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Formulation Of A Fruit Slush Using Evaporated Sweet Whey

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FORMULATION OF A FRUIT SLUSH USING

EVAPORATED SWEET WHEY

By

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Whey-fruit slush formulas were evaluated prior to consumer testing of pre-selected formulated beverages. Varying ingredients were prepared in accordance to a factorial design of maximum use of whey and minimum use of additional ingredients. Whey was obtained from the Mississippi State University Dairy Processing Plant and evaporated. The evaporated sweet whey was combined with blueberries, cherry concentrate, Splenda®, water and ice. Sensory tests were conducted to evaluate appearance, flavor and overall acceptability of formulations. Panelists were asked to participate in a survey to elaborate personal perceptions of the products. Formulation of 125mL whey and 30g blueberries had the higher acceptability score, but was not different from the beverage with 150mL whey and 30g blueberries. These two formulas were tested for chemical analysis. Adequate levels of antioxidants, total phenolics and neutral pH were observed. Results from proximate analysis showed minimal caloric levels with low presence of protein and carbohydrate.

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CHAPTER I

INTRODUCTION

Previously whey was considered a by-product of cheese production, but due to its value as a source of protein, minerals, and lactose, whey is now considered a co-product of cheese-making (Walzem et al. 2002). The disposal of whey has put tremendous pressure on wastewater treatment facilities and finding uses for whey has been a priority in the dairy industry. Two types of whey that are produced in the manufacture of cheese are sweet whey, which is produced from rennet-coagulated cheese manufacture, and acid or sour whey, which is produced from cottage cheese (Anonymous 2007). Along with major components, essential amino acids are present in whey including the branched chain amino acids (BCAA's) valine, leucine and isoleucine. These BCAA's play a vital role in regards to the nutritional value of whey due to their capability to act as metabolic regulators in protein and glucose homeostasis, lipid metabolism, and possibly weight control (Smilowitz et al. 2005; Smithers 2008; Zemel 2004). In addition, BCAA's enable the stimulation process regarding protein synthesis recovery (Cribb 2003).

Endless proto types have been made to benefit from the important nutritional properties of whey and increasing experimentation has been performed and product formulation has improved (Onwulata et al. 2004). Creating beverages that are included in the value-added food market is a priority for many food corporations and adding whey is an important step in many cases. Past attempts to create whey-based beverages have usually failed in the attempt to compete in the market due to the sedimentation problem, which occurs during shelf stability, but this can be avoided by the use of high methoxyl pectins which can prevent whey separation in the presence of small casein particles (Glahn and Rolin 1994; Jelen et al. 1987; Koffi et al. 2005; Parker et al. 1993).

It is known throughout the dairy industry that whey contains undesirable flavors and aroma, and attempts have been made to mask these flavors and aromas in order to enhance consumer acceptability. Djurić and others (2004) noted that because of the unappealing taste of whey, the high lactose to glucose ratio and excessive acidity, research has been conducted to enable the direct utilization of whey in food products to enhance human nutrition. The blending of tropical fruits and berries to the whey component has added value from a nutritional health standpoint (Djurić et al. 2004). Grapefruit juices and other citrus juices such as orange are compatible with acid whey but grapefruit juice consumption has declined in the U.S. (Branger et al. 1999; Florida Citrus Mutual 1997). However, acidic whey-based beverages have been associated with astringent type flavors which may cause undesirable appeal to consumers (Beecher 2006; Lee and Vickers 2008; Sano et al. 2005).

Berries add significant value to food products as well as beverages. Blueberries have been associated with very strong flavors, so blending them with other fruit juices or diluting the juice from blueberries with water has been considered in product development applications (Luh 1980; Tipton 1999). In addition, blueberries have a low sugar concentration and therefore, have little influence on sweetness. However, blueberries provide excellent nutrition due to their levels of antioxidant activity, which include flavonoids and anthocyanins, compounds that add value to food products (Main et al. 2001; Prior et al. 1998). Cherries are another fruit containing high antioxidant activity. Montmorency Tart Cherry, which comes from the species of *Prunus cerasus L.*, can also contribute to antioxidant capacity in food products. Like blueberries, Montmorency Tart Cherry also has high anthocyanin activity and contains high levels of polyphenolics (Chaovanalikit and Wrolstad 2004).

Since blueberries have a low sugar concentration, additional sweetening attributes have to be included when making a beverage. To uphold nutritional value, as well as to attempt to keep undesirable flavors from whey at a minimum, Splenda® can be utilized to sweeten beverages containing whey. In addition, Montmorency Tart Cherry concentrate can also be used in beverages as a source of antioxidants that may offset off-flavors due to whey incorporation by contributing a strong sweet and sour flavor.

A nutritional food product was developed with blueberries, cherry concentrate, and concentrations of whey to determine the maximum amount of whey that can be used without negatively impacting consumer acceptability. Fresh evaporated whey is a potentially good medium for microbial growth (Marek et al. 2003) and is known to be very shelf unstable, so this product was designed as a frozen slush to avoid microbial growth. In addition, keeping the beverage near a frozen state may reduce sedimentation problems from the separation of the liquid variable and lactose within the whey.

CHAPTER II

REVIEW OF LITERATURE

Whey is currently considered a co-product from cheese production, but was previously considered a by-product of the dairy industry until approximately 1998. Prior to 1998, whey was disposed of which caused pollution problems due to its heavy organic nature and high chemical oxygen demands (Gannoun et al. 2007; Mockaitis et al. 2006). In addition, information regarding compositional quality of whey has been inconsistent (Philippopoulos and Papadakis 2008). Whey is derived from cheese production in which butterfat, casein, and important mineral components separate from the milk in the formation of curds (Bilgin et al. 2006). The production of whey from rennet-coagulated casein is referred to as sweet whey, and that which is produced from mineral or lactic acid coagulated casein is referred to as acid whey (Fuente et al. 2001). Upon separation, the whey stream is formed which consists of whey proteins, lactose and other mineral components, and is the liquid that is drained from the curd and used for further processing or manufacture (Punidadas et al. 1999). These further processes are identified as modified whey, dry whey or simple whey in the formation of animal feed.

According to Shon and Haque (2007), there is an environmental dilemma regarding the disposal of whey into lakes or spraying onto agricultural land due to the biological oxygen demand at 35-45 kg m^{-3} of whey arising from cheese and casein manufacture. However, it has also been reported by Lehrsch et al. (2008) that the use of whey on certain soils provides natural soluble salts that reduce the diffuse double-layer thicknesses of clay and incorporates lactose and protein in the soil. The lactose and protein may stimulate the growth of aerobic microbes that produce polysaccharides to promote fungal growth, which in turn, may improve the structure of eroded or non-sodic soil (Lehrsch et al. 2008).

The composition of liquid whey following drainage is approximately 10% to 12% total solids, which varies according to cheese type with protein representing only 0.7% to 0.8% within a water basis (Anonymous 2007). The chemical composition of cheese whey directly relies on the chemical composition of the milk that is obtained, which fluctuates due to the types of feed, breed, individual animal differences, environment and climate (Casper et al. 1998; Johansen et al. 2002; Quiles et al. 1994).

Whey Processing Methods

To enhance nutritional value, several processing techniques are used to utilize specific components of whey. These technical methods include pasteurization, vacuum evaporation, ultra filtration, reverse osmosis, ion exchange, gel filtration, electro dialysis, crystallization and spray drying (Ji and Haque, 2003; Speer 1998). The vacuum evaporation method is one of the most promising practical methods for the recovery of solid content while avoiding the depletion of the nutritive and functional properties of the whey proteins (Haque and Ji 2002; Smith et al. 1984). Following vacuum evaporation to remove substantial volumes of water, certain methods can be used to enhance the whey's functionality.

Two types of whey that are produced in the manufacture of cheese are sweet whey, which is produced from rennet-coagulated cheese manufacture, and acid or sour whey, which is produced from the production of cottage cheese (Anonymous 2007). Whey has been further separated into specific categories such as evaporated whey and dry whey, which are obtained when a substantial amount of water from sweet or acid whey is removed during manufacture. Whey protein concentrate (WPC) can then be produced from evaporated whey by ultra-filtration (Heino et al. 2007). After filtration, the protein content is approximately 25% to 89% (Davis 2004). This is achieved by a pressure-driven process in which substances with molecular weights between10^3 and 10^6 are separated and concentrated, while the solvent and other components that are present pass through the membrane and are collected as permeate (Atra et al. 2004).

To increase the purity of the protein within the whey, ion exchange or micro-filtration can be performed to produce whey protein isolates (WPI), valuable proteins that are present in low concentrations of cheese but are concentrated to 90% protein and higher (Davis 2004; Neville et al. 2001). WPI and WPC are differentiated by protein concentration and the fact that lactose is removed from WPI but not WPC, which decreases WPI's sweetness. Whey protein isolates are well known for their functional and biological applications and the composition of the protein is standardized by technology that is used for the recovery and analysis of protein (Ounis et al. 2008). Such technology can be derived from a variety of methods which include electrophoretic techniques (Bonfatti et al. 2008; Hang and Kroener, 1984; Kim and Florez, 1994), isoelectric focusing (Bonfatti et al. 2008; Kim and Florez, 1994) and capillary electrophoresis and capillary zone electrophoresis (Bonfatti et al 2008; Ferreira and

Cacote 2003; Miralles et al. 2001). For high resolution accuracy, and reproducible results, high performance liquid chromatography combined with mass spectrometry can be performed to utilize rapid results that includes automated analysis in which significant separation is characterized (Bonfatti et al. 2008).

In addition to these categorical processes, the two major protein components that are present in whey are α-lactalbumin and β-lactoglobulin. The α-lactalbumin component is one of the main proteins in human milk and contains readily digestive capabilities regarding amino acids. The β-lactoglobulin component is represented by approximately half of all protein within the whey of cow's milk. However, it is absent from human milk (Séverin and Wenshui 2005). Minor components present, but of equal importance, are immunoglobulins and sphingolipids which have powerful antimicrobial properties. These components survive digestion processes and reach the large intestine where they implement their biological effects (Causey and Thomson, 2003). Further noted, specific roles of concentrated whey components have bioactive capabilities that promote and enhance intestinal health.

Proper equipment to sufficiently process whey to maximize its nutritional value is an overwhelming obstacle for many companies within the dairy industry. Balagtas et al. (2003) noted that some cheese processors have yet to obtain the necessary technology and funding to purchase and install whey-processing equipment in smaller and older plants. As further noted, some dairy products are produced in conjunction with other products, such as cheese and whey. Manufacturers can only increase production of whey if they use more milk which leads to competition with other dairy producers. Thus, less milk will be available for other dairy products which can lead to higher prices. Such trade-offs are key factors for the determination of implications regarding the dairy economy of an increased demand for whey (Balagtas et al. 2003).

Nutritional Composition of Whey

Whey has significant nutritional value as well as containing important components that enhance human physiological function. In respect to this, many food industries whether they are dairy, meat, pasta or bakery manufacturers use whey in their food products. A thoroughly researched area within the nutritional composition of whey is that of the branched-chain amino acids (BCAA's), which are leucine, isoleucine and valine. According to Bos and others (2000); Ha and Zemel (2003), the BCAA's consist of approximately 26% of the total composition of whey (Bos et al. 2000; Ha and Zemel 2003). The amino acid leucine plays a vital role in protein metabolism, thus contributing to muscle protein synthesis. In addition, other important essential amino acids, including leucine, play important roles as signaling molecules and substrates for the synthesis of new proteins in the protein synthetic pathway (Ha and Zemel 2003).

The major proteins derived within the natural product of whey primarily consist of α-lactalbumin, which has excellent properties of emulsifying and foaming (Lopez et al. 2007), β-lactoglobulin, which accounts for approximately 50% by mass of whey protein and has good gelation mechanisms (Anandharamakrishnan et al. 2007), immunoglobulins, which are bioactive compounds that are effective in boosting immune protection and enhancing post-exercise recovery (Talbott and Hughes 2007), and bovine serum albumin, which is reported to have anti-mutagenic functions and cancer prevention mechanisms (Bosselaers et al. 1994; Laursen et al. 1990; Madureira et al. 2007).

Several methods and techniques for whey protein fractionation such as ion exchange chromatography and enzymatic isolation have been performed to isolate these proteins for further use. However, according to Konrad and Kleinschmidt (2007), aside from enzymatic isolation, these schemes of fractionation have been observed as impracticable outside of the laboratory. In addition, to obtain high purity of α-lactalbumin, a method of selective denaturation of β-lactoglobulin has been proposed, but this method still has disadvantages including irreversible denaturation of all other whey proteins present (Kiesner et al. 2000; Konrad and Kleinschmidt, 2007; Tolkach et al. 2005). Fuda and others note that most of these techniques effectively fractionate the protein for enrichment on a laboratory scale as well; however, these processes are not effective on a commercial scale due to inadequate yield or purification. In addition, the given conditions are incompatible with the maintenance of any intrinsic biological activity (DeSilva et al. 2003; Fuda et al. 2004).

Aside from the high protein content in whey, other nutritional values of whey are documented. Whey has antioxidant capabilities as well as antibody activity due to its immunoglobulin component and containment of lactoferrin, a non-enzymatic component that consists of 698 amino acid residues (Marshall, 2004). Tu et al. (2002) also observed that antimicrobial activity in lactoferrin was much more efficient when used in combination with immunoglobulin and enhanced iron bioavailability to intestinal cells (Hambraeus and Lönnerdale 1982; Tu et al. 2002). Studies done by Min and others noted that naturally occurring bioactive compounds promoting antimicrobial activity in lactoferrin, lysozyme and lactoperoxidase systems are important ingredients in food due to their ability to inhibit microorganisms by binding iron, making lactoferrin unavailable to microorganisms (Appelmelk et al. 1994; Jones et al. 1994; Losso et al. 2000; Min et al. 2005; Naidu 2000 Proctor and Cunningham 1988; Shah 2000; Tomita et al. 2002; Walzem et al. 2002). Other antioxidant activity in whey can also come from its ability to contribute cysteine-rich proteins that are able to aid in the synthesis of glutathione; a potent intracellular antioxidant (Marshall, 2004).

Health Benefits

Whey proteins are commonly used as a nutritional supplement in the athletic and health industries. However, other health attributes have been evaluated from consumer consumption of whey. Bioactive functions in collaboration with amino acids within whey components are vital to the immune system for individuals involved in high levels of intense physical activity. Ha and Zemel (2003) noted that certain amino acids and whey-derived bioactive compounds offer extended beneficial health potential to people with higher physical activity. In addition, most functions derived from these amino acids and whey components involve immune system functionality, which is suppressed when subjects are under rigorous training or participating in excessive physical activity. Supporting this, immunoglobulins, lactoperoxidase, and lactoferrin, which can be concentrated from whey, have been identified as contributors to immunity in the gastrointestinal tract, and are iron-binding proteins. Iron is an element that works as a prebiotic. These proteins survive passing through the stomach and small intestine and are able to seize iron from bacteria in the lower bowel. Since many pathogens have high iron requirements, this property of lactoferrin makes it broadly antimicrobial in nature (Geiser 2007). Immunoglobulin proteins have been reported to bind to bacterial toxins to rid the intestinal tract of pathogenic organisms. Lactoperoxidase and lactoferrin play important roles in host immunity through their antibacterial action on pathogenic microorganisms (Causey and Thomson 2003). These whey-derived bioactive compounds demonstrate the capability to enhance intestinal health. Certain proteins commonly found in whey have shown prebiotic effects, which are in foods that have beneficial affects in the stimulation of growth activity of a limited number of bacteria in the colon. The lactose in whey supports the growth of lactic acid bacteria such as *bifidobacteria and lactobacilli,* microorganisms which are capable of utilizing prebiotics. These bacteria are beneficial due to their antimicrobial effects against pathogenic bacteria, production of essential B vitamins and inhibition mechanisms that function against intestinal enzymes that are considered potential precarcinogens (Causey and Thomson 2003).

Other components of whey that add value include the sulfur-containing amino acids cysteine and methionine. These specific amino acids have the ability to act as precursors in the production of the tripeptide, glutathione (GSH), which has the capability to moderate oxidative damage and enhance immune function (Archibald 2002). Tseng and others concluded that WPC, which is known as a potential antioxidant, has the capability to protect cells from ethanol damage that results in oxidative damage, and this protection includes its capacity to stimulate GSH synthesis (Tseng et al. 2005). In support of these findings, Blouet and others reported that increased levels of dietary cysteine in rats which were given a high sucrose pro-oxidant diet had significantly improved cellular GSH synthesis status and decreased oxidative stress. In addition, these researchers found that higher levels of dietary cysteine improved glucose homeostasis which is the modulator for maintaining blood sugar balance at appropriate levels (Blouet et al. 2007). In another study, Kennedy noted that the supplementation of whey reduced the GSH that is available to cancerous cells. In addition, whey protein has the capability to reduce the GSH levels of cancer cells even though GSH levels and growth rates are increased in normal healthy cells. However, these same healthy cells are less prone to proliferation and more resistant to chemotherapy while the cancer cells are more vulnerable (Kennedy et al. 1995).

Aside from the immune enhancing effects of whey protein, cardiovascular health can also be enhanced. Bioactive compounds within whey protein have positive effects on cardiovascular health by exhibiting angiotensin converting enzyme (ACE) inhibitor, antithrombotic activity and cholesterol-reduction activity. Jacobucci and others observed that whey protein reduced cholesterol blood serum in rats compared to casein and soy proteins. In addition, their results showed strong evidence that rats fed a diet containing 20% WPC for 45 days had significantly lower cholesterol production within the liver when compared to diets with no WPC (Jacobucci et al. 2001).

Whey proteins have important effects when contributing to preventive cancer activity. Beneficial whey proteins have been reported to inhibit cancer cell growth (Walzem et al. 2002). Nukumi and others found that whey acid protein had strong inhibitory effects on invasive breast cancer cells, which they noted was a crucial event associated with the mortality of cancer patients. These researchers concluded that whey acid protein had demonstrated vital functions in the degradation of laminin, which depressed proliferation and tumorigenesis (Nukumi et al. 2006). Conjugated linoleic acid (CLA) is a component in whey and has been reported as a potent anticancer agent in studies performed on human malignant breast and colon cancer cell lines. In addition, CLA consumption may inhibit the growth and spread of mammary tumors, but necessary dietary intake levels have yet to be confirmed (Hoolihan 2004).

Whey is considered a dairy product and many people feel the need to consume dairy products for calcium intake to promote bone health. However, good bone health extends beyond calcium content with the clustering of essential nutrients which include vitamin D, phosphorous, magnesium, vitamin A, vitamin B_6, and trace elements such as zinc (Hoolihan 2004). However, vitamin D can be depleted during the pasteurization method or sterilization and by the removal of part of the milk cream which contains lipid-soluble vitamins (Banville et al. 2000). A study performed by Kruger and others, in which they observed that protein fractions derived from whey were important in the reduction of bone loss due in ovariectomy, which is the surgical removal of one or both ovaries. These researchers concluded that prepared fractions from WPC significantly reduced bone loss in rats, and further noted that this reduction might have been attributed to beneficial bioactive compounds in the whey that preserved bone mass (Kruger et al. 2005). Narva and others found similar results when observing the effects of whey fermented with *Lactobacillus Helveticas.* These researchers reported that components in the whey increased bone mass in the bone marrow of mice that was cultured (Narva et al. 2004). Supporting these data findings, Takada and others fed an increased whey protein diet to ovariectomy rats and observed a significant increase in bone marrow breaking strength. They noted that whey protein influenced bone metabolism by increasing the amount of bone proteins such as collagen, and that bone breaking force that is derived from strength seemed to be enhanced (Takada et al. 1997).

Whey Beverage Product Development

Due to consumer demand of low-carbohydrate, high-protein foods, the research and development branches in food industries have reformulated many of their products to have increased protein levels, particularly protein from whey. Since whey contributes nutritional benefits, food industries try to utilize whey applications along with upholding consumer supply and demand. Developmental beverage formulation prototypes using whey as the main ingredient in liquid products were reported as early as 1975. Djurić and others noted references of whey beverage concepts appearing in 1975, when Bangert suggested a whey-based orange drink concentrate with citric acid as an acidifier (Bangert 1975; Djurić et al. 2004).

Since technology has advanced, further analysis has been performed to optimize beverage formulations using whey. Beecher and others noted by controlling the pH in whey-protein based beverages; solubility of whey proteins could be maintained. This retained clarity throughout the beverage (Beecher et al. 2008). Solubility is extremely important when formulating value-added beverages. Whey protein contents are reported to be least soluble at a pH of 5.2 and increasing temperature will decrease solubility (Beecher et al. 2008; Phillips et al. 1994). However, lowering pH with the use of phosphoric acid will increase solubility of whey proteins (Beecher et al. 2008; Pelegrine and Gasparetto, 2005). Whey proteins are very sensitive with the increase in temperature, which leads to denaturation. When this occurs, the proteins become insoluble and aggregation develops (Pelegrine and Gasparetto, 2005). In addition, Koffi and others note that whey proteins such as α-lactalbumin and β-lactoglobulin are susceptible to heat-induced physical and chemical changes via heat-processes that occur at temperatures above 60° C (Alting et al. 2000; de la Fuente et al. 2002; de Wit 1981; Euston et al. 2000; Jelen and Bucheim 1984; Koffi et al. 2005; Law and Leaver 2000; Rattray and Jelen 1997).

Another challenge that is related to formulating whey-based beverages is the production of off-flavors that may occur due to the use of high concentrations of whey protein. It is important to use additional ingredients to try to mask such off-flavors to obtain good balance of all derived flavors. Depending on processing techniques used to obtain evaporated whey, WPC or WPI, flavor attributes will fluctuate. A study by Mortenson and others was conducted to detect differences in flavor attributes between WPI and WPC using gas chromatography–olfactometry (GC–O) and gas chromatography-mass spectrometry (GC–MS) using different processing methods and different types of cheese whey. They concluded that flavor did not vary between WPC and WPI despite different processing methods; however, WPC was slightly sweeter than WPI due to lactose content (Mortenson et al. 2007). These results are important for whey beverage formulation because a sweetening effect within beverages is a necessity in most cases for consumer acceptability.

A sweet flavor is desirable to most consumers and sweeteners are known to promote pleasurable taste attributes. However, at intake levels exceeding 25% total energy, dietary quality suffers (American Dietetic Association 2004). A popular sweetener used today in soda beverages is High Fructose Corn Syrup (HFCS). However, it is noted that HFCS is the leading source of fructose in the diet and has been correlated with dramatic increases in obesity (Melanson et al. 2006), but it is easily utilized and is very cost effective. Non-nutritive sweeteners, which do not provide a source of energy, include five compounds that are approved by the U.S. Food and Drug Administration (American Dietetic Association 2004; Kroger et al. 2006). These include acesulfame-K, aspartame, neotame, saccharin and sucralose (Table 1).

Sucralose is a non-caloric sweetener derived from sucrose in a five step process that selectively substitutes three atoms of chlorine for three hydroxyl groups in the sucrose molecule. In addition, it can be transcribed as a free flowing, white crystalline solid that is freely soluble in water and stable both in its crystalline and liquefied forms (Grice and Goldsmith 2000). According to absorption, distribution, metabolism and elimination studies, sucralose has very limited absorption, rapid urinary excretion, and minimal metabolism of absorbed material (Baird et al. 2000; John et al. 2000; Roberts et al. 2000; Sims et al. 2000; Wood et al. 2000). Sucralose is a very intense sweetener, usually 600 times sweeter than sucrose; therefore low doses can be applied to meet intensity levels comparable to sucrose (Binns 2003).

Table 1. Non-nutritive sweeteners approved in the United States.

Sweeteners Approved for use in the United States	Brand Names	Sweetening Intensity (times sweeter than sucrose)*
Acesulfame-K	Sunett, Sweet One	200
Aspartame	NutraSweet, Equal	160-220
Neotame	Information not available	7,000-8,000
Saccharin	Sweet and Low, Sweet Twin, Sweet'n Low	200-700
Sucralose	Splenda	300-600

*Sweetening intensity varies in different food applications. Information according to the American Dietetic Association (2004) and Kroger, Meister and Kava (2006).

In addition to a sweet taste, fruit flavors are generally liked by consumers also. Blueberries have increased in popularity in recent years due to their high antioxidant concentrations and ability to enhance health. Blueberries are known to contain some of the richest sources of antioxidant phytonutrients and reported to contain three times as much as the total amount found in red raspberries (Kalt et al. 1999; Nindo et al. 2005). There are several different species of blueberry, such as highbush blueberries (*Vaccinium corymbosum*) and lowbush blueberries (*Vaccinium augustifolium),* which are known to have potent scavenging activity against radical oxygen species due to their content of anthocyanins and certain phenolic compounds (Kalt et al. 2001; Mansour et al. 2005; Prior et al. 1998). Another type of blueberry species is rabbiteye (*Vaccinium ashei*), which also contains anthocyanin compounds.

A study was conducted by Kay and Bruce (2002) in which subjects were fed a high fat meal and the same high fat meal one week later followed by a 100g blueberry supplement. They concluded that a significant increase in antioxidants was observed increasing effectiveness against chronic degenerative diseases (Kay and Bruce 2002). However, a similar study was performed by Dunlap and others (2005), where they too, observed the effects of using blueberry supplements. These researchers evaluated if a blueberry supplement would elevate plasma total antioxidant power and help aid sled dogs in short term exercise stress recovery and prevent muscle damage. They concluded that muscle damage was increased after exercise regardless of blueberry supplementation (Dunlap et al. 2005).

Cherry fruit is another source of antioxidant activity and includes sweet cherry (*Prunus avium L.)* and sour cherry (*Prunus cerasus L.*) (Vursavus et al. 2005). According to Tural and Koca (2008), Cornelian cherry (*Cornus mas L.)* has a sour taste when ripe and contains significant amounts of anthocyanins, which have antioxidant and anti-inflammatory effects. In addition, the Cornelian cherry is used as a product of medical treatment of diarrhea and has been used to enhance liver and kidney functions (Celik et al. 2006; Tural and Koca 2008). Tural and Koca (2008) observed significant levels of natural antioxidants in the Cornelian cherry which have the ability to contribute good health benefits upon consumption. A study to support anti-inflammatory effects of cherries was performed by Tall and others (2003). These researchers observed the efficacy of using anthocyanins that were extracted from tart cherries on inflammation-induced edema and pain behavior in rats. Their results indicated that tart cherries suppress behaviors associated with acute inflammation and may have a beneficial role in the treatment of inflammatory pain, despite further studies that are needed to determine precise mechanisms of action (Tall et al. 2003).

Sensory Testing

In the food industry, sensory science is used to understand consumer likes and dislikes and determine preferences regarding food products to ensure the food company's success. A mass selection of techniques and methodology exists throughout the field of sensory science from which sensory practitioners are able to depict evaluation schemes to observe consumers liking or disliking towards specific products (Hein et al. 2008; Lawless and Heymann 1999; Stone and Sidel 2004). Aside from liking and disliking of specific products, further detail of methodology enables sensory practitioners to observe preferences of consumers for a variety of food products. Young and others note that in products where variability is induced by preparation methods such as formulation schemes, panelists may observe differences in appearance, aroma, noise, flavor, texture, and overall acceptability which do not reflect treatment differences (Young et al. 2008).

Sensory practitioners use various methods and analyze data from panelist responses to optimize product formulations. Rating scales, such as the 9-point hedonic scale, are often used since these scales are reported to have stability of responses and can be utilized as a sensory benchmark for any particular product category (Moskowitz et al. 2006). Preferences can be further analyzed through cluster analysis, in which panelists are grouped in a representative cluster by a latent variable that represents product preference and liking for each cluster (Sahmer et al. (2004; Vigneau et al. 2001). In addition, panelists may be grouped by their degree of correlated behavior or observations based on degrees of similarity among their ratings. Further noted, two classes of cluster analysis are used consisting of hierarchical and nonhierarchical methods. Distinguishing between the two, using the hierarchical method, an observation assigned to a cluster is unable to be moved to another cluster, while an observation is able to be moved using the nonhierarchical method (Meilgaard et al. 2000).

Using sensory testing, it is very important to have the given product meet the same criteria within the testing site as the subject would consume or use the product at home or preferred area. If not, there should be no environmental variables to promote bias during sensory evaluation. Lawless and Heymann note that testing sites should remain in their simplest form, and secluded to avoid interruptions and consistently remain in a quiet manner to achieve higher success. In addition, the avoidance of contact among panelists is an important task for them to avoid influence, so personal booths for sensory testing are a necessity (Lawless and Heymann 1997).

CHAPTER III

MATERIALS AND METHODS

Process Method

Edam cheese-whey was obtained from the Mississippi State University Edward W. Custer Dairy Processing Plant and transported to the Mississippi State University Ammerman-Hernsberger Food Processing Plant using 5-gallon plastic buckets with lids which held approximately 42 pounds of whey. Ten minutes later, arriving at the Ammerman-Hernsberger Food Processing Laboratory, the whey was immediately filtered using a Halco kitchen strainer to discard clumps within the liquid.

The Edam cheese-whey, when collected, was approximately 3% soluble solids to 97% liquid, as measured with a refractometer (Vee Gee ABT-32. Kirkland, WA). Following filtration, the whey was concentrated in a vacuum evaporator (Model 26061, Year 1990, APV Inc. Tonawanda, NY; Søborg, DK) for 60 minutes. The temperature of the evaporator was held between 140°F – 150°F and vacuum pressure was held at –0.3 kp/cm^3, which increased the concentration of soluble solids by reducing the water concentration. This process was stopped when the percentage of solids was 25 – 30% °brix by refractometer (Vee Gee ABT-32. Kirkland, WA) to avoid burnt sensory attributes. Following vacuum evaporation, the volume of cheese-whey concentrate was approximately 1800mL from the initial 5 gallons (42 pounds) and transported in 5-gallon buckets with lids to a laboratory (Room 258) in the Herzer Building within 15 minutes for product formulation.

Formulation

Frozen rabbiteye blueberries (Blueberry Growers CO-OP Association, Flora, MS) and blueberry juice (*Vaccinium ashei*) that were stored in the Mississippi State University Ammerman-Hernsberger Food Processing Plant and leftover from a project by Stojanovic and Silva (2007) were obtained. Additionally, Montmorency Tart Cherry Concentrate (Transverse Farms Inc. Bellaire MI), Splenda® (McNeil Nutritionals LLC, Fort Washington, PA), and shaved ice from an ice machine (Scotsman Fairfax Operation Model AF325AE-1B. Fairfax, SC) in the Mississippi State University Ammerman-Hernsberger Food Processing Plant were obtained. When formulating the whey-fruit slush products, a 500mL graduated cylinder was used to hold 500mL of the evaporated cheese-whey and a 100mL graduated cylinder was used to hold distilled water. Prior to mixing, a hot plate/stirrer was used to hold a 400mL beaker and a magnetic stirrer was used in the beaker for constant rotation of liquid mixture. Desired volumes of cheese-whey (50, 75, 100, 125, and 150mL) and distilled water (10, 20, 30, 35, 45, 55, 60, 70, 80, 85, 95, 105, 110, 120, and 130mL) were collected using a 10mL pipette and transferred to the 400mL beakers. Following this, a separate 10mL pipette was used to transfer 10mL of Montmorency Tart Cherry Concentrate to the cheese-whey and water mixtures. Varying amounts of frozen blueberries were weighed (10, 20, and 30g), using a weight balance (Mettler Toledo, Columbus, OH) followed by 5g of Splenda® added to each mixture and constantly stirred for 2 min. An additional 10mL pipette was used to transfer blueberry juice to a plastic container to equal the weight of frozen blueberries using a weight balance (Mettler Toledo, Columbus, OH). Amounts of blueberry juice equal to the weights of frozen blueberries were added to appropriate treatments to compare product quality between products made with whole blueberries and blueberry juice. Each ingredient/treatment combination equaled 200mL. When the liquid was thoroughly mixed, the 200mL formulated liquid sample was then transferred to a 200mL plastic container with snap-on top and immediately placed in a refrigerator for 24-hour cold storage (4.5°C). After storage, 347g (3cups of ice) were added to a kitchen blender, followed by the 200mL formulated liquid sample and blended 30 seconds on mix speed. This produced a slush product and these slush formulations were then poured into 2oz plain white sample cups (Sweetheart, Owings Mills, MD) and kept chilled at –18°C in a Frigidaire commercial freezer (Model FFC15C3AW2, Electrolux Home Products Inc. Cleveland, OH) for 20 minutes prior to semi-trained/consumer taste panels.

The objective was to make a 200mL frozen beverage (slush) and determine the maximum volume of cheese-whey and minimum concentration of fruit that could be added without sacrificing consumer acceptability. Varying concentrations of evaporated whey (25–30% soluble solids), water, and fruit (blueberry and cherry) were used with 5g of Splenda® for each beverage. For the first evaluation, two frozen beverages were prepared, one with frozen blueberries and the other with blueberry juice, and each also contained 100mL evaporated sweet whey, 70mL distilled water, 10mL cherry concentrate, 247g ice, and 5g Splenda®. The pre-selected sensory panel then tasted both slush products and the results were evaluated to see if there was a perceived difference ($p<0.05$) between the products to determine which blueberry source (whole blueberries or juice) should be utilized in the slush formulation. The next evaluation consisted of three separate sections of varying concentrations of blueberry juice or whole blueberries in the range of 10, 20 and 30mL, each representing a different section. Each of these concentrations of blueberry juice or whole blueberries was added to varying volumes of evaporated sweet whey in the range of 50, 75, 100, 125, and 150mL. All beverages were tasted by the pre-selected taste panel and the two beverages from each section with the highest acceptability scores were used for consumer testing.

Radical Scavenging Activity (%DPPH)

Chemical analyses of antioxidant activity, pH, and total phenolics were conducted according to Lee and others (2003). Antioxidant activity was analyzed by adding .0025g (0.041mM) of 2, 2-Diphenyl-1-picrylhydrazy (DPPH) (Sigma Chemical Company, St. Louis, MO) to 100mL of 100% ethanol HPLC solution (control) (Sigma-Aldrich Inc. St. Louis, MO). The solution was refrigerated (35°F) 24 hrs in a 125mL bottle with screw cap and wrapped in foil for protection against light. Two milliliters of DPPH solution were then transferred into each of 4 cuvettes and 0.4mL of the 125mL whey-30g blueberries sample was added to 3 cuvettes and 0.4mL of the 150mL whey-30g blueberries sample was added to the other 3 cuvettes. Each cuvette was placed into a UV-VIS spectrophotometer (Model UV-1201, Shimadzu Scientific Instruments. Columbia, MD) at 750nm and observed (Blois 1958; Lee et al. 2003).

The following calculation was used for the determination of antioxidant activity:

$$\% \text{ DPPH activity} = \frac{\text{Control optical absorbance} - \text{Sample optical absorbance}}{\text{Control optical absorbance}}$$

Measurement of pH

The pH was measured by using a Fisher Scientific pH meter (PD12-10. San Jose, CA). Two samples of 125mL whey-30g blueberries and two samples of 150mL whey-30g blueberries were observed separately by placing the meter into each sample for approximately 1 minute.

Total Phenolics

Total phenolics were analyzed and estimated by using the Folin-Ciocalteau reagent (Gutfinger 1981; Lee et al., 2003). One hundred µL of samples 125mL whey-30g blueberries and 150mL whey-30g blueberries were added to separate volumes of 900µL of HPLC water. Using cuvettes, 20µL of the solution were added to 1.58mL of HPLC water for further dilution. Following this, 100µL of Folin-Ciocalteau reagent were then added to each cuvette and incubated at room temperature for 5 minutes. After incubation, 300 µL of sodium carbonate were added to each cuvette and incubated for 2 hrs at room temperature. Following incubation, each cuvette was placed in a spectrometer and observed at 765nm.

The following calculations were used for the determination of antioxidant activity (gallic acid was used as the standard):

-y = .5791 (standard constant curve of slope) × - .0023 (standard constant of sample solution).

Standard constant (absorbance) – Standard constant = Total Phenolics

Proximate Analysis

The two most preferred whey-fruit slush samples were individually mixed into separate 200g batches for proximate analysis which was performed by the Mississippi State Chemical Laboratory located at Mississippi State University. Percentage moisture was measured in triplicate for each formulation using a drying oven (AOAC. 1995. Method 39.1.02). Percentage protein was determined in triplicate by the AOAC Method 4.2.08 (AOAC. 2000. Model FP-528, LECO Corp., St. Joseph, MI.). Crude fat content (%) was determined in triplicate using a fat extractor (AOAC. 2000. Method 39.1.05; Model 1043, Soxtec HT Extraction Unit, Tecator, Hoganas, Sweden) and percentage ash was measured in triplicate using a muffle furnace (Model Isotemp, Fisher Programmable Muffle Furnace; Fischer, Pittsburgh, PA). Percentage carbohydrate was calculated by subtracting percentage moisture, protein, fat and ash from 100%. Caloric content was determined by using a Parr Adiabatic Calorimeter (Model 1241, Parr Instrument Company, Moline, IL).

Institutional Review Board Approval

The Mississippi State University Institutional Review Board (IRB) approved all research protocol pertaining to subjects (Appendix A). Each subject provided written, informed consent to participate in the study (Appendix B).

Sensory Analysis

Sensory testing was conducted with semi-trained panel subjects (n=8) to evaluate and pre-screen appearance, flavor and overall acceptability (Appendix C) of product samples (n=17). Once the formulated samples were grouped according to the most preferred (n=6), consumer sensory testing of the product samples were evaluated for appearance, flavor, and overall acceptability (Appendix C) with an appropriate number of responses ($n \geq 150$) (Appendix C).

Additionally, a 7-item survey (Appendix D) was developed using principles recommended by Fowler (1993). The survey included questions regarding consumers' personal preferences and choices of the product samples. Additional consumer testing was completed at the Mississippi State University Joe Frank Sanderson Center operated by the Recreational Sports Administration and is referred to as the Fitness Center.

Statistical Analysis

A pre-selected sensory panel using eight panelists was conducted to evaluate appearance, flavor, and overall acceptability. Once the formulations were selected based on maximizing whey concentration without affecting consumer acceptability, consumer sensory testing of the product was performed using sensory panels ($n>100$). Three repetitions ($n \geq 50$ per replication) were performed to determine significance of hedonics as well as any observed differences. Data were analyzed using SAS (version 9.1.2, 2005, SAS Institute Inc., NC). A randomized complete block design with three replications was used to determine if differences existed within each treatment among replications. When significant differences ($p<0.05$) occurred among treatments, the Least Significant Difference (LSD) test was utilized to separate treatment means. Agglomerate hierarchical clustering was performed using Ward's Method to cluster consumers together based on their preference and liking of the products (treatments) (Everitt et al. 2001; Schilling and Coggins 2007; Ward 1963). A dendrogram and a dissimilarity plot were used to determine how many clusters should be utilized to group consumers (Schilling and Coggins 2007). After cluster analysis was performed, randomized complete block designs were utilized to determine differences ($p<0.05$) among treatments within each cluster. When significant differences ($p<0.05$) occurred for a response within a cluster, the LSD test was performed to separate treatment means.

CHAPTER IV

RESULTS AND DISCUSSION

Preliminary Research Team Sensory Evaluation

Prior to consumer panel sensory testing, a preliminary research team (n=8) was assembled to pre-screen whey-fruit slush beverage formulations using blueberry juice or whole blueberries. All other variables within the slush formulations, with the exception of water, were the same to control for bias. This testing was performed to determine if differences ($p<0.05$) existed in taste attributes between beverage formulations 1 and 2 (Table 2).

Table 2. Slush formulations pre-screened to determine preference of blueberry juice or whole blueberries for preliminary research team sensory testing.

Ingredients	Slush Formulation 1	Slush Formulation 2
Whey	100mL	100mL
Distilled Water	70mL	70mL
Blueberry Juice	20mL	0mL
Cherry Concentrate	10mL	10mL
Ice	347.0g	347.0g
Whole Blueberries	0.0g	20.5g
Splenda®	5.0g	5.0g

No difference ($p>0.05$) was detected between the two beverage formulations (Table 3). Therefore, whole blueberries were utilized in the study since blueberry juice is more expensive than whole blueberries due to additional processing that is necessary to extract the juice. Blueberries are also generally more expensive than other fruits (Main et al. 2001), so the beverage needs to be as cost-effective as possible.

Table 3. Effects of the usage of blueberry juice or whole blueberries within a fruit slush on preliminary research team acceptability of appearance, flavor and overall acceptability determined by using pre-selected panels (n = 8).

Sample 100mL of whey	Appearance Acceptability Mean ± SE[a]	Flavor Acceptability Mean ± SE[a]	Overall Acceptability Mean ± SE[a]
Blueberry Juice[b]	6.9 ± 0.22	7.1 ± 0.36	7.1 ± 0.32
Blueberry Whole[c]	7.0 ± 0.30	6.9 ± 0.27	7.1 ± 0.18

[a]Mean score ± Standard Error (SE). Hedonic scale was based on 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely)
[b]Blueberry juice = 20mL
[c]Whole blueberries = 20.5 grams

The preliminary research team evaluated 15 different whey-fruit slush beverage formulations to determine which slush formulations should be used for consumer sensory testing (Table 4). The objective was to identify the 2 beverage formulations from each set for use in consumer testing. Each set of samples was evaluated on a different day to prevent panelist fatigue. These formulations were selected based on maximizing the concentration of whey and minimizing the amount of blueberries to determine the most cost-effective treatment that was deemed acceptable by the preliminary research team. With each set, 5 different concentrations of evaporated sweet whey were used to identify the threshold of whey that could be used for each formulation.

In Set 1, 10g of whole blueberries were used with each of the 5 different concentrations of whey (Table 4). On a hedonic scale of 1 to 9 with 1 being dislike extremely and 9 being like extremely, all formulations were rated favorably for overall acceptability with scores between 6.3 and 7.3 with no differences (p>0.05) among the 5 formulations.

Table 4. Formulations of fruit slush beverages that were pre-screened to determine the formulations for consumer sensory testing.

Set 1*					
Whey	50mL	75mL	100mL	125mL	150mL
Water	130mL	105mL	80mL	55mL	30mL
Cherry	10mL	10mL	10mL	10mL	10mL
Ice	347.0g	347.0g	347.0g	347.0g	347.0g
Blueberry	10.0g	10.0g	10.0g	10.0g	10.0g
Splenda®	5.0g	5.0g	5.0g	5.0g	5.0g
Set 2*					
Whey	50mL	75mL	100mL	125mL	150mL
Water	120mL	95mL	70mL	45mL	20mL
Cherry	10mL	10mL	10mL	10mL	10mL
Ice	347.0g	347.0g	347.0g	347.0g	347.0g
Blueberry	20.0g	20.0g	20.0g	20.0g	20.0g
Splenda®	5.0g	5.0g	5.0g	5.0g	5.0g
Set 3*					
Whey	50mL	75mL	100mL	125mL	150mL
Water	110mL	85mL	60mL	35mL	10mL
Cherry	10mL	10mL	10mL	10mL	10mL
Ice	347.0g	347.0g	347.0g	347.0g	347.0g
Blueberry	30.0g	30.0g	30.0g	30.0g	30.0g
Splenda®	5.0g	5.0g	5.0g	5.0g	5.0g

*Set = Formulations used for preliminary research team sensory test (n=8).

The two beverage formulations with the highest numerical values regarding overall acceptability (50mL whey-10g blueberries, 125mL whey-10g blueberries) were chosen for consumer taste panels. Twenty grams of whole blueberries were used with each of the 5 different concentrations of whey in Set 2 (Table 4). The preliminary research team scored all formulations with 20g of blueberries between 6.6 and 7.3 for overall acceptability. No differences ($p>0.05$) existed among these formulations regarding acceptability (Table 5), and all scores were around like moderately. These formulations (125mL whey-20g blueberries and 150mL whey-20g blueberries) were chosen for further testing since their values were similar to all other treatments but maximized whey concentration in the formulation.

In Set 3 (Table 4), there were no differences ($p>0.05$) in acceptability of appearance among treatments, but differences ($p<0.05$) occurred among treatments for acceptability of flavor and overall acceptability. Acceptability scores increased ($p<0.05$) from 6.8 to 7.7 as whey concentration increased (Table 5). This demonstrates that the high concentrations of whey did not impart negative sensory properties. When adding larger amounts of whole blueberries (30g), taste attributes may fluctuate negatively or positively, depending on the consumer. Juices that are derived from blueberries are known to have flavor attributes that have been associated as being strong with no sweetening affect (Tipton et al. 1998). Recognizing these flavor attributes, unappealing flavors might have been produced when the concentration of blueberries was increased, but not to the point where the flavor became undesirable. The beverage formulations (125mL whey-30g blueberries and 150mL whey-30g blueberries) were also chosen for their higher concentrations of whey and higher scores ($p<0.05$) for overall acceptability when compared to other treatments. Based on sensory panel data and maximal whey usage, the following formulations (50mL whey-10g blueberries, 125-10, 125-20, 150-20, 125-30 and 150-30) were selected for consumer sensory testing.

Table 5. Effects of varying concentrations of whey and blueberries within a fruit slush on preliminary research team acceptability of appearance, flavor and overall acceptability determined by using pre-selected panels (n=8).

Sample (mL whey - g blueberry)	Appearance Acceptability*	Flavor Acceptability*	Overall Acceptability*
50Ml - 10.0g	7.1[a]	6.9[a]	7.0[a]
75mL - 10.0g	7.0[a]	6.5[a]	6.3[a]
100mL - 10.0g	7.3[a]	6.9[a]	6.7[a]
125mL - 10.0g	7.2[a]	7.2[a]	7.3[a]
150mL - 10.0g	7.1[a]	6.7[a]	6.8[a]
Standard Error	0.15	0.21	0.25
50mL - 20.0g	7.4[ab]	6.8[a]	6.8[a]
75mL - 20.0g	7.1[b]	6.5[a]	6.6[a]
100mL - 20.0g	7.4[ab]	7.2[a]	7.1[a]
125mL - 20.0g	7.2[b]	7.2[a]	7.2[a]
150mL - 20.0g	7.5[a]	7.0[a]	7.3[a]
Standard Error	0.08	0.26	0.32
50mL - 30.0g	7.4[a]	6.6[c]	6.8[c]
75mL - 30.0g	7.6[a]	7.0[bc]	7.1[bc]
100mL - 30.0g	7.3[a]	7.3[abc]	7.4[ab]
125mL - 30.0g	7.5[a]	7.5[ab]	7.6[a]
150mL - 30.0g	7.4[a]	7.8[a]	7.7[a]
Standard Error	0.18	0.18	0.08

[a-c] Means within a column, for each concentration of blueberry, with the same letter are not significantly different ($p>0.05$)
*Hedonic scale was based on a 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely
Sample = milliliters of whey and grams of blueberries

Consumer Acceptability (Sensory Center)

Three replications of consumer taste panels were performed on 3 separate days using the beverage formulations chosen from the preliminary research team panel. The 150mL whey-30g blueberries treatment was liked ($p<0.05$) more than all other treatments aside from the 125mL whey-30g blueberries treatment. This reveals that blueberries concentration appeared to be the factor that was influencing acceptability. In addition, it appeared that data from the pre-selected panel was not synonymous with data from the consumer panel. Acceptability of appearance was different ($p<0.05$) among samples. Scores increased from 5.9 to 6.8 as concentrations of whole blueberries increased. Results for flavor acceptability were also observed; samples were rated between 5.9 and 6.7. In addition, appealing flavors seemed to be enhanced with larger concentrations of whole blueberries. Results indicate that beverages were liked slightly to liked moderately with regards to overall acceptability with scores between 5.8 and 6.7 (Table 6).

Table 6. Effects of varying concentrations of whey within a fruit slush on consumer acceptability of appearance, flavor and overall acceptability determined by using consumer panels (n = 156).

Sample (mL whey – g blueberry)	Appearance Acceptability*	Flavor Acceptability*	Overall Acceptability*
50mL – 10.0g	6.1^{c}	5.9^{bc}	6.0^{cd}
125mL – 10.0g	5.9^{c}	5.7^{c}	5.8^{d}
125mL – 20.0g	6.6^{ab}	6.3^{ab}	6.3^{bc}
150mL – 20.0g	6.5^{b}	6.2^{abc}	6.2^{bc}
125mL – 30.0g	6.8^{ab}	6.7^{a}	6.7^{a}
150mL – 30.0g	6.8^{a}	6.4^{ab}	6.5^{ab}
Standard Error	0.09	0.16	0.12

$^{a-d}$ Means within a column with the same letter are not significantly different ($p>0.05$)
*Hedonic scale was based on a 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely
Sample = milliliters of whey and grams of blueberries

Cluster Analysis (Sensory Center)

A dendrogram (Table 7) was used to group Sensory Center consumers (n=156) into 4 cluster segments based on their preference of the beverage formulations. Cluster 1, differed ($p<0.05$) in their preference of formulations, and made up 25% of the consumer panel. For Cluster 1 acceptability scores, the beverage containing 125mL whey and 30g blueberries was slightly acceptable with a mean score of 6.0. This treatment is more acceptable ($p<0.05$) than all treatments but the 125mL whey-20g blueberries and 125mL whey-30g blueberries treatments. All samples in this cluster were in the neither like nor dislike to like slightly range. The beverage rated the lowest (mean score=4.9) contained 125mL whey and 10g blueberries and was liked less ($p<0.05$) than the 125-20, 125-30 and 150-30 treatments (Table 6). This indicates that these consumers preferred a higher amount of blueberries in the beverage. Cluster 2 contained 34.6% of the panelists. It was evident that these panelists liked all the beverage formulations, with mean scores (7.2 to 7.8) that were between like moderately and like very much. However, the beverage formulations that maximized blueberry concentration (125mL whey-30g blueberries and 150mL whey-30g blueberries) were rated higher ($p<0.05$) in acceptability than other treatments (Table 6). Cluster 3 contained 33.3% of panelists. Their mean scores for the beverage formulations were between 6.1 and 6.8, like slightly to like moderately. The sample containing 125mL whey and 10g blueberries was liked less ($p<0.05$) than the other beverages and had the lowest score of 5.7 (Table 7). Results indicate that a certain amount of blueberry flavor is necessary to increase consumer acceptability in this cluster. Cluster 4 contained 7.1% of the panelists. This group did not like any of the beverages regardless of ingredient formulation. Their mean scores were between 3.3 and 4.6 (Table 7). Many consumers dislike the flavor that whey imparts into products. These findings were similar to a study done by Childs et al. (2007) in which it was observed that consumer acceptability of whey-based beverages were unacceptable to consumers with hedonic scores between 3.0 and 4.4. According to Table 6, beverage formulation samples containing 125mL whey and 30g blueberries and 150mL whey and 30g blueberries had the highest numerical values of overall acceptability, 6.7 and 6.5, respectively. These beverage samples were chosen for further consumer testing since they maximized whey usage without negatively affecting consumer acceptability.

Table 7. Mean hedonic scores for overall consumer acceptability of whey-fruit slush samples with varying concentrations of whey and blueberries according to different clusters of consumer segments.

Cluster	Panelist (%)	Sample (mL-g) 50-10	Sample (mL-g) 125-10	Sample (mL-g) 125-20	Sample (mL-g) 150-20	Sample (mL-g) 125-30	Sample (mL-g) 150-30
1	25.0	5.3[bc]	4.9[c]	5.7[ab]	5.0[c]	6.0[a]	5.6[ab]
2	34.6	7.2[b]	7.2[b]	7.5[b]	7.4[b]	7.8[a]	7.8[a]
3	33.3	6.1[c]	5.7[d]	6.2[c]	6.6[ab]	6.8[a]	6.4[bc]
4	7.1	3.6[ab]	3.9[b]	3.5[ab]	3.3[ab]	4.3[ab]	4.6[a]

[a-d]Means within a row with the same letter are not significantly different ($p>0.05$)
Hedonic scale was based on a 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely
Sample (mL-g) = milliliters of whey and grams of blueberries

Consumer Acceptability (Fitness Center)

Additional consumer testing was conducted at the Joe Frank Sanderson Center on the Mississippi State University campus that houses the University Fitness Center that is used by students, staff, and faculty. Table 8 shows the beverage formulations and the mean consumer responses from the panels that were conducted at the Fitness Center. This location was chosen to conduct additional consumer panels because potential panelists that exercise at the fitness center are more likely to have interest in sport type beverages that include whey protein. According to consumer response, no difference ($p<0.05$) was detected between beverage formulations. Each beverage was rated for appearance, flavor, and overall acceptability with average ratings between 6.4 and 6.7 which gave the conclusion that these beverage formulations (125mL whey-30g blueberries and 150mL whey-30g blueberries) were acceptable to consumers.

Table 8. Effects of varying concentrations of whey within a fruit slush on consumer acceptability of appearance, flavor and overall acceptability determined by using consumer panels (n = 104).

Sample (mL whey – g blueberry)	Appearance Acceptability*	Flavor Acceptability*	Overall Acceptability*
125mL - 30.0g	6.5[a]	6.5[a]	6.7[a]
150mL - 30.0g	6.4[a]	6.6[a]	6.6[a]
Standard Error	0.18	0.40	0.30

[a]Means within a column with the same letter are not significantly different (p>0.05)
*Hedonic scale was based on a 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely).
Sample = milliliters of whey and grams of blueberries

Cluster Analysis (Fitness Center)

A dendrogram was used to group panelists from the Fitness Center into 6 clusters according to preferred responses. Cluster 1 contained 21.2% of panelists. These panelists slightly liked (5.9–6.5) both slush samples but preferred the beverage sample containing 150mL whey-30g blueberries (Table 9). These panelists may have liked the flavor attributes that were imparted by increasing whey content. The largest group of panelists (33.7%) was in Cluster 2. This group showed no difference ($p<0.05$) in preference between beverages. This group liked each beverage formulation very much and scores were 7.7 and 7.9 for 125mL whey-30g blueberries and 150mL whey-30g blueberries, respectively. This cluster is probably the consumer group that would be most likely to purchase and consume this product. In addition, this cluster of consumers may have not only liked the beverage samples for taste, but may have been more health conscious of their active lifestyles and ignored specific flavor attributes whether they were bad or good. A similar study was performed by Temelli and others, where they too, targeted consumers at a local fitness center thought to have an active lifestyle. They evaluated orange-flavored beverages with whey protein isolates using consumer taste panels. They observed a high percentage of the panelists scored 7.0 or higher on all beverage formulations giving them the conclusion that consumers with active lifestyles found whey beverage formulation products acceptable (Tamelli et al 2004).

Cluster 3, 26% of panelists, liked the beverage formulations between slightly (6.7) and very much (7.1). Consumers preferred ($p<0.05$) 125mL whey-30g blueberries over the 150mL whey-30g blueberries treatment. This reveals that this cluster may have not liked the flavor change due to increased whey concentration but still liked the product in general. Cluster 4, 10.6% of panelists, slightly disliked (4.2) the beverage sample with 125mL whey-30g blueberries and neither liked nor disliked to liked slightly (5.5) the beverage sample with 150mL whey-30g blueberries. These consumers would not likely be purchasers of this product. For Cluster 5 (7.7% of consumer panelists), there was a major difference between beverage sample preferences. The treatment with 125mL whey-30g blueberries was liked between moderately (7.6) and very much and preferred ($p<0.05$) over the sample with 150mL whey-30g blueberries, which was disliked slightly (4.0). This indicates that the increasing whey concentration had a major impact on overall consumer acceptability. The smallest group of consumer panelists (1.0%), cluster 6, disliked both beverage formulations extremely despite varying concentrations of whey.

Table 9. Mean hedonic scores for overall consumer acceptability of whey-fruit slush samples with the two highest concentrations of whey, according to different clusters of consumer segments in the Fitness Center.

Cluster	Panelists (%)	Sample 125mL whey-30g blueberries Hedonic scores*	Sample 150mL whey-30 g blueberries Hedonic scores*
1	21.2	5.9^{a}	6.5^{b}
2	33.7	7.7^{a}	7.9^{a}
3	26.0	7.1^{a}	6.7^{b}
4	10.6	4.2^{b}	5.5^{a}
5	7.7	7.6^{a}	4.0^{b}
6	1.0	1.0^{a}	1.0^{a}

$^{a-b}$Means within a row with the same letter are not significantly different (p>0.05)
*Hedonic scale was based on a 9-point scale (1 = dislike extremely, 5 = neither like nor dislike, and 9 = like extremely).

Chemical Analyses

Since there were no differences between beverage formulations, both were used for chemical analyses of optical absorbance, antioxidant activity percentage, total phenolics, pH measurement and proximate analysis were determined for the 125mL whey-30g blueberries and 150mL whey-30g blueberries treatments. These treatments maximized whey concentration without negatively affecting consumer acceptability. No difference (p>0.05) was detected between formulations. Antioxidant activity was present in both formulations (Table 10). For beverage sample 125mL whey-30g blueberries, antioxidant activity was at 0.24 optical absorption which converts to 44.3% and for beverage sample 150mL whey-30g blueberries; antioxidant activity was at 0.25 optical absorption which converts to 41.3%, which means the absorbance of DPPH acting as a free radical was decreased from .431 due to the presence of antioxidants. These values indicate that the presence of antioxidants would provide health benefits after consumption. Comparing these figures to Lohachoompol and others (2004), percentage of antioxidant activity for frozen blueberries that was stored for 1 month was approximately 60% and frozen blueberries stored for 3 months was approximately 30%. They concluded even though there was a reduction in antioxidant activity, there was no difference in content from fresh blueberries (Lohachoompol et al. 2004). In addition to blueberries contributing antioxidant activity, the cherry concentrate may have also contributed to antioxidant activity.

Total phenolics within the beverage formulations had minimal nutritional added value when compared to fresh blueberries and cherries. The 125mL whey-30g blueberries treatment had 0.25mg/mL and the 150mL whey-30g blueberries treatment had 0.30mg/mL. Comparing these figures to Chaovanalikit and Wrolstad (2004), who determined total phenolics levels directly from the skins of Montmorency cherries at 4.07mg/g, the total phenolics capacity for the slush beverages, were at much lower levels. However, only 10mL of Montmorency cherry concentrate was used in a 200mL slush beverage and upon testing for total phenolics, a further dilution scheme was done.

The pH was measured to estimate how safe the beverage samples were upon 48-hour storage. The 125mL whey-30g blueberries and 150mL whey-30g blueberry beverages had pH levels of 4.18 and 4.24, respectively. Therefore, both samples had safe pH level, which is understood to be below 4.6 before the possible growth of *Clostridium botulinum* (Lund et al. 1990). In addition, Beecher and others note astringent type flavors were produced when pH decreased to 3.4–2.6, which produces undesirable off-flavors and limited acceptance among consumers (Beecher et al. 2008).

Table 10. Chemical analysis observation using the two whey-fruit slush samples that were most accepted by consumers for antioxidant activity, pH and total phenolics.

Sample (mL-g)	O. A.	A. A. (%)	pH	Total Phenolics
125mL - 30.0g	.240	44.3	4.18	.25 mg/mL
150mL - 30.0g	.253	41.3	4.24	.30 mg/mL

O. A. = optical absorbance
A. A. (%) = percentage of antioxidant activity
Sample (mL-g) = milliliters of whey and grams of blueberries

Proximate Analysis

Proximate analysis (Table 11) was determined for the beverage formulations with the two highest concentrations of whey and the highest concentration of whole blueberries (125mL whey and 30g blueberries, 150mL whey and 30g blueberries) since their sensory acceptability scores were equal to or greater than the other treatments. Minimal differences existed between the formulations (Table 2). Each of the two formulations was predominantly made up of moisture due to the usage of ice and evaporated whey which had a ratio of 25-30% solids to 70-75% water. The protein content was minimal at approximately 1%. Carbohydrate content was moderately higher due to the presence of lactose within the whey and other simple sugars from the blueberries and cherry concentrate, but had limited impact on the nutritional value of the overall product. The alternative sweetener, sucralose (Splenda®) was used to sweeten the products which kept the kcal low. Within a 100g mixture, there were only 34 kcal in the beverage formulation with 125mL of whey and only 38 kcal in the beverage formulation with 150mL of whey.

According to these results, kcal slightly increased when concentration of whey increased which is due to the sugars derived from lactose present in whey. According to Frank and others (2008), this issue has been negligible due to an unspecified amount of calories contributed from maltodextrin, which is understood to be mixed with sucralose as a bulking agent. Other data notes high intensity sweeteners such as sucralose can be up to 600 times sweeter than that of sucrose, however may give undesirable aftertastes that can limit application (Zhao and Tepper 2006).

Table 11. Proximate analysis results of the two most preferred whey-fruit slush products.

Proximate Analysis Determinations	Sample 125mL whey – 30.0g blueberries	Sample 150mL whey – 30.0g blueberries
Moisture (%)	91.2	90.0
Ash (%)	0.4	0.4
Crude Protein (%)	1.0	0.9
Crude Fat (%)	0.3	0.5
Crude Fiber (%)	0.2	0.6
Carbohydrates (%)	6.9	7.6
Energy (kcal)	34.3 kcal/100g	38.5 kcal/100g

<u>Survey Results</u>

Consumers' personal preferences regarding whey-fruit slush beverages were investigated in conjunction with the consumer sensory testing through the use of a survey. Consumers were asked, "Overall do you think this whey-fruit slush is a healthy food item?" The majority (77.3%) of the Sensory Center consumer panelists indicated that the beverages were healthy, 20% of the panelists were unsure of the products health value and only 2.7% of panelists indicated that it had no health value. Consumers at the Fitness Center had somewhat similar responses to the Sensory Center consumers; 64.1% stated that the beverage products were healthy, 33.0% were unsure and almost identical to Sensory Center consumers, 2.9% felt the beverage products were not healthy (Figure 1). This left the assumption that the majority of consumers had knowledge that whey and blueberries enhance the nutrition of a variety of food products, including whey-fruit slush products.

Consumer panelists were asked about their willingness to purchase the whey-blueberry beverages they evaluated. At the Sensory Center and Fitness Center consumers, 46.6% and 46.0% reported they would be willing to purchase the products. Approximately 20% of consumers in both groups indicated they would not purchase the products, and 33.3% of Sensory Center consumer panelists and 33.9% of Fitness Center panelists were unsure if they would purchase this product type (Figure 2).

Consumer panelists were asked their willingness to purchase the whey-fruit slush products in place of other beverage products such as cola beverages, fruit juices, fruit smoothies, and sports drinks. The largest percentages (42.7% both Sensory Center and Fitness Center groups) felt somewhat likely they would purchase the products over other beverage products. Additionally, consumer panelists that were very likely to purchase this beverage product over other beverage products included 11.3% of the Sensory Center consumers and 17.5% of the Fitness Center consumers (Figure 3).

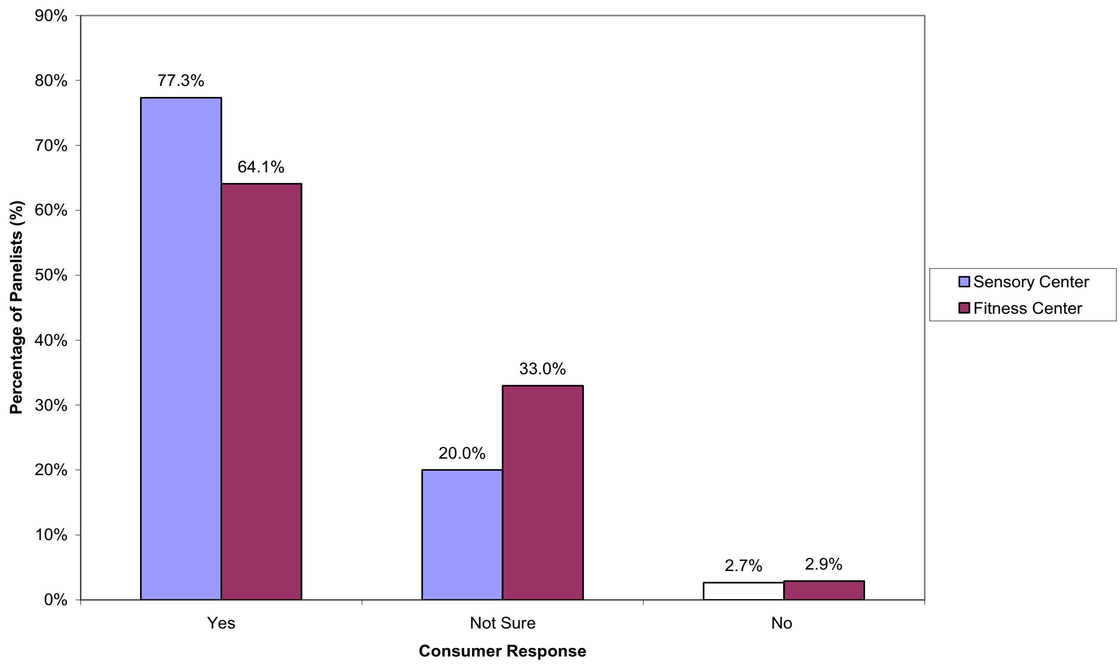

Figure 1. Percentage of panelists that perceived that the whey-fruit slush is a healthy product

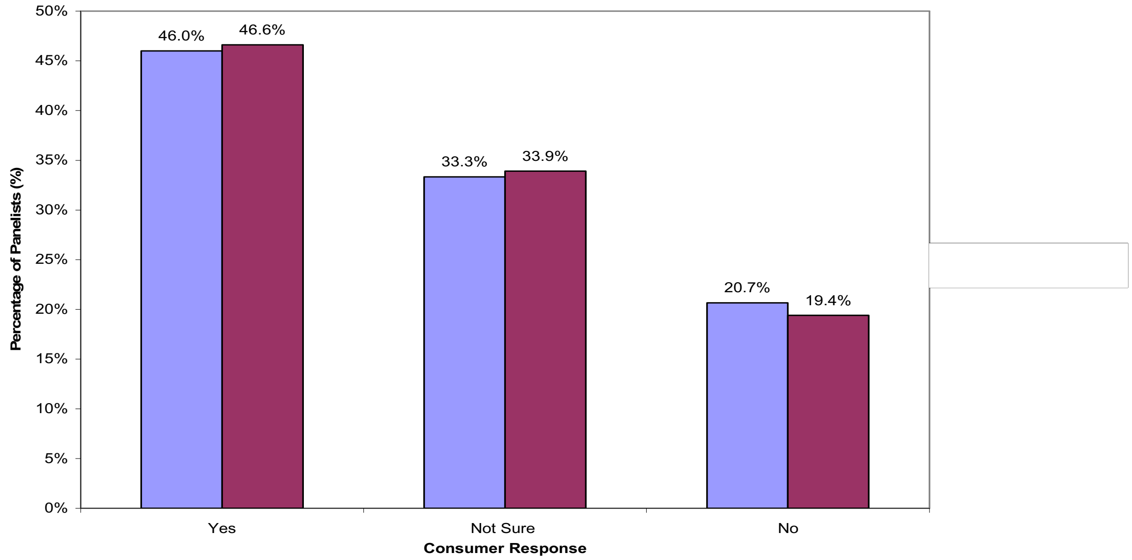

Figure 2. Consumer willingness to purchase whey-fruit slush products

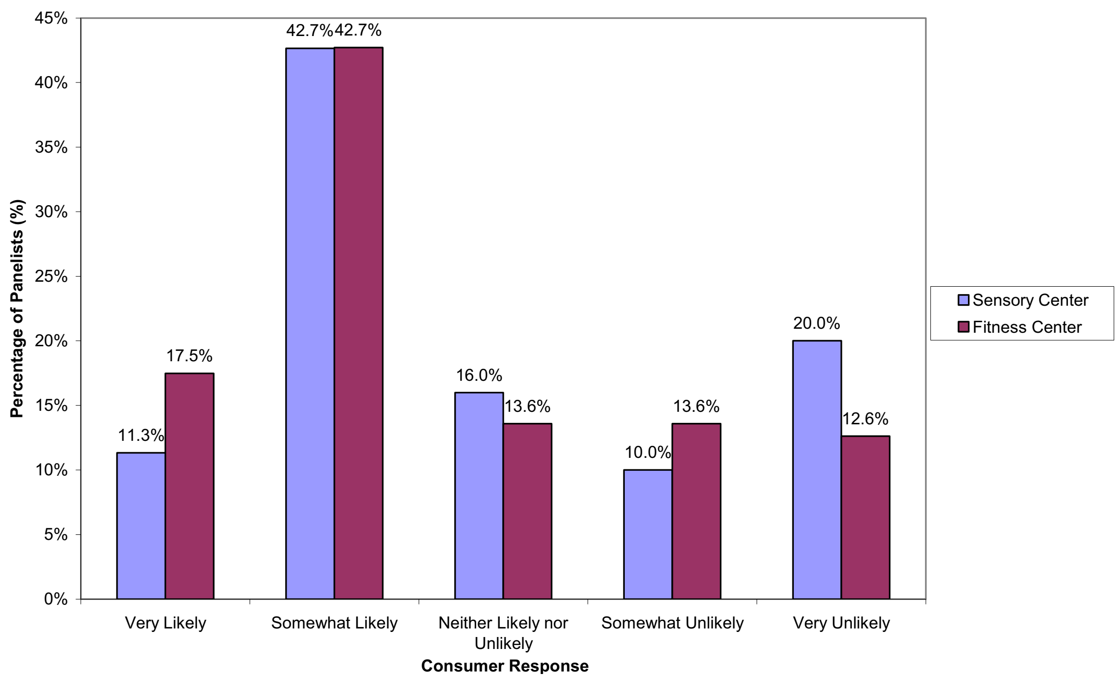

Figure 3. The likelihood that panelists are willing to purchase the whey-fruit slush product over other beverage products

Overall, 54.0% of the Sensory Center consumers and 60.2% of the Fitness Center consumers indicated they were very likely or somewhat likely to purchase the whey-fruit beverages in place of other beverages. Observing these numerical percentages, the majority of consumers may have appreciated the perceived nutritional added value of this product. Also, the consumer panelists at the Fitness Center may be more health conscious of their dietary intake. In comparison, 16.0% of the Sensory Center consumer panelists and 13.6% of the Fitness Center consumer panelists did not know if they would be willing to purchase this beverage product over other beverage products. Thirty (Sensory Center) and 26.2% of panelists (Fitness Center) indicated that they were somewhat unlikely and very unlikely to purchase the whey-fruit products in place of other beverage products (Figure 3).

Consumer panelists were asked how often they consume fruit slush products. A majority of Sensory Center consumer panelists (59.3%) and a majority of Fitness Center consumer panelists (45.6%) responded that they consume fruit-slush products rarely, less than once a month. This could be due to the lack of locations that supply such products due to unpopularity or limitation of availability. Consumers that indicated that they consume fruit slush products about once a month included 26.7% of Sensory Center panelists and 35.9% of Fitness Center panelists. A large difference was observed between Sensory Center consumer panelists at only 1.3% and Fitness Center consumer panelists at 26.2% for consuming fruit slush products once or twice a week. Perhaps people performing increased physical activity have a slush product as a refresher or thirst quencher more often than the other consumers. The remaining consumer panelists that may consume slush products often or very often, three to seven times a week, included

0.6% of Sensory Center consumer panelists and 5.8% of Fitness Center consumer panelists. Consuming a slush product on a daily basis had the least number of responses (Figure 4).

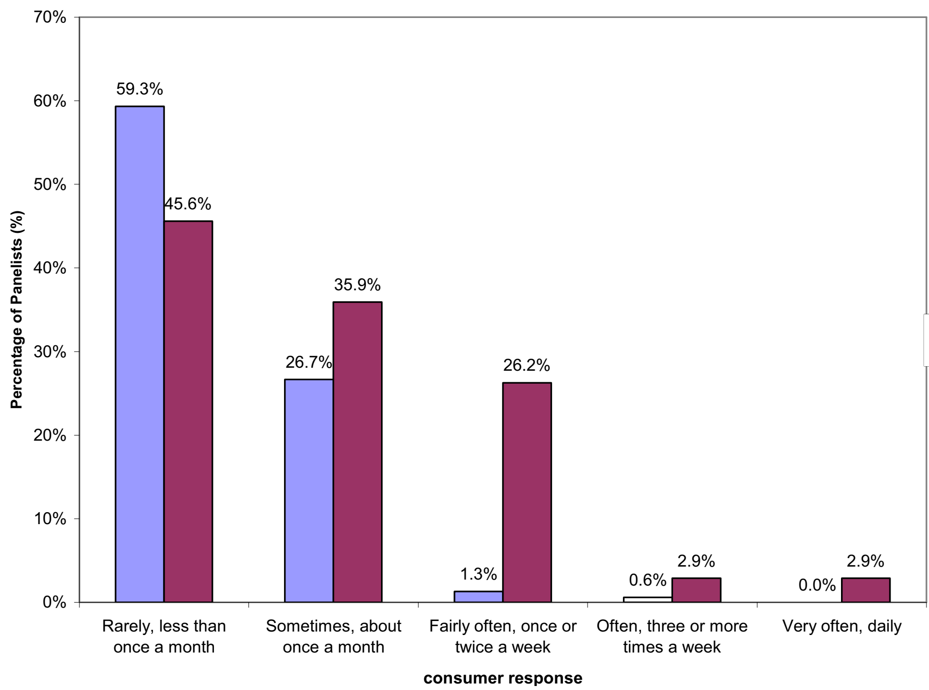

Figure 4. Consumer purchases of fruit slush products

Panelists were asked when they would most likely consume the products they were sampling A majority of Sensory Center consumer panelists (72.8%) responded as an afternoon snack and a majority of Fitness Center consumer panelists (54.3%) responded the same. Only 1.8% of Sensory Center consumer panelists and 10.6% of Fitness Center consumer panelists preferred the whey-fruit slush products with breakfast (Figure 5). This could possibly be due to panelists at the Fitness Center perceiving that this beverage product may be a good source of protein due to the whey or energy due to nutritional added value, and may want to replace it from a breakfast meal.

Few consumers indicated they were likely to consume the whey beverage at lunch or dinner; 0.3% of Sensory Center consumer panelists and 0.9% of Fitness Center consumer panelists indicated they would consume such a product at dinner. Likewise, only 0.3% of Sensory Center consumer panelists and 5.8% of Fitness Center consumer panelists stated that they would consume this whey-fruit slush product at lunch. Consumer panelists indicating they were not likely to consume the product included 11.6% of Sensory Center consumer panelists and 9.7% of Fitness Center consumer panelists (Figure 5).

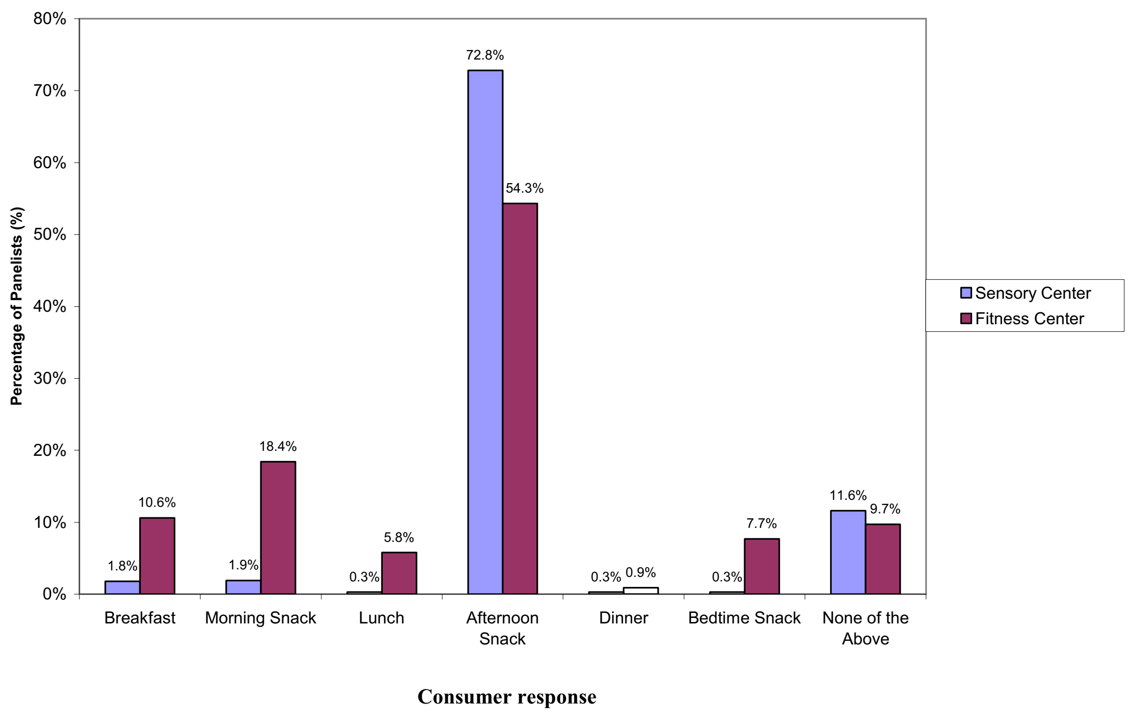

Figure 5. Consumer preference for consumption

Demographics of the consumer panelists indicated that 44.6% of the Sensory Center group and 58.2% of the Fitness Center group were male, and 55.4% of Sensory Center consumer panelists and 41.8% of fitness Center consumer panelists were female. (Table 12). Consumers reported their age and the largest age group included 59.2% of the Fitness Center consumer panelists who were 18–23 years old. The percentage of Sensory Center consumer panelists who were 18–23 years old was 28.6%. Those aged 24-29 included 21.3% of Sensory Center consumer panelists and 26.2% of Fitness Center panelists; 10.0% of Sensory Center consumer panelists and 6.7% of Fitness Center consumer panelists were the ages of 30-35 years, 39.9% of Sensory Center consumer panelists and 7.7% of Fitness Center consumer panelists were 35 years of age and older. There were no associations between age and consumer preference of the whey-fruit slush products. Similarly, there was no effect of gender on preference regarding willingness to purchase whey-fruit products or consumer acceptability of whey-fruit slush products.

Table 12. Gender and age groups of participating consumers from the Sensory Center and Fitness Center.

Demographic Variables of Participants	Sensory Center (n=156) (%)	Fitness Center (n=104) (%)
Gender:		
Male	44.6	58.2
Female	55.4	41.8
Age Group (years):		
18–23	28.6	59.2
24–29	21.3	26.2
30–35	10.0	6.7
35–40	9.3	1.9
41 and older	30.6	5.8

CHAPTER V

CONCLUSIONS

Many studies have shown that cheese whey has superior nutritional value and mechanisms that promote human health. Utilization of cheese whey in food product development has promising applications and benefits that contribute to value added food research. Despite literature reporting that whey-based beverages have astringent flavors and do not meet consumer expectations, whey-fruit slush products may be an acceptable product with positive health benefits if formulated with slight modifications. Even though fresh whey is unstable and carries undesirable flavors and aromas, this study has shown that these negative attributes can be minimized when properly formulated.

Understanding that whey has strong off-flavors, formulations need other contributing factors to obtain flavor balance. This was accomplished by adding additional ingredients that have strong contributing factors of flavor such as blueberries and cherry concentrate, which have sour attributes. Sucralose, marketed as Splenda®, is 300 to 600 times sweeter than sucrose was a contributing factor to mask the off-flavor of whey and intensify the sweetening effect at low concentrations. These added ingredients in combination with dilution schemes were utilized for additional masking in the prevention of off-flavors that could over-power the final product.

In this study, the formulated whey-fruit slush (150mL whey-30g blueberries) which maximized whey usage was considered acceptable by the majority of participating consumers. Statistical data showed that most consumers slightly and moderately liked the beverage schemes with a small percentage that found it unacceptable. Considering these findings, further modifications of adding ingredients, such as more fruit and/or sugar may persuade consumers to find the whey-fruit slush beverage more acceptable. Since the final product was slightly unstable due to separation of ice from liquid, the adding of certain stabilizers and/or methoxyl pectins (Koffi et al. 2005; Parker et al. 1993) and shelf life studies should be considered in future studies. In addition, to increase the intensity of fruit flavor, the usage of citric acid could be used and the adding of additional sweetening factors such as honey. However, making these modifications will make it a necessity to continue sensory evaluation tests to determine the impact of these additional variables on consumer acceptance.

Whey is a valuable resource of cheese production and should be utilized. Further research methods and utilization of cost-effective processes need to be considered to avoid large concentrations of whey being discarded. Since the demand of whey has increased in recent years (Beecher et al. 2008), application usages have to be further enhanced to meet a vast market of consumers who demand products that are both healthy and have great flavor.

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APPENDIX A

INSTITUTIONAL REVIEW BOARD (IRB) APPROVAL LETTER

March 26, 2008

Reginald Johnson 110 Lynn
Lane Apt 31A Starkville. MS
39759

RE: IRB Study #08-089: Acceptability of whey-fruit products

Dear Mr. Johnson:

The above referenced project was reviewed and approved via administrative review on 3/26/2008 in accordance with 45 CFR 46.101(b)(6). Continuing review is not necessary for this project. However, any modification to the project must be reviewed and approved by the IRB prior to implementation. Any failure to adhere to the approved protocol could result in suspension or termination of your project. The IRB reserves the right, at anytime during the project period, to observe you and the additional researchers on this project.

Please refer to your IRB number (#08-089) when contacting our office regarding this application.

Thank you for your cooperation and good luck to you in conducting this research project. If you have questions or concerns, please contact irb@research.msstate.edu or 325-3294.

Sincerely,

Katherine Crowley
Assistant IRB Compliance Administrator

cc: Dr. Diane Tidwell
Dr. Wes Schilling

Office for Regulatory Compliance
R 0. Box 6223 • 70 Morgan Avenue • Mailstop 9563 • Mississippi State, MS 39762 • (662) 325-3294 • FAX (662) 325-8776

APPENDIX B

INFORMED CONSENT FORM

Informed Consent Form - WHEY-FRUIT SLUSH PRODUCTS
(You must be at least 18 years old to participate)

Title of Study: Acceptability of whey-fruit products
Study Site: Joe Frank Sanderson Center, Mississippi State University
Researchers & University affiliation: Mr. Reginald Johnson, Dr. Diane K. Tidwell, Dr. M. Wes Schilling, Dr. Patti C. Coggins, and Ms. Julie Wilson, all are affiliated with Mississippi State University
What is the purpose of this research project? To determine the acceptability of two different whey-fruit slush products with varying concentrations of whey and fruit within a slush form.
How will the research be conducted? You will be provided with 2 whey-fruit slush samples. Please taste them and record your responses on the provided score sheets.
Are there any risks or discomforts to me because of my participation? There are no anticipated risks or discomforts. A list of all ingredients will be provided to you to prevent a possible food allergy. You may discontinue your participation at any point.
Does participation in this research provide any benefits to others or myself? Yes. Valuable information will be obtained that will help the dairy industry, Mississippi State University, and consumers understand the effect of endpoint concentration of whey on product quality, acceptability, and yields.
Will this information be kept confidential? Yes. Only the researchers who designed this study will have access to this information. Also, please note that these records will be held by a state entity and therefore are subject to disclosure if required by law.
Who do I contact with research questions? If you should have any questions about this research project, please feel free to contact (Dr. Diane Tidwell) at 662-325-0239 or (Dr. M. Wes Schilling) at 662-325-2666. For additional information regarding your rights as a research subject, please feel free to contact the MSU Regulatory Compliance Office at 662-325-5220.
What do I do if I am injured at a result of this research?
In addition to reporting an injury to Dr. Diane Tidwell, 662-325-0239 or Dr. Wes Schilling, 662-325-2666, and to the Regulatory Compliance Office, 662-325-5220, you may be able to obtain limited compensation from the State of Mississippi if the injury was caused by the negligent act of a state employee where the damage is a result of an act for which payment may be made under §11-46-1, et seq. Mississippi Code Annotated 1972. To obtain a claim form, contact the University Police Department at MSU UNIVERSITY POLICE DEPARTMENT, Stone Building, Mississippi State, MS 39762, (662) 325-2121.
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What if I do not want to participate? Please understand that your participation is voluntary, your refusal to participate will involve no penalty or loss of benefits to which you are otherwise entitled, and you may discontinue your participation at any time without penalty or loss of benefits. Additionally, you may skip any portion of the taste evaluation process.
ALL INGREDIENTS INVOLVED IN MAKING THIS FOOD PRODUCT ARE APPROVED BY THE FOOD & DRUG ADMINISTRATION FOR CONSUMPTION ACCORDING TO THEIR REGULATIONS
You will be given a copy of this form for you records.

Participant Signature **Date**

Investigator Signature **Date**

APPENDIX C

SCORE SHEETS

PRELIMINARY TEAM ACCEPABILITY OF WHEY-FRUIT PRODUCTS

Samples: Whey-fruit Slush Products **Date:**

Please taste each whey-fruit slush sample provided. After tasting, if you do not wish to swallow the sample, you may expectorate it in the cup and rinse with the water provided.

Rate each sample in each of the three categories listed.

Each column will need one check mark if you choose to evaluate all samples.

372	571	APPEARANCE
		Like extremely
		Like very much
		Like moderately
		Like slightly
		Neither like nor dislike
		Dislike slightly
		Dislike moderately
		Dislike very much
		Dislike extremely

372	571	FLAVOR
		Like extremely
		Like very much
		Like moderately
		Like slightly
		Neither like nor dislike
		Dislike slightly
		Dislike moderately
		Dislike very much
		Dislike extremely

372	571	OVERALL ACCEPTABILITY
		Like extremely
		Like very much
		Like moderately
		Like slightly
		Neither like nor dislike
		Dislike slightly
		Dislike moderately
		Dislike very much
		Dislike extremely

CONSUMER ACCEPABILITY OF WHEY-FRUIT PRODUCTS

Samples: Whey-fruit Slush Products **Date:**

Please taste each whey-fruit slush sample provided. After tasting, if you do not wish to swallow the sample, you may expectorate it in the cup and rinse with the water provided.

Rate each sample in each of the three categories listed.

Each column will need one check mark if you choose to evaluate all samples

825	410	137	294	781	649	APPEARANCE
						Like extremely
						Like very much
						Like moderately
						Like slightly
						Neither like nor dislike
						Dislike slightly
						Dislike moderately
						Dislike very much
						Dislike extremely

825	410	137	294	781	649	FLAVOR
						Like extremely
						Like very much
						Like moderately
						Like slightly
						Neither like nor dislike
						Dislike slightly
						Dislike moderately
						Dislike very much
						Dislike extremely

825	410	137	294	781	649	OVERALL APPEARANCE
						Like extremely
						Like very much
						Like moderately
						Like slightly
						Neither like nor dislike
						Dislike slightly
						Dislike moderately
						Dislike very much
						Dislike extremely

APPENDIX D

SURVEY

<u>Acceptability of Whey-Fruit Slush Products</u>

Please answer the following by placing one checkmark for each item.

1. Overall, do you think this whey-fruit slush is a healthy food item?

   ___ Yes ___ Not Sure _____No

2. Would you be willing to purchase this product?

   ____ Yes ___ Not Sure ___No

3. Please indicate your willingness to purchase this product over other products such as colas beverages, fruit juices, fruit smoothies, sport drinks, etc.

   ____Very Likely
   ____Somewhat Likely
   ____Neither Likely nor Unlikely
   ____ Somewhat Unlikely
   ____ Very Unlikely

4. How often do you consume fruit-slush products?

   ____ Rarely, less than once a month
   ____ Sometimes, about once a month
   ____ Fairly often, once or twice a week
   ____ Often, three or more times a week
   ____ Very often, daily

5. When would you most likely consume this product?

   ____ Breakfast
   ___ Morning Snack
   ____ Lunch
   ___Afternoon Snack
   ____ Dinner
   ____ Bedtime Snack
   ___None of the Above

6. Please check your age group.

   ___18-23 ___ 24-29 ___30-35
   ___35-40 ___41 and older

7. What is your gender?

   ___Male _____Female

Thank you for your participation!