. Emulsification of low-melt and medium-melt butteroils by components into 20% creams was conducted as described in Chapter III. Also, creams were homogenized and pasteurized as described in Chapter III.

Creaming Stability. The emulsion stability of each cream was analyzed by creaming rate over a two week storage period (Elling and Duncan, 1996). On day 0, each cream was placed in 100 ml graduated cylinders, capped, and stored at 3.3°C. Initial fat content was determined on day 0 by the modified Babcock test (Marshall, 1993). The top and bottom portions of each cream were tested on days 1, 3, 5, 7, 11, and 13 of storage using the modified Babcock test (Marshall, 1993). The bottom layer was obtained by pipetting with a 10 ml glass pipette. Duplicate measurements of each formulation were made. The following equation was used to determine the changes occurring in fat percentage of each layer compared to the initial fat content:

\[
\text{Changes in fat percentage} = \left(\frac{\text{Fat content of top or bottom layer}}{\text{initial fat content}}\right) \times 100 - 100.
\]

Viscosity. Viscosity measurements on creams were made on days 1, 7, and 13. On day 0, each treatment was poured into 16x150 mm glass tubes and stored at 3.3°C. Measurements were made with a Haake Rotovisco RV-12 viscometer equipped with a Haake NV spindle and cup (Haake-Buchler Instruments, Paramus, NJ) (Elling and Duncan, 1996). Measurements were made at 7°C maintained by a Haake A82 cooling unit. Shear stress measurements were taken at the following shear rates with 40 second intervals between readings (shear rates (s^{-1}): 173->346->692->1385->2770->1385->692->346->173). Viscosity was determined by dividing the shear stress by the shear rate.

Feathering Stability. The feathering assay is a visual test for cream feathering in sodium acetate buffers at different pH values (pH = 4.70-5.60) at high temperatures (approximately 85°C). The feathering test was conducted on days 1, 7, and 13 of storage at 3.3°C. On day 0, each treatment was poured into 16x150 mm glass test tubes, capped, and stored at 3.3°C. Duplicate measurements were made for each cream. The assay was performed as described by Anderson et al. (1977). The test is designed to mimic feathering conditions which are associated with coffee. Scores ranging from -5 to 5 were used to assess the degree of feathering in each sample. In the feathering assay, the lowest concentration value at which feathering failed to occur was recorded as the
feathering score. Highly stable creams are typically denoted by scores of 5 to 3 whereas stable creams were given feathering scores of 2 to 1 (Atherton and Newlander, 1977). Moderately stable creams had feathering scores of 0 and slightly unstable creams had feathering scores of –1 to –2. Creams with feathering scores of –3 to –5 were unstable and considered to be unmarketable to consumers (Atherton and Newlander, 1977).

Microbiological Analysis. Standard plate count, modified psychrotrophic bacteria count, and coliform bacteria count methods of enumeration were conducted on Day 0 of reformulation to insure creams were pasteurized efficiently and low in spoilage bacteria count prior to sensory evaluation. Aerobic Count and Coliform Count Petrifilm (3M, St. Paul, Minnesota) were used for plating samples. Dairy blanks consisted of solutions of MgPO4, KCl, and deionized water (Marshall, 1993). Dilutions selected for microbial analysis were 10⁰, 10⁻¹, 10⁻², and 10⁻³ and were done in duplicate measurements according to methods outlined in the Standard Methods for the Examination of Dairy Products (Marshall, 1993). Due to cream thickness, a 1:5 dilution of cream in water was made for preparation of the 10⁰ dilution and counts were multiplied by a factor of 5 to express as a 10⁰ dilution. The dilution was made by combining 24.75 gm of cream with 99 ml dairy blank. Petrifilm was incubated at 21°C for 25 h for the modified psychrotrophic bacteria count and at 32°C for 48 h for the standard plate count and coliform plate count methods of enumeration.

Sensory Evaluation of Cream Quality. Creams were evaluated for sensory characteristics within the first four days of refrigerated storage at 3.3°C. Twelve panelists from the Department of Food Science and Technology used the In/Out Method to evaluate the creams (Munoz et al., 1992). Training consisted of familiarizing panelists with the In/Out Method of Specification as well as with potential flavor defects associated with cream products (Appendix C). This particular method was used to determine if creams met quality specifications. Since cream is very susceptible to biochemical degradation and microbial contamination, slight levels of off-flavors were permitted. However, as off-flavors reached moderate levels, creams were deemed "Out" of specification. Training sessions, at total of one per replication, were conducted a week prior to each taste panel. Creams were deemed acceptable in quality if 65% of total responses were "In" specification. Approximately 20 ml each cream was poured into a 1 oz. portion size plastic soufflé cup the day of sensory analysis and refrigerated at 4°C prior to evaluation by panelists. Samples were identified with three digit codes and randomized for presentation to panelists. Panelists were given a total of 8 samples, one sample per cream. Panelists evaluated cream samples in the sensory laboratory of the Department of Food Science and Technology, Virginia Tech.

Statistical Analyses. The effects of separation temperature in obtaining components, cream formulation, and melting range characteristics of butteroil on the physical properties and sensory quality of reformulated and control creams were determined. Creaming stability, viscosity, feathering, and sensory evaluation were analyzed over three replications. Testing of creaming stability, viscosity, and feathering was done in duplicate. Since results from the feathering assay were ordinal data, it was not
statistically analyzed. Responses from all three replications of sensory evaluation were combined and analyzed as percent values. A split plot design was used for analysis of data generated from creaming stability and viscosity testing. A split plot design was chosen for analysis because the effects of several parameters were being tested (formulation, separation temperature in obtaining components, and melting characteristics of butteroil). Also, each formulation was tested on the same day with equal numbers of days between subsequent testing days. Statistical contrasts were made in formulation: Skim milk creams were tested against buttermilk creams; buttermilk creams against natural creams; and natural creams against skim milk creams. Within treatments without regard to melting range characteristics of butteroil, the cream set processed from components obtained at 49°C were compared to the 2 creams manufactured from components obtained at 55°C. Within reformulated cream treatments, the two creams consisting of low-melt fraction butteroil, one consisting of components obtained at 49°C and the other processed from 55°C components, were statistically compared to medium-melt butteroil creams processed from 55°C components. A p-value of 0.01 was used to determine significance. With 0.01 as a p-value, there is a minimum 16% [(0.01 x 2 x 8 contrasts) x 100%] chance of committing a Type 1 Error. Using a p-value of 0.05, however, results in a minimum 80% [(0.05 x 2 x 8) x 100%] chance of making a Type I Error. Statistical analyses were conducted using SAS (Cary, NC).

Results and Discussion. Processing techniques and surface active components, such as proteins and phospholipids, affect emulsion stability and physical properties of creams. Proteins, particularly caseins, tend to associate with lipid globules at pasteurization temperatures (McPherson et al., 1984; Houlihan et al., 1992; Kim and Jimenez-Flores, 1995). Also, creaming associated with milkfat emulsions is delayed since heat processing halts cold agglutination. Heat treatment also causes a decrease in apparent viscosity with increasing temperatures (Langley, 1984; Prentice, 1992). Creams obtained at separation temperatures exceeding 40°C are more Newtonian in behavior due to the loss of cryoglobulins, causing less cold agglutination. The homogenization process enhances emulsion stability and physical properties of creams by reducing the size of the fat globules and dispersing them throughout the emulsion, preventing the formation of a cream plug and improving creaming stability. During homogenization, the original milkfat globule membrane is destroyed. However, proteins from the aqueous phase become adsorbed to the surfaces of lipid globules, increasing surface area. Thus, homogenization increases the viscosity of creams. The formation of homogenization clusters also contribute to an increase in viscosity.

Components used in formulation of creams are rich in surface active components which improve emulsion stability. Skim milk is a good source of casein and whey proteins. Sweet buttermilk and butter-derived aqueous phase, also rich in proteins, are abundant in phospholipids (Elling et al., 1996). Sweet buttermilk and butter-derived aqueous phase obtained from churning contain fragments of original milkfat globule membrane material. Elling et al. (1996) found that when used in formulations, these components yielded creams with very good creaming stability. Creams formulated with skim component and creams formulated with sweet buttermilk and butter-derived aqueous phase components did not differ (p ≤ 0.01) in the amount of protein associated with lipid globules or total
protein in cream (Tables 3-5 and 3-7) (Scott, 1999) (Chapter III). However, all formulations consisting of buttermilk and butter-derived aqueous phase were significantly higher in total phospholipid content and than all creams formulated with skim component (Tables 3-5 and 3-7). Also, buttermilk+butter-derived aqueous phase formulations were comparable to the natural creams in phospholipid concentration.

Creaming Stability. A decrease in creaming stability is noted by the formation of a cream plug at the surface of the milkfat emulsion. Over time, milkfat progressively rises to the top of the emulsion. Changes in creaming stability can be monitored by analyzing fat contents of both top and bottom layers of the milkfat emulsion.

Creaming stability of reformulated and natural creams was analyzed every 48 hours over a two week storage period at 3.3°C. Over the two week storage period all creams displayed a decrease in creaming stability, denoted by an increase in percent change in fat content of the top layer of the emulsion (Figure 4-1). Formulation, separation temperature used in obtaining components, and melting range characteristics did not significantly (p > 0.01) affect creaming stability of the top layer of reformulated and natural creams.

Length of storage period, however was very significant (p < 0.01) in determining the fat content of the top and bottom layers (Table 4-1). Changes in creaming stability of the top layer of reformulated and natural creams are somewhat small when considering the first week of refrigerated storage. After a week of refrigerated storage, however, changes in the fat content of the top layer became more apparent. For reformulated creams, day of significance in determining creaming stability was greater for creams formulated with low-melt fraction than was for those creams formulated with medium-melt fraction butteroil.

Fat content of the bottom layer was also analyzed over the two week storage period (Figure 4-2). As storage period increased, fat content of the bottom layer decreased. Creaming stability of the bottom layer of each cream was not significantly (p > 0.01) affected by formulation, separation temperature in obtaining components, or melting range characteristics. Like the top layer, creaming stability of the bottom layer was significantly (p ≤ 0.01) affected by day (Table 4-2). For most creams, significant (p ≤ 0.01) differences in creaming stability were detected earlier than those associated with the top layer, indicating that some interaction may have occurred between the top and bottom layers of the emulsions. For most creams, significant (p ≤ 0.01) differences in creaming stability were noted as early as Day 3 or 5.

Homogenization at 13.6/3.4 MPa yielded fairly stable creams. By day 13 the total percent change in fat of the top layer ranged from 9.5% to 21.1% (Table 4-1). Elling and Duncan (1996) used two homogenization pressures, 10.2/3.4 MPa and 13.6/3.4 MPa, to determine the efficiency of homogenization in yielding stable creams. Elling and Duncan (1996) found that the higher homogenization pressure of 13.6/3.4 MPa yielded more stable creams than homogenization at 10.2/3.4 Mpa. By Day 13 creaming stability values of the top layer of creams homogenized at 13.6/3.4 ranged from
Figure 4-1. Creaming stability (percent change in the fat content of the top layer) of natural creams and reformulated creams formulated with low-melt or medium-melt butteroils over a two week storage period at 3.3°C.
Table 4-1. Creaming stability (total percent change in fat content of top layer) for reformulated creams formulated with low-melt and medium-melt butteroils and natural creams processed from components obtained at two separation temperatures (49°C and 55°C)

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Separation temperature(°C)</th>
<th>Day of significance (Top)</th>
<th>Total % change in fat on day of significance</th>
<th>Total % change in fat over 13 day storage</th>
</tr>
</thead>
<tbody>
<tr>
<td>20%lmbo + 80%skim</td>
<td>49</td>
<td>9</td>
<td>12.0%</td>
<td>21.1%</td>
</tr>
<tr>
<td>20%lmbo + 80%skim</td>
<td>55</td>
<td>9</td>
<td>6.2%</td>
<td>9.5%</td>
</tr>
<tr>
<td>20%mmbo + 80%skim</td>
<td>55</td>
<td>7</td>
<td>4.2%</td>
<td>12.0%</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>49</td>
<td>13</td>
<td>14.1%</td>
<td>14.1%</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>11</td>
<td>9.6%</td>
<td>12.1%</td>
</tr>
<tr>
<td>20%mmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>11</td>
<td>9.4%</td>
<td>15.4%</td>
</tr>
<tr>
<td>Natural cream</td>
<td>49</td>
<td>9</td>
<td>15.0%</td>
<td>18.9%</td>
</tr>
<tr>
<td>Natural cream</td>
<td>55</td>
<td>13</td>
<td>11.5%</td>
<td>11.5%</td>
</tr>
<tr>
<td>Standard error</td>
<td></td>
<td></td>
<td>1.715</td>
<td></td>
</tr>
</tbody>
</table>

1lmbo = low-melt butteroil; mmbo = medium-melt butteroil; bm = buttermilk; ap = aqueous phase
Figure 4-2. Creaming stability (percent change in the fat content of the bottom layer) of natural and reformulated creams formulated with low-melt and medium-melt butteroils over a two week storage period at 3.3°C.
<table>
<thead>
<tr>
<th>Formulation</th>
<th>Separation temperature(°C)</th>
<th>Day of significance (Botm.)</th>
<th>Total % change in fat on day of significance</th>
<th>Total % change in fat over 13 day storage</th>
</tr>
</thead>
<tbody>
<tr>
<td>20%lmbo + 80%skim</td>
<td>49</td>
<td>5</td>
<td>3.3%</td>
<td>22.7%</td>
</tr>
<tr>
<td>20%lmbo + 80%skim</td>
<td>55</td>
<td>5</td>
<td>5.0%</td>
<td>16.2%</td>
</tr>
<tr>
<td>20%mmbo + 80%skim</td>
<td>55</td>
<td>5</td>
<td>2.5%</td>
<td>12.4%</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>49</td>
<td>5</td>
<td>3.2%</td>
<td>19.0%</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>3</td>
<td>1.2%</td>
<td>16.4%</td>
</tr>
<tr>
<td>20%mmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>1</td>
<td></td>
<td>17.7%</td>
</tr>
<tr>
<td>Natural cream</td>
<td>49</td>
<td>1</td>
<td></td>
<td>17.1%</td>
</tr>
<tr>
<td>Natural cream</td>
<td>55</td>
<td>1</td>
<td></td>
<td>17.0%</td>
</tr>
<tr>
<td>Standard error</td>
<td></td>
<td></td>
<td></td>
<td>1.646</td>
</tr>
</tbody>
</table>

1lmbo = low-melt butteroil; mmbo = medium-melt butteroil; bm = buttermilk, ap = aqueous phase
4.7% to 24.1%, with the highest value associated with the skim milk formulation and the lowest value associated with the control (Elling, 1995).

Although significant differences were not detected among treatments, natural cream processed from components obtained at 55°C separation had the best creaming stability. The greater stability to creaming associated with the natural cream is indicated by low percent change in fat content of the top layer during the storage period (Table 4-1). When comparing reformulated creams, all formulations consisting of sweet buttermilk and butter-derived aqueous phase closely resembled natural cream manufactured from components obtained at 55°C in creaming stability. Differences between natural cream from components at 55°C and creams formulated with sweet buttermilk and butter-derived aqueous phase became apparent, however, as storage time progressed. Better creaming stability associated with natural cream and creams consisting of sweet buttermilk and butter-derived aqueous phase can be attributed to presence of more native milkfat globule membrane fragments. Unlike skim milk formulations, creams composed of buttermilk and butter-derived aqueous phase are abundant in native milkfat globule membrane constituents such as phospholipid material. These particular constituents remain after the churning process. Since buttermilk and butter-derived aqueous phase components are rich in native milkfat globule membrane constituents, they probably demonstrate better emulsifying characteristics than skim milk. Natural creams and creams formulated with sweet buttermilk and butter-derived aqueous phase had higher (p ≤ 0.01) amounts of total phospholipids and phospholipids occurring in the surface material associated with lipid globules than creams formulated skim component (Tables 3-5 and 3-7) (Scott, 1999. Chapter III). Elling and Duncan (1996) found that buttermilk formulated creams homogenized at 13.6/3.4 MPa and refrigerated at 3°C displayed better creaming stability than skim milk formulated creams homogenized and stored under the same conditions. Elling et al. (1996) found that creams formulated with sweet buttermilk or sweet buttermilk and butter-derived aqueous phase had higher (p ≤ 0.05) total phospholipid than natural creams and creams formulated with skim. However, no difference (p ≤ 0.05) existed in amount of phospholipid associated with surface material occurring in natural creams, creams formulated with sweet buttermilk (80%), or sweet buttermilk (70%) and butter-derived aqueous phase (10%). Also, Smith and Dairiki (1975) found that creams emulsified with phospholipids were stable to creaming. Furthermore, Smith and Dairiki (1975) found that increasing phospholipid concentration enhanced creaming stability.

Viscosity. Apparent viscosity of reformulated and natural creams was monitored at different shear rates over a two week storage period at 3.3°C. The effects of formulation and separation temperature in obtaining components were not significant (p > 0.01). Within formulations, however, creams processed from components obtained at 55°C had higher apparent viscosity values than creams processed from components obtained at 49°C (Figure 4-3). When analyzing apparent viscosity values among formulations processed from components obtained at 55°C and low-melt fraction butteroil, higher apparent viscosity values were associated with natural creams, followed by creams formulated with skim milk and creams formulated with sweet buttermilk and butter-
Figure 4-3. Hysteresis curve at 7°C of apparent viscosity of natural creams and creams formulated with low-melt butteroil (LM) and components obtained at two separation temperatures on Day 1 of storage (— increasing shear; - - decreasing shear)
derived aqueous phase (Figure 4-3). Lower apparent viscosity values occurred with creams consisting of buttermilk. This observation may be attributed to a higher phospholipid and unsaturated fatty acid content which contributes to increased fluidity. In addition to low-melt fraction butteroil, the sweet buttermilk and butter-derived aqueous phase were sources of unsaturated fatty acids (Scott, 1999. Chapter III).

Depending on shear rate, however, butteroil characteristics had significantly different effects within formulations \( (p \leq 0.01) \). For creams formulated with skim component and creams formulated with sweet buttermilk and butter-derived aqueous phase, at a shear rate of 692.48 s\(^{-1}\) creams formulated with low-melt fraction butteroil were significantly different \( (p \leq 0.01) \) than those formulated with medium-melt fraction butteroil (Figure 4-4). At shear rate 1384.96 s\(^{-1}\) and 2769.92 s\(^{-1}\), only creams consisting of skim and low-melt fraction significantly \( (p \leq 0.00625) \) differed from their skim component and medium-melt fraction formulated counterparts. Higher apparent viscosity values occurring in creams formulated with medium-melt fraction butteroil resulted from the higher saturated fat content. In comparison to low-melt fraction butteroil, medium-melt fraction butteroil had a higher degree of saturated fatty acids, resulting in a more compact, crystalline structure with an increased melting point (Kaylegian, 1998).

Hysteresis curves were produced to describe the flow characteristics of reformulated and natural creams (Figures 4-3 and 4-4). For the majority of creams apparent viscosity was fairly consistent throughout the storage period. Therefore, the hysteresis curves for Day 1 will be discussed. Failing curves (increase in shear rate) proved that creams displayed non-Newtonian behavior which is characterized by decreases in apparent viscosity due to increases in shear rate. Additionally, rising curves (decrease in shear rate) demonstrated an increase in viscosity as shear rate was lowered. When comparing rising curves to failing curves, higher apparent viscosity curves were associated with failing curves. This observation can be attributed to break down of colloidal aggregate particles as increasing shear was applied during the first stage of viscosity measurement (Fox and McSweeney, 1998).

Apparent viscosity curves were produced to determine if apparent viscosity was consistent for reformulated and natural creams during the two week storage period. Since significant effects \( (p \leq 0.01) \) were observed at a shear rate of 692.48 s\(^{-1}\), graphs were plotted at this particular shear rate. Values at this shear rate are also noted (Appendix D, Table D-3). Apparent viscosity values for most creams, with little fluctuation, were consistent from Day 1 to Day 13 of refrigerated storage (Figure 4-5). Both natural creams, however, increased drastically in apparent viscosity by Day 7. By Day 13 of storage, the viscosity of the natural cream processed from skim and cream obtained at 49°C decreased. The natural cream processed from skim and cream obtained at 55°C increased linearly. Apparent viscosity did not differ \( (p > 0.01) \) throughout the 13 storage period for reformulated creams. The reason behind the behavior of the natural creams is not quite understood. Inspection of some electron micrographs of natural creams processed from 55°C components indicate some coalescence which may contribute to
Figure 4-4. Hysteresis curve at 7°C of natural cream and creams formulated with low-melt (LM) and medium-melt (MM) butteroils and components obtained at separation temperature of 55°C on Day 1 of storage (--- increasing shear; - - decreasing shear)
Figure 4-5. Apparent viscosity (shear rate = 692 s⁻¹) at 7°C of natural creams and reformulated creams with low-melt or medium-melt butteroil over a two week storage period at 3.3°C.
increases in viscosity. Overall, day within storage did not have a significant effect on apparent viscosity. However, a treatment*day effect was found to be significant ($p < 0.01$). In a similar study, Elling and Duncan (1996) found that formulation was the only factor to have a significant effect ($p \leq 0.05$) on apparent viscosity at a shear rate of 692.48 s$^{-1}$. Creams consisting of sweet buttermilk or combinations of sweet buttermilk and butter-derived aqueous phase had higher viscosity values, followed by skim milk formulations. By Day 13, creams consisting of sweet buttermilk and butter-derived aqueous phase homogenized at 13.6/3.4 MPa were not statistically significant ($p > 0.05$) from natural creams homogenized at both pressures while creams of other formulations were different ($p \leq 0.05$). Higher apparent viscosity values associated with reformulated creams was attributed to the presence of homogenization clusters. Although not statistically ($p > 0.05$) significant, creams homogenized at 13.6/3.4 MPa had higher apparent viscosity values than similar creams homogenized at 10.2/3.4 MPa over the two week storage period. Also, very little fluctuation occurred in apparent viscosity of both reformulated and natural creams during the 13 day refrigerated storage period.

Feathering Stability. Feathering is a defect occurring in cream upon addition to coffee. Feathering is characterized by the formation of undesirable particulate matter, particularly destabilized proteins and fat globules (Fox and McSweeney, 1998). The incidence of feathering in coffee is attributed to heat and acidity which causes protein destabilization and aggregation. Regardless of formulation, separation temperature used to obtain components, melting range characteristics, and length of storage period, reformulated and natural creams tended to feather between pH 4.70-5.09 (Appendix D, Table D-4). Therefore, feathering scores of 0 to –1 were issued for most creams, and natural and reformulated creams were characterized as moderately stable or slightly unstable. Overall, natural creams were slightly more stable to feathering than reformulated creams. Creams processed from sweet buttermilk and butter-derived aqueous phase components seemed to be more comparable to natural creams in incidence of feathering than creams formulated with skim component. Anderson et al. (1977) found that 18% milkfat creams which were UHT pasteurized feathered in a pH range of 4.70-5.20. Elling and Duncan (1996) found that reformulated creams processed from a reduced cholesterol butteroil and components like those used in this study and natural creams feathered between pH 4.86 to 5.09.

Susceptibility to feathering can be related to use of a higher homogenization pressure of 13.6/3.4 MPa and multiple heat processing steps. During the homogenization process, proteins become adsorbed to lipid globules. Most of these newly adsorbed protein, particularly casein and whey, are destabilized by increases in heat and acidity. Multiple heating processing steps used during processing of reformulated and natural creams (i.e. pasteurization of components and creams) most likely contributed to sensitivity to denaturation when added to hot buffer solutions.

Microbiological analysis. Cream is an excellent reservoir for microbial contamination. Cream has the ideal nutrients, pH, and water activity for microbial growth and
proliferation. Spoilage bacteria contribute to various deteriorating reactions, resulting in off-flavors. Shelf-life is improved by pasteurization and refrigeration. Aerobic, modified psychrotrophic bacteria count, and coliform bacteria count methods of enumeration were conducted on Day 0 of reformulation. Microbiological analysis was used to insure creams were pasteurized efficiently and low in spoilage bacteria prior to sensory evaluation. Due to efficient pasteurization and subsequent refrigeration, bacterial counts for all creams were low (< 50 cfu/ml) (Appendix D, Table D-5). Higher counts were associated with the aerobic plate count, which are expected since pasteurization insures complete destruction of pathogenic microorganisms, but not all spoilage microbes.

Sensory Evaluation of Cream Quality. Very little research has been conducted on sensory evaluation of cream, and even less on reformulated creams. Cream should have a clean, slightly sweet, and slightly cooked flavor (Jensen and Poulsen, 1992). The body and texture of cream should be smooth, free of lumps and fat plugs. The potential occurrence of off-flavor and texture defects, however, is possible. The presence of off-flavors and poor texture may be indicative of various factors, including low emulsion stability, temperature abuse, processing conditions, and storage conditions.

The majority of creams were considered to be "In" specification (Table 4-1). During training sessions, panelists were familiarized with off-flavors common to cream resulting particularly from biochemical degradation (i.e. oxidation or lipolyzed) and microbial contamination (i.e. fruity or malty). Samples were considered to be "In" specification if a slight off-flavor was noticed. However, as off-flavor intensity increased to moderate levels, samples were considered "Out" of specification. Textural problems were not considered in determining if natural or reformulated were "In" specification. Apparent viscosity was utilized to determine textural information about natural and reformulated creams. Natural cream processed from components obtained at 55°C and creams formulated with sweet buttermilk obtained at 55°C, butter-derived aqueous phase, and low-melt fraction or medium-melt fraction were consistently rated "In" specification (Table 4-1).

Panelists noted that all creams had a cooked flavor. Cooked flavor was caused by high temperature short time pasteurization of components and creams, resulting from \( \text{H}_2\text{S} \) formation. At temperatures above 70°C, proteins begin to denature, resulting in the exposure of cysteine residues (Fox and McSweeney, 1998). As a result, proteins engage in Maillard browning reactions with lactose, causing cooked flavors. In regard to all creams formulated with sweet buttermilk and butter-derived aqueous phase components, panelists used descriptors such as rich and creamy to characterize flavor. Both natural creams were described as having good dairy flavors. The skim component formulations processed from low-melt butteroil fractions were considered "Out" of specification by experienced panelists (Table 4-1). The formulations consisting of skim component and low-melt fraction butteroil were criticized as being oxidized. The low-melt butteroil was the source of oxidative flavor characteristics as determined by smelling of the butteroil by the principal investigator prior to reformulation.
Table 4-3. Percent of "In" specification responses for 20% natural and reformulated creams within 4 days of refrigerated storage at 3.3°C.

<table>
<thead>
<tr>
<th>Formulation</th>
<th>Temperature(°C)</th>
<th>Percent &quot;In&quot; Specification Responses</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>20%lmbo + 80%skim</td>
<td>49</td>
<td>52.8*</td>
<td>Oxidized, flat, cooked</td>
</tr>
<tr>
<td>20%lmbo + 80%skim</td>
<td>55</td>
<td>58.3*</td>
<td>Oxidized, cooked</td>
</tr>
<tr>
<td>20%mmbo + 80%skim</td>
<td>55</td>
<td>72.2</td>
<td>Flat, cooked</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>49</td>
<td>72.2</td>
<td>Rich cream flavor, cooked</td>
</tr>
<tr>
<td>20%lmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>83.3</td>
<td>Rich cream flavor, cooked</td>
</tr>
<tr>
<td>20%mmbo + 70%bm + 10%ap</td>
<td>55</td>
<td>83.3</td>
<td>Rich, sweet flavor, cooked</td>
</tr>
<tr>
<td>Natural cream</td>
<td>49</td>
<td>66.7</td>
<td>Good dairy flavor, cooked</td>
</tr>
<tr>
<td>Natural cream</td>
<td>55</td>
<td>83.3</td>
<td>Good dairy flavor, cooked</td>
</tr>
</tbody>
</table>

1lmbo = low-melt butteroil; mmbo = medium-melt butteroil; bm = buttermilk; ap = aqueous phase
* Denotes "Out" of specification (percent "In" < 65%)
Oxidation flavors were expected to occur in low-melt fraction butteroil, as opposed to medium-melt butteroil since it has a higher degree of unsaturated fatty acids. Skim component, having a lower fat content than butter-derived aqueous phase and sweet buttermilk, was more sensitive in detection of oxidation flavors associated with low-melt butteroil and more susceptible to being described as flat in flavor. Sweet buttermilk and butter-derived aqueous phase components have many flavor compounds associated with rich and creamy dairy notes. Statistics indicate that an increase in milkfat sales has accompanied a 25% decrease in margarine sales, due to demand of good dairy flavors (Honer, 1993). Therefore, oxidation of creams formulated with low-melt fraction butteroil emulsified by sweet buttermilk and butter-derived aqueous phase was undetectable by panelists.

Conclusions. Length of storage period was the only significant (p < 0.01) factor affecting emulsion stability of natural and reformulated creams. Good emulsion stability displayed throughout the two week storage period can be attributed to effective homogenization at 13.6/3.4 MPa. Although significant (p > 0.05) differences were not detected in creaming stability of creams, skim milk and natural creams processed from components obtained at 55°C had better creaming stability than those obtained at 49°C separations. Utilization of a higher separation temperature could have contributed to more efficient separation of components and inactivation of cold agglutination.

All creams displayed non-Newtonian behavior, marked by a decrease in viscosity due to an increase in shear rate. The melting range characteristics of butteroils used in reformulated creams had a significant (p ≤ 0.01) impact on apparent viscosity. Creams formulated with medium-melt butteroil had higher viscosity values than creams formulated with low-melt butteroil at higher shear rates. This may be attributed to a higher degree of unsaturated fatty acids in the low-melt butteroil which enhance fluidity. Although separation temperature did not have a significant effect (p ≤ 0.01), reformulated and natural creams processed from components obtained at 55°C had higher apparent viscosity values than creams processed from components obtained at 49°C. Sensory evaluation provided great insight about the flavor characteristics of individual components used in processing reformulated creams. Sweet buttermilk and butter-derived aqueous phase components have a higher fat content and possibly more lipid derived flavor components than skim component. Thus, sweet buttermilk and butter-derived aqueous phase component formulated creams successfully masked observed oxidation flavors of low-melt fraction butteroil. Natural creams were described as having typical rich dairy flavors. The next step would be to conduct more involved sensory testing on the flavor attributes of the individual components used in formulating creams.

The creams most effective in mimicking the physical and organoleptic characteristics of natural cream were the creams formulated with buttermilk and butter-derived aqueous phase components. Similarities existed in creaming stability, feathering stability, and sensory quality. Future studies should focus on ratios of milk-derived components required to optimize the physical and organoleptic characteristics. More in depth
knowledge is also required regarding the texture and flavor attributes of fractionated butteroils having different melting properties.
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