Comparison of composition, sensory, and volatile components of thirty-four percent whey protein and milk serum protein concentrates¹

J. Evans,* J. Zulewska,† M. Newbold,‡ M. A. Drake,* and D. M. Barbano‡²

*Department of Food Science, Bioprocessing, and Nutritional Sciences, Southeast Dairy Foods Research Center, North Carolina State University, Raleigh 27695

†Faculty of Food Sciences, University of Warmia and Mazury, Olsztyn, Poland

‡Northeast Dairy Foods Research Center, Department of Food Science, Cornell University, Ithaca, NY 14853

ABSTRACT

The objectives of this study were to identify and compare the composition, flavor, and volatile components of serum protein concentrate (SPC) and whey protein concentrate (WPC) containing about 34% protein made from the same milk to each other and to commercial 34% WPC from 6 different factories. The SPC and WPC were manufactured in triplicate with each pair of serum and traditional whey protein manufactured from the same lot of milk. At each replication, SPC and WPC were spray dried (SD) and freeze dried (FD) to determine the effect of the heat used in spray drying on sensory properties. A trained sensory panel documented the sensory profiles of rehydrated SD or FD powders. Volatile components were extracted by solid-phase microextraction (SPME) and solvent extraction followed by solvent-assisted flavor evaporation (SAFE) with gas chromatography-mass spectrometry and gas chromatography-olfactometry. Whey protein concentrates had higher fat content, calcium, and glycomacropeptide content than SPC. Color differences (Hunter L, a, b) were not evident between SPC and WPC powders, but when rehydrated, SPC solutions were clear, whereas WPC solutions were cloudy. No consistent differences were documented in sensory profiles of SD and FD SPC and WPC. The SD WPC had low but distinct buttery (diacetyl) and cardboard flavors, whereas the SD SPC did not. Sensory profiles of both rehydrated SD products were bland and lower in overall aroma and cardboard flavor compared with the commercial WPC. Twenty-nine aroma impact com-

pounds were identified in the SPC and WPC. Lipid and protein oxidation products were present in both products. The SPC and WPC manufactured in this study had lower total volatiles and lower concentrations of many lipid oxidation compounds when compared with commercial WPC. Our results suggest that when SPC and WPC are manufactured under controlled conditions in a similar manner from the same milk using the same ultrafiltration equipment, there are few sensory differences but distinct compositional and physical property differences that may influence functionality. Furthermore, flavor (sensory and instrumental) properties of both pilot-scale manufactured protein powders were different from commercial powders suggesting the role of other influencing factors (e.g., milk supply, processing equipment, sanitation).

Key words: whey protein, serum protein, flavor, microfiltration

INTRODUCTION

Whey proteins are highly functional and nutritious proteins used in a variety of products that are a by-product of cheese making. Whey proteins can be found in sports and nutrition bars and beverages, infant formula, dairy foods, meat, and other foods (National Dairy Council, 2008). Concentrated whey, whey powder, lactose, lactalbumin, whey protein fractions, whey protein concentrate (**WPC**), and whey protein isolate (**WPI**) are a few of the products formed from processing of liquid whey. The 3 most prominent proteins in whey from cheeses produced with rennet are β -LG, α -LA, and glycomacropeptide produced by the action of rennet on κ -CN. The most commonly used forms of whey in the food industry are whole whey (about 11 to 13% protein), WPC (34 to 89% protein), and WPI (>90% protein). Whey proteins have many functional characteristics such as high solubility, dispersibility, water binding, foaming, whipping, emulsification, gelation, and buffering power (Davis and Foegeding, 2007) and are used frequently in food applications (National

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²Corresponding author: dmb37@cornell.edu

Dairy Council, 2008). Spray drying is the most common method of drying and has been in use since the early 1900s (Masters, 1997). Whey proteins should ideally have a bland flavor to facilitate their application in foods (Drake et al., 2009), but flavor of these products is highly variable because of many factors, including the original whey source, processing, and storage (Mahajan et al., 2004; Carunchia Whetstine et al., 2005a; Gallardo-Escamilla et al., 2005; Wright et al., 2006, 2009). Off-flavors may carry through into ingredient applications and limit food applications (Drake, 2006; Drake et al., 2009; Wright et al., 2009).

Milk minus milk fat globules is called milk plasma; milk minus milk fat globules and casein micelles is called milk serum (Walstra et al., 1999). Milk serum proteins (SP) are soluble milk proteins present in milk outside of the colloidal dispersion of the much larger casein micelles (Walstra et al., 1999). They are present in molecular form or as very small aggregates with naturally bound counter ions and bound water. Soluble proteins can be separated from casein and removed from skim milk (Pierre et al., 1992; Le Berre and Daufin, 1996) by microfiltration (MF). The extent of removal of SP from skim milk by microfiltration is a function of concentration factor and extent of diafiltration (Nelson and Barbano, 2005a). Once removed from milk by MF, the milk soluble proteins can be concentrated to produce milk serum (i.e., soluble) protein concentrates (SPC). The milk soluble proteins are similar to the family of proteins that are found in cheese whey from cheese made with rennet and can be concentrated to make SPC, but there are some distinct differences between SPC and WPC. Two major differences are that SPC does not contain glycomacropeptide (GMP) from the action of rennet on κ -CN and SPC contains very little fat. Because of a lack of familiarity with the term "milk serum protein," these proteins have also been referred to as "native, virgin, and ideal" whey proteins. Because SPC have a different composition from WPC and are produced by a different process, they are not exposed to the enzymatic or chemical reactions of the cheese-making process that can lead to off-flavors. The reality is that the SPC products produced by MF of skim milk have nothing to do with cheese making or cheese whey. However, because of the familiarity of dairy industry with whey products it is useful to compare these products with respect to their flavor and functionality.

Recent research has demonstrated the feasibility of removal of SP from skim milk before Cheddar cheese making (Nelson and Barbano, 2005a,b). Nelson and Barbano (2005a) developed a combined MF and diafiltration with UF permeate that removed 95% of serum proteins from milk before Cheddar cheese making and subsequently demonstrated that serum protein

removal from fluid milk had no effect on the quality of Cheddar cheese (Nelson and Barbano, 2005b). Serum protein concentrates, because they are not subjected to a cheese-making procedure, are not exposed to as many processing techniques that may alter flavor; thus, SPC may have the potential to offer a blander flavor profile compared with WPC. To our knowledge, no studies have compared flavor of SPC and WPC. The objective of this research was to compare the composition and sensory properties of 34% WPC and 34% SPC made from the same milk. A second objective was to compare the composition and sensory properties of the SPC and WPC made in our study with those of commercial 34% WPC from 6 different companies.

MATERIALS AND METHODS

Experimental Design

One batch of whole, raw bovine milk (about 1,800 kg) was received from the Cornell University dairy farm (Ithaca, NY). The milk was divided into 2 portions. One portion of the milk was used for Cheddar cheese manufacture to produce 34% WPC. The other portion was centrifugally separated at 4°C into raw cream and raw skim, and the skim milk was used to produce 34% SPC. This was replicated 3 times with different batches of raw milk. The timeline for processing was as follows: raw whole milk was received on Monday and split into 2 portions. On Monday, one portion of the whole raw milk was pasteurized and used for Cheddar cheese making. The Cheddar whey was pasteurized and run through a cream separator and cooled to 4°C by the end of the day Monday. The other portion of raw milk was cold separated to produce raw skim milk. On Tuesday, the raw skim milk from Monday was pasteurized, cooled to 4°C, and stored; the separated pasteurized whey was ultrafiltered to produce a liquid 34% WPC that was cooled and split into 2 batches: one was freeze-dried (this material was immediately frozen in trays) and the other was held overnight at 4°C and spray dried on Wednesday. On Wednesday, the pasteurized skim was microfiltered to produce 65% SP reduced casein concentrate and MF permeate. The MF permeate was cooled to 4°C and held overnight. The characteristics of the 65% SP reduced native micellar casein concentrate will be reported in a separate publication. On Thursday, MF permeate was ultrafiltered to produce 34% liquid SPC that was cooled and split into 2 batches: one was freeze-dried (this material was immediately frozen in trays) and the other was held overnight at 4°C and spray dried on Friday. Commercial samples of 34% WPC were obtained from 6 different companies (2 lots per company) and analyzed under the same conditions for comparison to the samples produced in this study.

WPC Manufacture

Raw whole milk for Cheddar cheese production was pasteurized with a plate heat exchanger (model 080-S, AGC Engineering, Manassas, VA) at 72°C and a holding time of 16 s. The pasteurized milk was cooled to 31°C and weighed into a cheese vat (model DLHD8SSS, Kusel Equipment Company, Watertown, WI). If necessary, the milk was standardized to a 0.70 casein to fat ratio by cream addition. Samples of milk were taken for chemical and microbiological analysis before and after the standardization for Cheddar cheese making. The average fat and protein contents of the milk for cheese making were $3.44\% \pm 0.03$ and $2.98\% \pm 0.03$, respectively. The milk was inoculated with the starter culture including *Lactococcus lactis* ssp. *lactis* and *Lactococcus lactis* ssp. *cremoris* (911 DVS, Chr. Hansen Inc., Milwaukee, WI) at a rate of 0.1 g/kg. The milk was agitated for 5 min and allowed to ripen for 30 min. A small amount of Annatto food color (AFC WOS 550, Rhodia Inc., Madison, WI) was added (0.0033 mL/kg of milk) to provide a consistent color to the white Cheddar. The ripened milk, 31°C, was coagulated with double-strength chymosin (Chymax Extra, Chr. Hansen Inc.) for 30 min at a rate of 0.1 mL/kg of milk. The coagulum was cut with 1.6-cm wire knives, and the curd and whey was allowed to rest for 5 min. Then, the curd plus whey was gently stirred for 10 min without added heat. The temperature was increased gradually from 31 to 33°C over 15 min and then from 33 to 38°C over an additional 15 min. The curd was continuously stirred at 38°C until the target whey draining pH of 6.35 was attained. The whey was drained and immediately pasteurized using a plate heat exchanger (3 sections, regeneration, heating, and cooling; model 080-S, AGC Engineering) at 72°C for 16 s. The whey was cooled to 50°C at the exit of the pasteurizer and immediately processed with cream separator (model 619, DeLaval Inc., Kansas, MO) to reduce the fat content. The fat content of the whey before and after separation was $0.21\% \pm 0.02$ and $0.06\% \pm 0.01$, respectively. After separation, the whey was cooled to 4°C and held overnight at $\leq 4^\circ\text{C}$. The whey was mixed and sampled directly before and after cream separation. During cheese manufacturing, the pH of whey and cheese were measured with an electrode (model HA 405, Mettler Toledo, Columbus, OH) that was standardized at pH 6.97 and 4.03 at 38°C and kept immersed in 3 M KCl at 38°C between readings to keep its temperature equal to the temperature of the buffers and samples. All samples were at 35 to 38°C at the time of measurement.

On the next day, the separated whey was weighed into a vat and heated to 50°C using a plate heat exchanger (model A3, DeLaval Inc.). The separated Cheddar

cheese whey was fractionated using a pilot-scale UF system equipped with a polyethersulfone spiral wound membrane (model 3838, GEA Niro Inc., Hudson, WI; nominal separation cutoff: 10,000 Da, surface area: 6.8 m²) at 50°C. The retentate inlet pressure was 276 kPa and the retentate outlet pressure was 103 kPa with no back pressure on the permeate side.

Immediately before processing, the spiral wound UF membrane was given a short cleaning cycle. First, the soak solution (0.26% vol/vol, Ultrasil MP, Ecolab Inc., Food and Beverage Division, St. Paul, MN) was flushed from the system until the flush water was at neutral pH. The membrane was then washed for 20 min at 276 kPa inlet pressure and no permeate back pressure with a combination of Ultrasil 110 liquid alkaline membrane cleaner (0.40% vol/vol), and XY-12 liquid sanitizer (0.15% vol/vol; both Ecolab Inc.) diluted in 50°C reverse osmosis (RO) water at a pH of 11.0 to 11.4. After the wash cycle was completed, the membrane system was flushed with 50°C RO water until neutral pH was obtained. The membrane was cooled to $<24^\circ\text{C}$ and sanitized with a solution of Ultrasil 110 liquid alkaline membrane cleaner (0.40% vol/vol) and XY-12 liquid sanitizer (0.15% vol/vol), at pH 11.0 to 11.4 and a chlorine level of 150 to 180 ppm, and RO water. This solution was circulated through the membrane for 10 min at 276 kPa inlet pressure and no permeate back pressure. The membrane was then flushed with 50°C RO water to neutral pH and the clean water flux was determined by operating only the inlet pump with an inlet pressure of 172 kPa. The water flux (typically about 54 kg/m² per hour) was calculated based on the weight of permeate collected in 30 s and the total membrane surface area.

Approximately 573 kg of separated whey was heated to 50°C and processed with the UF system in batch recirculation mode. When starting the process, before directing the retentate back to the feed tank, approximately 10 to 15 L of liquid was collected and discarded to flush water out of the system. Then, the retentate was returned to the feed vat and the process continued until 34% protein concentration as a percentage of solids was reached. The flux was determined by weight every 15 min, and samples of permeate and retentate (sample taken from feed vat) were taken for analysis using an infrared milk analyzer (Lactoscope FTIR, Delta Instruments, Drachten, the Netherlands) to control the ratio of protein to lactose in the retentate. Ultrafiltration was continued until the measured protein content of the retentate was 41% of the protein plus lactose in the retentate. The total time of UF was approximately 110 min. The infrared milk analyzer was calibrated using modified milk samples as described by Kaylegian et al. (2006). After UF was complete, the UF retentate in the

feed vat was combined with the UF retentate drained from the dead volume of the UF system, mixed, and sampled. The final concentration factor was about $5.2\times$. The final liquid WPC was weighed and divided into 2 portions. One portion was frozen at -40°C in stainless steel freeze-dryer pans in preparation for freeze drying. The other portion was cooled to 4°C and held overnight for spray drying.

After whey processing, the UF system was cleaned as follows: first, the UF system was rinsed with two 70-L lots of 50°C RO water at 276 kPa retentate inlet pressure and 103 kPa retentate outlet pressure with no permeate back pressure. During the second rinse, the recirculation pump was turned off and the inlet pressure was adjusted to 172 kPa to determine the fouled water flux, which was typically about 38% of the initial clean water flux (21 vs. 53 kg/m^2 per hour). Next, the membrane was washed for 30 min with a combination of a 50°C aqueous Ultrasil 110 liquid alkaline membrane cleaner (0.40% vol/vol, pH 11.0 to 11.4) and Ultrasil 01 liquid high-surfactant cleaner (0.08% vol/vol; both Ecolab Inc.) at 276 kPa inlet pressure and 103 kPa retentate outlet pressure. These inlet and outlet pressures were used throughout all cleaning procedures. After the 30-min wash, the membrane was flushed to a neutral pH with 50°C RO water and then washed with a 50°C aqueous Ultrasil 76 liquid acid cleaner (0.30% vol/vol, pH 1.9 to 2.2; Ecolab Inc.) for 30 min followed by a flush to a neutral pH with 50°C RO water. The membrane was then washed for 30 min at 50°C with a combination of an aqueous Ultrasil 110 liquid alkaline membrane cleaner (0.40% vol/vol, pH 11.0 to 11.4) and XY-12 liquid sanitizer (0.15% vol/vol, chlorine 150 to 180 ppm) and flushed to a neutral pH with 50°C RO water. When the rinse water pH was neutral, the clean water flux (typically about 53 kg/m^2 per hour) was determined by operating only the feed pump with an inlet pressure of 172 kPa. After the clean water flux was determined, the membrane was cooled to $<24^{\circ}\text{C}$ with a 10-min recirculation (276 kPa inlet pressure) of a 24°C aqueous solution of Ultrasil 110 liquid alkaline membrane cleaner (0.40% vol/vol, pH 11.0 to 11.4) and XY-12 liquid sanitizer (0.15% vol/vol, 150 to 180 ppm chlorine). The membrane was then flushed with room temperature RO water to a neutral pH, followed by a 10-min recirculation of an aqueous storage solution of Ultrasil MP soak solution (0.26% vol/vol, pH 3.5 to 4.0; Ecolab Inc.) that remained in the system until the next processing run.

SPC Manufacture

Raw whole milk was separated in the Cornell University dairy plant at 4°C using a model 590 Air Tight

Centrifuge (DeLaval Co., Chicago, IL). The raw skim milk was pasteurized (1,380 kg/h) on the following day with a plate heat exchanger (3 sections, regeneration, heating, and cooling; model 080-S, AGC Engineering) at 72°C and a holding time of 16 s. The milk was cooled to 4°C and stored refrigerated overnight at $\leq 4^{\circ}\text{C}$. The skim milk (about 1,040 kg) was heated to 50°C with a DeLaval model A3 plate heat exchanger and microfiltered using a pilot-scale, uniform transmembrane pressure (UTP) MF system (Tetra Alcross MFS-7, Tetra-Pak Filtration Systems, Aarhus, Denmark) equipped with ceramic Membralox (EP1940GL0.1 μA , alumina, Pall Corp., East Hills, NY) membranes (pore diameter: 0.1 μm ; surface area: 1.7 m^2). The MF process was a continuous bleed and feed at a concentration factor of $3\times$ with retentate and permeate removal rates of 45 and 90 L/h , respectively, and a transmembrane pressure in the range of 24 to 28 kPa during the processing run. A more detailed description of the UTP system and process is provided in Zulewska et al. (2009). The total time of milk processing was about 8 h. The MF permeate was cooled to 4°C and held overnight.

On the following day, MF permeate (about 670 kg) was weighed into a vat, heated to 50°C using a DeLaval model A3 plate heat exchanger, and ultrafiltered. Before processing, the UF membrane was cleaned following the same procedure as mentioned previously for whey processing. The MF permeate was processed using a polyethersulfone spiral wound UF membrane (model 3838, GEA Niro Inc.) with a nominal pore size of 10,000 Da. The conditions and parameters of UF processing of MF permeate were the same as described above for whey processing. The total processing time was about 120 min at a concentration factor $6.5\times$. After producing the 34% SPC liquid concentrate, the UF system was cleaned as described above for 34% WPC processing. The fouled water flux after processing MF permeate was typically about 35% of the clean water flux (19 vs. 54 kg/m^2 per hour).

Spray Drying

The 34% WPC and 34% SPC were spray dried (SD) using a model 1 atomizer (Niro Atomizer Inc., Columbia, MD). The feed material (about 55 kg) was kept at or below 7°C . The spray dryer was equipped with an FU11 atomizer (Niro Atomizer Inc.) rotating at 23,000 rpm, and the feed rate was 16 kg/h . The inlet temperature was 200°C and the outlet temperature was 95°C . The powder from the first 10 min of the run was discarded. Thereafter, the dried product was collected and packaged every half hour. The total time of the drying run was approximately 3.5 h. The material for sensory examination was packaged in VWR

TraceClean 950-mL amber glass, wide-mouth jars with polytetrafluoroethylene-lined caps (VWR International, West Chester, PA). The powders for chemical and microbiological analysis were stored at 21°C in clear 84-mL plastic vials (Capitol Vial, Auburn, AL).

Freeze Drying

The liquid 34% WPC and 34% SPC were held at -40°C and freeze-dried (**FD**) in a model 101-SRC-5 freeze dryer (VirTis Company, Gardiner, NY). The frozen liquid for freeze-drying was transferred from the -40°C freezer to the freeze dryer where the shelves had been cooled to -30 to -40°C. When the vacuum reached approximately 100 μ m, the shelf temperature was increased to 30 to 35°C and maintained at that temperature throughout the drying. The time of freeze-drying was approximately 72 to 96 h. The dried material for sensory examination was packaged in VWR TraceClean 950-mL amber glass, wide-mouth jars with polytetrafluoroethylene-lined caps (VWR International). The dried material for chemical and microbiological analysis was stored at 21°C in 84-mL clear plastic vials (Capitol Vial).

Chemical Analyses

Milk for cheese making was analyzed using infrared spectrophotometer (Lactoscope FTIR, Delta Instruments) for fat content and true protein content (Kaylegian et al., 2006). The fat content of unseparated and separated whey was determined by ether extraction (AOAC, 2000; method 989.05, 33.2.26).

Fresh samples of the final liquid 34% WPC and liquid 34% SPC were analyzed for fat, TS, total N, and NPN content using ether extraction (AOAC, 2000; method 989.05; 33.2.26), forced air oven drying (AOAC, 2000; method 990.20; 33.2.44), Kjeldahl (AOAC, 2000; method 991.20; 33.2.11), and Kjeldahl (AOAC, 2000; method 991.21; 33.2.12), respectively. The GMP content (which is soluble in 12% TCA) of WPC was calculated as a difference in NPN content between WPC and SPC powders on a dry basis. The commercial 34% WPC samples were reconstituted to 10% solids, and the liquids were analyzed for fat and total N by the methods indicated above. The 34% WPC, 34% SPC, and 6 commercial powders were reconstituted to 10% solids: pH was measured with an electrode (model Electrolyte 9823, Mettler Toledo) that was standardized at pH 7.01 and 4.00 at room temperature (22°C). The TS content was measured by forced-air oven drying (AOAC, 2000; method 990.20; 33.2.44), and moisture content of the powders was calculated.

Contents of Ca, P, K, Mg, Na, and S were measured in duplicate by the North Carolina State University Analytical Services Laboratory (Raleigh) using a standard dry ash method with inductively coupled plasma optical emission spectroscopy (**ICP-OES**). For this analysis, 2.5 g of WPC or SPC was ashed overnight at 500°C in a muffle furnace, cooled, mixed with approximately 1.5 mL of distilled water and 4 mL of 6 *N* HCl (VWR, West Chester, PA), heated to dryness on a steam plate to remove silicates, transferred to a 50-mL volumetric flask with approximately 4 mL of 6 *N* HCl, and brought to volume with distilled water for ICP-OES analysis.

Microbiological Analyses

Samples of pasteurized skim milk for cheese making before and after standardization, pasteurized whey from cheese making, pasteurized skim milk for MF, liquid WPC, liquid SPC, and spray-dried and freeze-dried powders were taken for total bacterial and coliform counts (Wehr and Frank, 2004; methods 6.020 and 7.020, respectively).

Color Analysis of Powders

The Hunter L (lightness), a (red-green), and b (yellow-blue) values for the spray and freeze-dried powders were determined in duplicate with a MacBeth Color-Eye spectrophotometer (model 2020, Kollmorgen Instruments, Corp., Newburgh, NY) with Optiview software from the same company. Hunter values were computed from the diffuse reflectance data in the 360 to 740 nm range, at 20-nm intervals, based on illuminant A. The measurements were done at 23 to 25°C.

SDS-PAGE

The gels and the samples were prepared using the same approach as described in Zulewska et al. (2009) with the exception that the powder samples were reconstituted to 10% solids. The loading of the samples was 8.5 μ L for milk and 7 μ L for 34% SPC and WPC samples. Three slots were loaded on the same gel for WPC and SPC along with the milk sample within each replicate. The gels were scanned and analyzed with USB GS 800 Densitometer using Quantity One 1-D Analysis Software (Bio-Rad Laboratories Inc., Hercules, CA). The maximum optical density of the predominant protein in each sample was in the range of 1.0 to 1.4 optical density to avoid exceeding the linear range of response of the detection system for the analysis. For each lane, the background was adjusted separately using the rolling disk method of subtraction to obtain

a flat base on the pop-up trace. The line that defines each lane was adjusted using the lane tools functions (add/adjust anchors) in the software, so that the red lane line crossed each band at the center. The bands for all lanes were detected using the same detection settings that were chosen to detect the main bands in the milk sample. The adjust band function of the software was used with brackets to set the leading and trailing edge for each band as visually observed on the image of the gel, not based on the beginning and end of the peak in the pop-up trace. The width of the brackets within each lane was set for all bands in that lane to be slightly wider than the widest band in that lane. A reference milk sample was run on every gel as a qualitative and quantitative reference. The relative proportion of casein and serum protein in pasteurized milks from this study was determined by Kjeldahl and compared with the same total casein and serum protein values calculated by electrophoresis as a quality control tool. The estimate of casein as a percentage of true protein was within 1% between the 2 methods.

Sample Shipping and Storage

Liquid products were shipped on ice by overnight carrier to North Carolina State University (Raleigh) and dry products were shipped at ambient temperature. Subsamples (1 kg) were stored in glass jars at -80°C and analyzed within 3 mo of receipt. For all analyses, samples were reconstituted at 10% solids. Sensory analysis samples were reconstituted using deodorized water (prepared by boiling 4 L of distilled water until its volume was decreased by one-third). Instrumental analysis samples were reconstituted using HPLC-grade water (EMD Chemicals Inc.). Six commercial 34% WPC (less than 2 mo old) were also received on 2 separate occasions and reconstituted using the same methods above for comparison to pilot-plant 34% WPC and SPC.

Descriptive Sensory Analysis

Sensory testing was conducted in compliance with North Carolina State University Institutional Review Board for human subject approval. A trained sensory panel ($n = 10$; 7 females, 3 males, ages 22 to 37 yr) evaluated the flavor attributes of the reconstituted serum and whey proteins using a previously published lexicon for dried dairy ingredients (Drake et al., 2003). Each panelist had over 150 h of experience with descriptive analysis of dried dairy ingredients and additional training with WPC and SPC aroma and flavor. Consistent with Spectrum descriptive analysis training, panelists were presented with reference solutions of sweet, sour, salty, and bitter tastes to learn to use the

universal intensity scale (Meilgaard et al., 1999; Drake et al., 2003). Panelists then evaluated and discussed flavor attributes of rehydrated dairy ingredients with a focus on 80% WPC and 34% WPC using an established sensory language (Drake et al., 2003; Russell et al., 2006; Wright et al., 2009). Analysis of variance of data collected in preliminary sessions confirmed that the panel and the panelists could consistently identify and scale flavor attributes (data not shown). Attribute intensities were scaled using the 0 to 15 universal intensity scale characterized by the Spectrum descriptive analysis method (Meilgaard et al., 1999; Drake and Civille, 2003).

Reconstituted products (approximately 30 mL) were dispensed into lidded 58-mL soufflé cups with 3-digit codes and stored at 3°C overnight. Products were tempered to 20°C and served at this temperature with spring water and unsalted crackers for palate cleansing. Panelists evaluated each sample individually in booths in a positive-air-pressure room dedicated to sensory analysis. Each product replication was evaluated by each panelist in duplicate in a randomized balanced block design on separate occasions. Products were scored using paper ballots or computerized ballots using Compusense 5.0 release 4.8 (Compusense Inc., Guelph, Canada).

Volatile Compound Extraction

Solid-Phase Microextraction GC-MS of WPC and SPC Powders. The solid-phase microextraction (SPME) GC-MS was conducted using a modified method of Wright et al. (2006). Spray-dried and freeze-dried powders were reconstituted at 10% solids, with 10% NaCl and 10 μL of internal standard solution (2-methyl-3-heptanone in methanol at 81 ppm) in 20-mL autosampler vials with steel screw tops containing silicone septa faced in Teflon (Microliter Analytical, Sawanee, FL). Samples were injected using a CombiPal autosampler (CTC Analytics, Zwingen, Switzerland) attached to an Agilent 6890N GC with 5973 inert MSD (Agilent Technologies Inc., Santa Clara, CA). Samples were maintained at 5°C before fiber exposure. Samples were equilibrated at 40°C for 25 min before 30 min fiber exposure of a 1-cm DVB/CAR/PDMS fiber at 31 mm with 4-s pulsed agitation at 250 rpm. Fibers were injected for 5 min at a depth of 50 mm. The GC method used an initial temperature of 40°C for 3 min with a ramp rate of $10^{\circ}\text{C}/\text{min}$ to 250°C held for 5 min. The SPME fibers were introduced into the split/splitless injector at 250°C . An Rtx-5ms column (Rtx-5ms 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μm film thickness; Restek, Bellefonte, PA) was used for all analysis at a constant flow rate of 1 mL/min. Purge time was set at

1 min. The MS transfer line was maintained at 250°C with the Quad at 150°C and Source at 250°C.

Direct Solvent Extraction of WPC and SPC Powders. Solvent extraction was conducted using the modified methods of Carunchia-Whetstine et al. (2005b). One hundred grams of powder was divided into 4 Teflon bottles (Nalgene, Rochester, NY, capacity of 250 mL) with Tefzel closures (Nalgene). Five milliliters of HPLC water (EMD Chemicals Inc.) was added to each bottle, along with 20 μ L of internal standard at 81 ppm (2-methyl-3-heptanone, 2-methylpentanoic acid in methanol, Sigma Aldrich, Milwaukee, WI). Seventy-five milliliters of diethyl ether (EMD Chemicals Inc.) was added to each bottle. The bottles were then shaken for 30 min on a Roto mix (type 50800, Thermolyne, Dubuque, IA) at high speed and centrifuged at $1,459 \times g$ for 10 min to separate the solvent phase from the mixture. After centrifugation, the solvent phase containing the extracted volatile components was removed by pipette to an amber glass jar. This procedure was repeated twice with the addition of 40 mL of diethyl ether to each bottle each time. After the third round of solvent was removed, the bottles were centrifuged a fourth time and any remaining solvent was removed. The solvent extracted from the bottles was then concentrated to 150 mL using a Vigreux column placed inside a water bath at 40°C.

Solvent-Assisted Flavor Evaporation. Volatile compounds from serum and traditional whey protein extracts were distilled using solvent-assisted flavor evaporation (SAFE; Ace Glassware, Vineland, NJ). The assembly used was similar to that described by Engel et al. (1999). A rough pump/diffusion pump combination was used as the vacuum source. The SAFE apparatus was connected to a separate receiving tube and a waste tube, both submerged in liquid nitrogen. The distillation procedure was carried out over 2 h under vacuum (10^5 Torr). The liquid sample was poured into the SAFE apparatus and introduced drop wise into the vacuum until all of the liquid extract had been placed under vacuum conditions. The SAFE apparatus was kept at 40°C with a circulating water bath.

Phase Separation. Following SAFE, the distillate was concentrated under a nitrogen stream to 20 mL. The concentrated distillate was then washed twice with 3 mL of sodium bicarbonate (Fisher Scientific, Fairlawn, NJ), mixed thoroughly and the bottom (water phase) removed to a separate test tube. The concentrated solvent was then washed with 2 mL of saturated sodium chloride solution 3 times. Each time, the solution was mixed thoroughly and the water phase removed to the same test tube. The upper layer (ether) containing the neutral-basic (NB) fraction was collected, dried over anhydrous sodium sulfate (VWR International, West

Chester, PA) and concentrated to 0.5 mL under a gentle stream of nitrogen gas. Acidic (AC) volatiles were recovered by acidifying the bottom layer (water phase) with hydrochloric acid (18% w/vol; Sigma Aldrich) to a pH of 2 to 2.5 and extracting the sample 3 times with 15 mL of ethyl ether. The acidified extract was dried over anhydrous sodium sulfate before concentration to 0.5 mL under a nitrogen gas stream.

GC-MS of SAFE Extracts. An Agilent 6890N GC with 5973 inert mass selector detection (MSD; Agilent Technologies Inc.) was used to analyze NB solvent extracts. Separations were performed on a fused-silica capillary column (Rtx-5ms 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness (Restek, Bellefonte, PA). A carrier gas (helium) at a constant flow rate of 1 mL/min was used. The oven temperature was programmed from 40 to 200°C at a rate of 5°C per min with initial and final hold times of 5 min, respectively. Each extract (2 μ L) was injected in the splitless mode. Duplicate analyses were performed on each sample.

A Varian CP-3380 GC with Saturn 2000 inert MSD (Varian Inc., Palo Alto, CA) was used to analyze AC solvent extracts. Separations were performed on a fused-silica capillary column (Rtx-wax 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness; Restek). A carrier gas (helium) at a constant flow rate of 1 mL/min was used. The oven temperature was programmed from 40 to 200°C at a rate of 5°C per min with initial and final hold times of 5 min, respectively. Each extract (2 μ L) was injected in the splitless mode. Duplicate analyses were performed on each sample.

GC-Olfactometry. The GC-olfactometry (GC-O) analysis was performed on NB and AC extracts from SAFE using an HP 5890 series II gas chromatograph (Hewlett-Packard Co., Palo Alto, CA) equipped with a flame-ionization detector (FID), a sniffing port, and a split/splitless injector. Neutral/basic and AC fractions were analyzed from each solvent extraction. Two microliters were injected onto a polar capillary column (Rtx-wax 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness; Restek) and a nonpolar capillary column (Rtx-5ms 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness; Restek). Column effluent was split 1:1 between the FID and the sniffing port using deactivated fused-silica capillaries (1 m length $\times$ 0.25 mm i.d.). The GC oven temperature was programmed from 40 to 200°C at a rate of 10°C/min with an initial hold for 3 min and final hold of 20 min. The FID was maintained at a temperature of 300°C. The sniffing port was maintained at a temperature of 105°C. The sniffing port was supplied with humidified air at 30 mL/min. The post-peak intensity of aroma active compounds was evaluated (Grosch, 1993; van Ruth, 2001). Each extract was sniffed in duplicate by 2 experienced sniffers, each

Table 1. Mean ($n = 3$) composition (% by weight) of the serum protein concentrate (SPC) and whey protein concentrate (WPC) liquids after UF and before drying

Product	Composition ¹				
	TS	Fat	TN	NPN	TP
SPC	9.71	0.02 ^b	3.43	0.23 ^b	3.20 ^a
WPC	9.68	0.19 ^a	3.33	0.37 ^a	2.96 ^b
R ²	0.59	>0.99	0.87	>0.99	0.97
SE	0.068	0.003	0.026	0.003	0.023

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹TN = total nitrogen $\times 6.38$; NPN = nonprotein nitrogen $\times 6.38$; TP = true protein (TN – NPN).

with >50 h training on GC-O of dairy products on both polar and nonpolar columns.

Aroma Extract Dilution Analysis. The aroma extract dilution analysis (AEDA) was performed under the same conditions as those for post peak intensity GC-O. Representative samples of each of the pilot-plant products (SD WPC, SD SPC, FD WPC, FD SPC) were analyzed. The NB fractions were injected onto the DB-5ms (Rtx-5ms 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness; Restek) capillary column, and AC fractions were injected onto the DB-WAX (Rtx-wax 30 m length $\times$ 0.25 mm i.d. $\times$ 0.25 μ m film thickness; Restek) capillary column. Each sample was diluted stepwise at a ratio of 1:3 (vol/vol) with diethyl ether. Samples were evaluated by 2 experienced sniffers until no odors were detected. The greatest dilution in which a compound was sniffed was reported as the flavor dilution ($\log_3$ FD) factor (Grosch, 1993).

Volatile Compound Identification. Volatile compounds from SAFE and SPME were identified using the NIST 2005 library of spectra and comparison of spectra of authentic standards injected under identical conditions (NIST, 2005). Relative abundance of compounds was calculated using the calculated recovery of the internal standard concentration to determine relative concentrations of each compound. Retention indices were calculated (Van den Dool and Kratz, 1963) using an alkane series (Sigma Aldrich). For aroma active compound identity verification, retention index, and aroma properties were also compared with authentic standards injected under identical conditions.

Statistical Analyses

To determine if there were significant differences in UF flux, color, composition, or sensory between WPC and SPC, all data were analyzed by ANOVA using the Proc GLM procedures of SAS (SAS version 8.02, 1999–2001, SAS Institute Inc., Cary, NC). A SAS GLM model was used to determine if UF flux changed among treatments (WPC, SPC) with the time of processing run after the first 30 min of running. Time

was treated as a continuous variable in the split-plot ANOVA model. Distortion of the ANOVA by multicollinearity in the model was minimized by centering the time of running using a mathematical transformation (Glantz and Slinker, 2001). Time was transformed as follows: time transformed = running time – [(last time – first time)/2]. This transformation made the data set orthogonal with respect to time. The model was dependent variable (UF flux) = treatment + replicate + treatment $\times$ replicate + time + time $\times$ replicate + time $\times$ treatment + error, with treatment and replicate as category variables and time as a continuous variable. The GLM model for analysis of flux data at 60 min of UF and for the direct comparison of composition from WPC and SPC produced in this study was treatment (WPC, SPC) + replicate + error. The GLM model for analysis of sensory data for the direct comparison of composition from WPC and SPC produced in this study was treatment (WPC, SPC) + replicate + panelist + error. The GLM model for comparison of composition among samples produced in this study and the commercial powders was dependent variable = treatment (WPC, SPC, commercial powders 1, 2, 3, 4, 5, 6) + error. For descriptive sensory analysis the GLM

Figure 1. Mean ($n = 3$) UF flux during the production of 34% whey protein concentrate (WPC) and 34% serum protein concentrate (SPC) using a spiral-wound 10 kDa polyethersulfone membrane.

Table 2. Mean ($n = 3$) composition (% by weight) of spray-dried serum protein concentrate (SPC) and whey protein concentrate (WPC) calculated on a dry basis and pH

Product	Composition ¹					
	Fat	TN	NPN	TP	GMP	pH
SPC	0.25 ^b	35.29 ^a	2.37 ^b	32.92 ^a	0.00 ^b	6.74 ^a
WPC	1.93 ^a	34.38 ^b	3.82 ^a	30.56 ^b	1.45 ^a	6.44 ^b
R ²	>0.99	0.99	>0.99	>0.99	>0.99	0.99
SE	0.03	0.06	0.03	0.03	0.003	0.011

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹TN = total nitrogen $\times$ 6.38; NPN = nonprotein nitrogen $\times$ 6.38, includes GMP for WPC; TP = true protein (TN – NPN); for WPC this does not include GMP; GMP = glycomacropeptide, calculated as NPN of WPC minus NPN of SPC; pH of the reconstituted powders (10% wt/vol).

model was dependent variable = treatment (WPC, SPC, commercial powders 1, 2, 3, 4, 5, 6) + panelist + error. If the F-test for the model was significant (i.e., $P < 0.05$), then the least squares means were compared to determine if they were significantly different. Principal component analysis using the Proc Princomp command using the correlation matrix was also applied to sensory and volatile component data of products produced in this study and commercial powders to visualize how products were differentiated across sensory attributes or volatile components (SAS version 9.2, SAS Institute Inc.).

RESULTS AND DISCUSSION

Processing

The largest decrease in the UF flux occurred during the initial 30 min of the processing for both Cheddar cheese whey and MF permeate. During the initial 15 min of the UF run, the flux decreased at about 33 and 38% during WPC and SPC processing, respectively, with an additional 10% decrease during the subsequent 15 min (Figure 1). No difference in mean UF flux (after 60 min of processing) when processing whey (15.71 ± 0.23 kg/m² per hour) or MF permeate from skim milk (14.62 ± 0.74 kg/m² per hour) was detected ($P > 0.05$). Time-dependent differences ($P < 0.05$) in UF flux after 30 min of processing were detected when comparing processing MF permeate from skim milk and whey. The

mean flux during the short time of UF processing in this study was higher for WPC than for SPC (Figure 1). However, flux was decreasing at faster rate per hour for the WPC than for the SPC (i.e., significant time $\times$ treatment interaction; 8.4% of the variation in the ANOVA model was explained by time $\times$ treatment interaction). In our experiment after 105 min of UF processing, the slope of flux for WPC intersected the flux for SPC (Figure 1) and it can be expected that in a longer run the flux for WPC might be lower than that for SPC. All 34% WPC and 34% SPC SD and FD products conformed to the dry whey grade standards (ADPI, 2002) of <30,000 cfu/g for standard plate count and <10 cfu/g for coliforms. The actual maximum counts for our products were 3,300 cfu/g for standard plate count and <1 cfu/g for coliforms.

Composition

No significant difference in the TS content of SPC and WPC liquids was detected (Table 1). The fat content of liquid WPC (Table 1) was higher ($P < 0.05$) than the fat content of liquid SPC, because most of the fat left in the skim milk by the cream separator was retained in the MF retentate during MF processing of milk using a membrane with a pore size of 0.1 μ m, whereas the fat that was not removed from the whey by the cream separator was concentrated in the liquid WPC by the UF. The difference in fat content between SPC and WPC may influence the sensory and functional proper-

Table 3. Mean ($n = 3$) mineral composition (% by weight) of spray-dried serum protein concentrate (SPC) and whey protein concentrate (WPC) as calculated on a dry basis

Product	Ca	P	K	Mg	Na	S
SPC	0.44 ^b	0.56	1.89	0.12	0.43	0.57
WPC	0.55 ^a	0.63	1.93	0.12	0.45	0.52
R ²	0.99	0.95	0.81	0.48	0.84	0.91
SE	0.006	0.008	0.022	0.003	0.005	0.009

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

Table 4. Composition (% by weight) and pH of spray-dried ($n = 3$) serum protein concentrate (SPC), spray-dried ($n = 3$) whey protein concentrate (WPC) produced in this study, and 6 commercial ($n = 2$) 34% WPC on a dry basis

Source	Product	Moisture	Fat	Total N	pH ¹
Experimental	SPC	4.60 ^a	0.25 ^e	35.29 ^{cd}	6.74 ^a
Experimental	WPC	4.08 ^a	1.93 ^f	34.38 ^d	6.44 ^{bcd}
Commercial WPC	1	5.05 ^a	3.41 ^{ab}	39.08 ^a	6.13 ^d
	2	4.93 ^a	3.52 ^a	36.16 ^{bc}	6.42 ^{acd}
	3	4.86 ^a	3.23 ^{abc}	36.53 ^{bc}	6.28 ^{cd}
	4	5.84 ^a	2.81 ^{cd}	36.81 ^b	6.64 ^{ab}
	5	5.63 ^a	3.03 ^{bc}	35.61 ^{bcd}	6.68 ^{ab}
	6	5.08 ^a	2.50 ^d	36.29 ^{bc}	6.57 ^{abc}
R ²		0.65	0.98	0.89	0.73
SE: commercial		0.3745	0.1371	0.4312	0.1113
SE: experimental		0.3058	0.1119	0.3521	0.0909

^{a-f}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹pH of the reconstituted powders (10% wt/vol).

ties. Whey protein concentrate contained more ($P < 0.05$) NPN than SPC (Table 1), which was because of the presence of GMP in the whey (which is soluble in 12% TCA) as a result of κ -CN hydrolysis by rennet. The true protein content, expressed as a difference of total N (TN) minus NPN, was higher ($P < 0.05$) for SPC liquid (Table 1) than for WPC liquid.

There was no significant difference detected ($P > 0.05$) in mean ($n = 3$) moisture content of SPC (SD 4.6 and FD 3.33%) versus WPC (SD 4.08 and FD 3.62%) within each drying method. Spray-dried WPC contained more fat and GMP ($P < 0.05$) on a dry basis than SPC, whereas SPC contained more protein on a TN basis (Table 2). The pH of the reconstituted liquid WPC was lower ($P < 0.05$) than that of liquid SPC (Table 2) because of the lactic acid from cheese making that was present in the WPC. The lower pH of the WPC would be expected to cause some differences between WPC and SPC in mineral content. The calcium content of the WPC powder on a dry basis was higher than that of SPC ($P < 0.05$; Table 3). A higher calcium content in WPC would be expected because calcium was released from casein micelles into the cheese whey as pH decreased due to lactic acid production by the starter culture during cheese making. There was a trend for higher phosphorus content in WPC ($P < 0.07$).

Commercial 34% WPC were obtained from 6 different cheese plants and their composition was compared with experimental 34% SPC and WPC. No significant difference was detected ($P > 0.05$) in moisture content among powders (Table 4). Spray-dried 34% WPC produced commercially with an average fat content of 3.08% were higher ($P > 0.05$) in fat than the SPC (0.25%) and WPC (1.93%) produced in this study. The higher fat content of WPC compared with SPC may have negative effects on flavor and functionality. All powders contained more than 34% protein. Most of the commercial powders had significantly higher TN content compared with the

experimental 34% SPC and WPC. Controlling protein concentration closer to the target level of protein for commercial WPC products would increase WPC yield. The pH of 34% WPC produced in our study was similar to pH of all of the commercial WPC. The 34% SPC had the highest pH but was not different ($P > 0.05$) from the pH of several of the commercial WPC. The WPC had lower pH than SPC because of the lactic acid from cheesemaking. The WPC from plants 1, 2, and 3 were from Cheddar cheese whey that had been bleached, the WPC from plants 4 and 5 were from Mozzarella whey that was not bleached, and the WPC from plant 6 was

Figure 2. Sodium dodecyl sulfate-PAGE of proteins in 34% serum protein concentrate (SPC), 34% whey protein concentrate (WPC), and skim milk. The loading of the samples was 7 μ L (8.5 μ L for milk) and samples were run in triplicate. Bands in lanes are identified on the gel: SP1, SP2, SP3 = serum proteins; CN1 = α _S-CN (combination of α _{S1}- and α _{S2}-CN); CN2, CN3, CN4 = casein proteolysis products, β -LG, α -LA.

Table 5. Mean ($n = 3$) relative percentage of casein and serum proteins for 34% whey protein concentrate (WPC) and 34% serum protein concentrate (SPC) powders by the densitometry analysis of the SDS-PAGE gels

Product	Casein	Serum proteins
SPC	3.97 ^a	96.03 ^b
WPC	2.89 ^b	97.11 ^a
R ²	0.89	0.89
SE	0.11	0.11

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

a blend of Mozzarella and bleached Cheddar whey. It is not known if the commercial WPC products were neutralized.

A typical SDS-PAGE gel for 34% WPC and SPC is shown in Figure 2. There was a significant difference in the relative percentage of casein and serum protein between SPC and WPC (3.97 and 2.89% and 96.03 and 97.11%, respectively; Table 5). The difference resulted mostly from the presence of α_{S1} -CN (band CN1 in Figure 2) in SPC, whereas WPC did not contain any α_{S1} -CN. Both 34% SPC and 34% WPC contained casein proteolysis products (band CN2 in Figure 2); however, a higher content of CN2 was found in SPC (3.53 vs. 2.89 for SPC and WPC, respectively). Verdi et al. (1987) studied somatic cell-associated proteolysis and presumed that, as a result of plasmin action on β -CN, the casein-proteolysis products consisted predominantly of γ -CN. Based on comparison of the mobility of band CN2 with the mobility of proteolysis products reported by Verdi et al. (1987), it is likely that CN2 represents secondary proteolysis products of γ_1 -CN = AA 28 to 209 of β -CN (molecular weight: 20,520 Da) and a fragment of α_{S1} -CN (molecular weight: 20,500 Da; Eigel and Keenan, 1979). A significant difference in β -LG and α -LA percentage was detected between WPC and SPC (Table 6) with higher β -LG content estimated for WPC. The typical percentage of β -LG and α -LA for the milk run on the same gels (not milk used in this study) were 78.10 to 21.90 and were similar to those of WPC. The presence of GMP in WPC and not SPC (Table 2) and the difference in fat content may have

Table 6. Mean ($n = 3$) relative percentage of β -LG and α -LA for 34% whey protein concentrate (WPC) and 34% serum protein concentrate (SPC) powders by the densitometry analysis of the SDS-PAGE gels

Product	β -LG	α -LA
SPC	77.24 ^b	22.76 ^a
WPC	78.54 ^a	21.46 ^b
R ²	0.80	0.80
SE	0.16	0.16

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

more effect on functional properties than the small differences in the percentage of β -LG and α -LA between SPC and WPC.

Color

There was an effect ($P < 0.05$) of drying method (SD vs. FD) on the Hunter L, a, b values of the powders (Table 7). Freeze-dried powders were less white (i.e., lower L value) and more brown ($P < 0.05$) than SD powders for both WPC and SPC, which was not expected. The longer time of drying for freeze drying versus spray drying and a shelf temperature of 35°C during the freeze-drying process probably caused more nonenzymatic browning in the FD powders compared with the SD powders. No differences ($P > 0.05$) in L, a, b color values were detected between WPC and SPC powders within the SD or FD powders (Table 8); however, when the powders were reconstituted in water at 10% solids, the liquid solutions produced from the 34% WPC and SPC powders looked very different. The liquids produced from the SPC powders were clear and the liquids produced from the WPC powders were cloudy (Figure 3).

Sensory Analysis

Serum and Whey Proteins. Both WPC and SPC products were characterized by low flavor intensities (Table 9), all of which have been previously reported in dried whey ingredients (Drake et al., 2003, 2009; Wright et al., 2009). Sweet aromatic, cooked flavor, and sweet taste were the most intense attributes found in both of the pilot-plant products. Diacetyl flavor was absent from SPC, but was detected in WPC. Diacetyl flavor, associated with the chemical compound diacetyl, may be due to starter culture fermentation and has been previously documented in the volatile profiles of sweet whey powders and WPC with 80% protein (Mahajan et al., 2004; Carunchia Whetstine et al., 2005a). Both pilot-plant products also displayed low intensities

Table 7. Mean ($n = 3$) Hunter L (lightness), a (red-green), b (yellow-blue) color values of spray-dried and freeze-dried powders

Treatment	Hunter value		
	L	a	b
Spray dried	90.15 ^a	-0.46 ^b	6.16 ^b
Freeze dried	83.37 ^b	1.55 ^a	11.41 ^a
R ²	>0.99	>0.99	>0.99
SE	0.19	0.03	0.21

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

Table 8. Mean ($n = 3$) Hunter L (lightness), a (red-green), b (yellow-blue) color values of spray-dried and freeze-dried serum protein concentrate (SPC) and whey protein concentrate (WPC)

Product	Spray dried			Freeze dried		
	L	a	b	L	a	b
SPC	90.05	−0.50	5.92	83.59	1.32	10.68
WPC	90.25	−0.42	6.40	83.15	1.77	12.15
R ²	0.71	0.94	0.79	0.52	0.95	0.83
SE	0.16	0.01	0.15	0.23	0.05	0.35

(scores of 1 on a 15-point scale) of cereal flavors, and WPC also had a low but detectable cardboard flavor.

SD and FD Powders. The process of drying the products by either spray drying or freeze drying had minimal effects on the sensory profiles of the pilot-plant products (Table 10). Cooked/milky flavor was lower ($P < 0.05$) in SD products compared with FD products, whereas sweet taste was higher in SD products compared with FD products. Cooked/milky flavors are typically associated with Maillard browning and caramelization products. It is possible that the spray-drying process drives off volatiles and decreases perception of cooked/milky flavor, while also causing increased caramelization and increased sweetness. Alternatively, it is possible that the prolonged time for freeze drying the samples at 35°C resulted in additional nonenzymatic browning that imparted additional cooked flavor, just as color was affected. Cardboard flavor was also detected in both FD products and only detected in SD WPC, which may also have been because of the prolonged time required to freeze dry the samples. Typically, freeze drying is a more costly process on a large scale (Resch et al., 2004). However, spray drying can be difficult to perform on a small sample size in a bench-top process. Because flavor properties of FD and SD products were similar, FD may serve as an acceptable alternative for screening bench-top processes for flavor.

Commercial and Pilot-Plant Products. Commercial SD WPC powders of similar composition (Table 4) were compared with pilot-plant products. The 6 com-

mmercial samples demonstrated similar aroma and flavor characteristics as the pilot-plant products but generally with greater intensities (Table 11). One exception was cereal flavor, which was absent in the commercial WPC but present in pilot-plant products. This flavor has been previously documented in commercial WPC containing 80% protein (Russell et al., 2006) so it was not unique to the pilot-plant products. The principal component analysis biplot of the sensory profiles (Figure 4) further demonstrated differences between the pilot-plant products and commercial products. Previous studies have demonstrated wide variability among commercial fluid whey, whey powders, and whey proteins (Carunchia Whetstine et al., 2003, 2005a; Karagul-Yuceer et al., 2003; Mahajan et al., 2004). These differences were attributed to different milk supplies, whey sources, and processing parameters as well as to minor but distinct differences in composition and might represent sources of difference between pilot-plant and commercial products. Fat content of commercial samples was higher than that in pilot products (Table 4). The higher fat content may lead to a higher amount of lipid oxidation products, which are purportedly responsible for many of the off-flavors found in whey proteins (Morr and Ha, 1991; Carunchia Whetstine et al., 2005a).

Instrumental Volatile Analysis

SPME. Selected volatile compounds in commercial and pilot-plant products were quantified by SPME

Table 9. Mean ($n = 3$) sensory attributes¹ of spray-dried serum protein concentrate (SPC) and spray-dried whey protein concentrate (WPC) produced in this study

Product	Aroma intensity	Sweet aromatic	Diacetyl	Cardboard	Cereal	Cooked/milky	Sweet taste	Astringent
SPC	1.7 ^a	0.7 ^b	ND ²	ND	1.0 ^a	1.5 ^a	2.0 ^a	1.7 ^a
WPC	1.8 ^a	1.1 ^a	0.5	0.6	0.7 ^a	1.4 ^a	2.0 ^a	1.7 ^a
R ²	0.26	0.36			0.20	0.25	0.02	0.18
SE	0.06	0.08			0.06	0.08	0.08	0.09

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹Intensities were scored on a 0 to 15-point universal scale where 0 = none and 15 = very high intensity (Meilgaard et al., 1999).

²ND = not detected. Dried whey ingredient intensities generally fall between 0 and 4 (Drake et al., 2003; Wright et al., 2009); attributes not listed were not detected.

Table 10. Mean ($n = 3$) sensory attributes¹ of spray-dried (SD) and freeze-dried (FD) serum protein concentrate (SPC) and whey protein concentrate (WPC) produced in this study

Product	Aroma intensity	Sweet aromatic	Diacetyl	Cardboard	Cereal	Cooked/milky	Sweet taste	Astringent
SD WPC	1.8 ^b	1.1 ^b	0.5 ^a	0.6 ^a	0.7 ^b	1.4 ^a	2.0 ^a	1.7 ^a
SD SPC	1.7 ^{bc}	0.7 ^c	ND ²	ND	1.0 ^a	1.5 ^a	2.0 ^a	1.7 ^a
FD WPC	2.3 ^a	1.9 ^a	0.5 ^a	0.7 ^a	1.0 ^a	2.0 ^a	1.6 ^b	1.5 ^a
FD SPC	1.6 ^c	1.2 ^b	0.5 ^a	0.5 ^a	1.0 ^a	2.0 ^a	1.5 ^b	1.8 ^a
R ²	0.31	0.48	0.14	0.14	0.19	0.25	0.24	0.17
SE	0.07	0.08	0.08	0.08	0.06	0.08	0.08	0.09

^{a-c}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹Intensities were scored on a 0 to 15-point universal scale where 0 = none and 15 = very high intensity (Meilgaard et al., 1999).

²ND = not detected. Dried whey ingredient intensities generally fall between 0 and 4 (Drake et al., 2003; Wright et al., 2009); attributes not listed were not detected.

(Tables 12 and 13). Compounds were selected based on previous literature and because they represented a range of lipid oxidation or fermentation (i.e., diacetyl) compounds and they were consistently detected within quantification limits from 3 or more whey products. The SD WPC had higher ($P < 0.05$) concentrations of hexanal, heptanal, pentanal, nonanal, and diacetyl (2, 3-butanedione) compared with SD SPC, whereas SD SPC had higher concentrations of decanal (Table 12). The higher concentration of lipid oxidation compounds in the SD WPC compared with SD SPC was not surprising, given the higher fat concentration (Table 4) in the WPC. The higher concentration of diacetyl in SD WPC was likely because of starter culture fermentation. Starter culture fermentation has also been associated with lipid oxidation flavors and lipid oxidation volatile compounds in fluid cheese whey (Tomaino et al., 2004; Gallardo Escamilla et al., 2005). Concentrations of diacetyl were higher in FD WPC and FD SPC compared

with SD WPC and SD SPC, respectively, and lipid oxidation compounds (hexanal, heptanal, pentanal, and nonanal) were higher in FD SPC compared with SD SPC. It is possible that the longer time to freeze dry the powders allowed or promoted lipid oxidation. Cardboard flavors were detected in all commercial WPC products (Table 11) and (at threshold) in FD SPC but not in SD SPC (Table 10), and lipid oxidation products have been associated with cardboard flavors in whey and whey protein (Tomaino et al., 2004; Wright et al., 2009), which is in agreement with our results.

Commercial products were characterized by higher total concentrations of volatile compounds based on peak areas (results not shown) and generally had higher concentrations of hexanal, heptanal, and pentanal compared with pilot-scale products manufactured in this study (Table 14 and Figure 5). There was a large amount of variation (Figure 5) among the 6 commercial WPC powders. The principal component analysis bi-

Figure 3. Liquid 34% whey protein concentrate (WPC) and 34% serum protein concentrate (SPC) rehydrated at 10% solids. From left to right: WPC replicate 1, SPC replicate 1, WPC replicate 2, SPC replicate 2, WPC replicate 3, SPC replicate 3.

Figure 4. Principal component biplot of sensory attributes of pilot plant-produced ($n = 3$) spray-dried (SD) 34% whey protein concentrates (WPC) and 34% serum protein concentrates (SPC) and SD commercial 34% WPC (numbers 1 to 6 represent 6 different commercial factories) ($n = 2$ per factory). PC1 and PC2 are principal components 1 and 2.

plot shows that the profile of volatile compounds in SD SPC and SD WPC produced from the same milk were similar to each other but were distinctly different from commercial products. The large amount of variability

among the commercial samples (Figure 5) may indicate that in commercial WPC there could be significant influences of milk source or processing. These results are consistent with sensory profiles of these products as well

Table 11. Mean sensory attributes¹ of spray-dried ($n = 3$) serum protein concentrate (SPC), spray-dried ($n = 3$) whey protein concentrate (WPC) produced in this study, and 6 commercial ($n = 2$) 34% WPC

Source	Treatment	Aroma intensity	Sweet aromatic	Diacytyl	Cardboard	Cereal	Cooked	Oxidized	Sweet taste	Sour taste	Salty taste	Astringent
Experimental	SPC	1.7 ^{bc}	0.7 ^d	ND ²	ND	1.0 ^a	1.5 ^d	ND	2.0 ^b	ND	ND	1.7 ^c
Experimental	WPC	1.8 ^{bc}	1.1 ^c	0.5 ^c	0.6 ^e	0.7 ^b	1.4 ^d	ND	2.0 ^b	ND	ND	1.7 ^c
Commercial WPC	1	2.0 ^b	1.0 ^d	1.0 ^b	2.0 ^b	ND	1.4 ^d	1.5 ^a	1.4 ^c	0.8 ^a	0.5 ^c	3.0 ^a
	2	1.4 ^c	1.5 ^b	ND	1.4 ^d	ND	1.9 ^{bc}	1.6 ^a	1.9 ^b	ND	ND	2.0 ^c
	3	2.3 ^{ab}	2.0 ^a	1.9 ^a	1.7 ^d	ND	2.0 ^b	ND	2.0 ^b	0.7 ^a	2.0 ^a	2.2 ^{bc}
	4	2.0 ^b	1.7 ^b	1.8 ^a	2.9 ^a	ND	1.7 ^{cd}	ND	1.5 ^c	ND	ND	2.5 ^b
	5	2.4 ^a	1.7 ^b	1.0 ^b	2.9 ^a	ND	2.0 ^b	ND	1.5 ^c	ND	1.2 ^b	2.5 ^b
	6	1.7 ^{bc}	2.2 ^a	ND	0.5 ^e	ND	2.3 ^a	ND	2.4 ^a	ND	ND	2.5 ^b
R ²		0.27	0.53	0.76	0.78	0.47	0.35	0.17	0.30	0.22	0.72	0.44
SE: commercial		0.11	0.10	0.08	0.11	0.08	0.10	0.06	0.10	0.05	0.08	0.11
SE: experimental		0.08	0.08	0.06	0.09	0.06	0.08	0.06	0.08	0.05	0.08	0.09

^{a-e}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹Intensities were scored on a 0 to 15-point universal scale where 0 = none and 15 = very high intensity (Meilgaard et al., 1999).

²ND = not detected. Dried whey ingredient intensities generally fall between 0 and 4 (Drake et al., 2003; Wright et al., 2009).

Table 12. Mean ($n = 3$) concentrations of selected aroma compounds ($\mu\text{g/L}$) of spray-dried serum protein concentrate (SPC) and spray-dried whey protein concentrate (WPC) produced in this study isolated using solid-phase microextraction

Product	Dimethyl sulfide	Hexanal	Heptanal	Diacetyl	Acetic acid	Pentanal	Octanal	Nonanal	Decanal
WPC	0.17 ^a	0.72 ^a	0.14 ^a	0.82 ^a	0.24	0.06 ^a	0.40 ^a	0.72 ^a	0.33 ^b
SPC	0.18 ^a	0.19 ^b	0.04 ^b	0.41 ^b	ND ¹	0.02 ^b	0.29 ^b	0.31 ^b	1.04 ^a
R ²	0.65	0.82	0.70	0.30		0.72	0.75	0.61	0.90
SE	0.03	0.06	0.02	0.13		0.01	0.02	0.12	0.06

^{a,b}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹ND = not detected.

as the differences in composition and different processing histories. Both sensory and instrumental volatile compound variability among commercially produced whey proteins has been previously documented (Drake et al., 2009; Wright et al., 2009; Carunchia Whetstine et al., 2005a).

GC-O. The GC-O was conducted on the SD and FD manufactured pilot-scale powders to more closely evaluate and compare flavor-contributing compounds. Twenty-nine aroma-active compounds were detected in the pilot plant SD and FD WPC and SPC (Table 15). The compounds identified were aldehydes, ketones, esters, sulfur-containing compounds, and free fatty acids. Of these compounds, 14 were positively identified by mass spectra, retention index (RI), and odor properties of authentic standards; 13 compounds were tentatively identified by comparing RI and odor properties with authentic standards; 1 was tentatively identified by RI and previously published data; and 1 remained unknown. These compounds are lipid or protein oxidation products and have been previously documented in dried whey products (Mahajan et al., 2004; Carunchia-Whetstine et al., 2005a; Gallardo-Escamilla et al., 2005; Wright et al., 2006, 2009; Javidipour et al., 2008). It is important to keep in mind that AEDA is a semiquantitative technique and therefore the values do not represent actual concentrations of compounds, only their aroma activity in the extracts. As such, AEDA results provide a snapshot or profile of the most po-

tent aroma (flavor) contributing compounds in a given extracted sample (FD WPC, SD WPC, FD SPC, SD SPC) (Audouin et al., 2001). Log₃ FD factors for the same compound differing by ≥ 2 log₃ are considered suggestive of concentration differences. Similarly, SAFE is a different volatile compound extraction approach and recovers different classes of compounds compared with SPME so that compounds not recovered by SPME may be recovered by SAFE with GC-O.

1-Octen-3-one, (E, Z)-2-6-nonadienal, (E)-2-undecanal, and tridecanal, all lipid oxidation compounds, were detected in all 4 products (Table 15). Other compounds were variable or were specific to FD or SD products and WPC or SPC. Five sulfur-containing compounds (dimethyl disulfide, dimethyl trisulfide, methional, dimethyl tetrasulfide, and thenylthiol) were identified in the pilot whey products. Each of these compounds with the exception of dimethyl tetrasulfide and thenylthiol have previously been documented in whey products (Mahajan et al., 2004; Carunchia Whetstine et al., 2005a; Wright et al., 2006, 2009; Mortenson et al., 2007). These sulfur-containing compounds have aromas associated with garlic, cabbage, and potato. Methional has been identified in dairy products as a Strecker degradation product formed from the degradation of methionine. Dimethyl disulfide and dimethyl trisulfide are degradation products of sulfur-containing amino acids (Wright et al., 2006). Thenylthiol has been demonstrated to be a reaction product of hydrogen

Table 13. Mean ($n = 3$) concentrations of selected aroma compounds ($\mu\text{g/L}$) of spray-dried (SD) and freeze-dried (FD) serum protein concentrate (SPC) and whey protein concentrate (WPC) produced in this study isolated using solid-phase microextraction

Product	Dimethyl sulfide	Hexanal	Heptanal	Diacetyl	Acetic acid	Pentanal	Octanal	Nonanal	Decanal
SD WPC	0.18 ^a	0.72 ^b	0.14 ^b	0.82 ^{bc}	0.24 ^a	0.06 ^b	0.28 ^a	0.72 ^a	0.33 ^b
SD SPC	0.17 ^a	0.19 ^b	0.04 ^b	0.42 ^c	ND ¹	0.02 ^b	0.40 ^a	0.31 ^b	1.04 ^a
FD WPC	0.11 ^a	0.18 ^b	0.21 ^b	7.03 ^a	0.07 ^a	0.03 ^b	0.32 ^a	0.82 ^a	0.96 ^a
FD SPC	0.15 ^a	4.56 ^a	0.75 ^a	1.53 ^b	ND	0.43 ^a	0.52 ^a	0.88 ^a	0.34 ^b
R ²	0.30	0.42	0.53	0.88	0.41	0.54	0.28	0.34	0.73
SE	0.36	0.84	0.98	0.37	0.06	0.06	0.05	0.13	0.09

^{a-c}Means in the same column not sharing a common superscript are different ($P < 0.05$).

¹ND = not detected.

Figure 5. Solid-phase microextraction principal components analysis biplot of instrumental analysis results for pilot plant-produced (n = 3) spray-dried (SD) 34% whey protein concentrates (WPC) and 34% serum protein concentrates (SPC) and SD commercial 34% WPC (numbers 1 to 6 represent 6 different commercial factories; n = 2 per factory). PC1 and PC2 are principal components 1 and 2.

sulfide (product of Strecker degradation of amino acid and 2-furfural carbohydrate caramelization product) in meat (Shibamoto, 1980). Dimethyl tetrasulfide, which has a garlic aroma, and has been identified in Cheddar

cheese, is the result of further degradation of methionine (Milo and Reineccius, 1997).

Some compounds were more prevalent in SD products compared with FD products and this may be because

Table 14. Mean concentrations of selected aroma compounds (µg/L) of spray-dried (n = 3) serum protein concentrate (SPC), spray-dried (n = 3) whey protein concentrate (WPC) produced in this study, and 6 commercial (n = 2) 34% whey protein concentrates isolated using solid-phase microextraction

Source	Treatment	Dimethyl sulfide	Hexanal	Heptanal	Diacetyl	Acetic acid	Pentanal	Octanal	Nonanal	Decanal
Experimental	WPC	0.18 ^c	0.19 ^b	0.04 ^c	0.82 ^a	ND ¹	0.02 ^d	0.40 ^a	0.32 ^{ab}	1.04 ^a
Experimental	SPC	0.17 ^c	0.72 ^b	0.14 ^c	0.42 ^a	0.24 ^a	0.06 ^d	0.29 ^a	0.72 ^a	0.33 ^a
Commercial	1	0.63 ^a	42.71 ^a	1.25 ^{ab}	12.11 ^a	1.97 ^a	1.35 ^{bc}	0.59 ^a	0.11 ^b	1.04 ^a
	2	0.40 ^b	39.90 ^a	1.29 ^{ab}	19.21 ^a	4.71 ^a	2.19 ^{ab}	0.81 ^a	0.19 ^b	1.84 ^a
	3	0.22 ^{bc}	39.04 ^a	1.41 ^a	6.25 ^a	0.62 ^a	2.23 ^a	1.08 ^a	0.36 ^{ab}	0.80 ^a
	4	0.26 ^b	48.76 ^a	1.18 ^{ab}	5.05 ^a	0.36 ^a	2.39 ^a	0.94 ^a	0.15 ^b	0.25 ^a
	5	0.40 ^b	36.04 ^a	0.46 ^{bc}	20.02 ^a	9.72 ^a	1.57 ^{abc}	0.98 ^a	0.20 ^b	1.73 ^a
	6	0.62 ^a	36.85 ^a	0.46 ^{bc}	1.95 ^a	0.21 ^a	1.07 ^c	0.69 ^a	0.14 ^b	0.12 ^a
R ²		0.61	0.38	0.37	0.23	0.18	0.64	0.13	0.29	0.17
SE: experimental		0.05	9.1	0.25	4.9	2.3	0.25	0.26	0.12	0.47
SE: commercial		0.07	12.3	0.34	6.6	3.1	0.34	0.35	0.16	0.63

^{a-d}Means in the same column not sharing a common superscript are different (P < 0.05).
¹ND = not detected.

Table 15. Aroma active compounds detected, mean aroma intensity ($n = 3$), and retention index (RI) in spray-dried (SD) and freeze-dried (FD) pilot plant serum protein concentrate (SPC) and whey protein concentrate (WPC) by gas chromatography-olfactometry with aroma extract dilution analysis

No.	Compound	Fraction ¹	Odor ²	Log ₃ flavor dilution ³ (aroma intensity ⁴)				RI ⁵		Methods of identification ⁶
				SD SPC	FD SPC	SD WPC	FD WPC	DB-5	DB-Wax	
1	2/3-Methylbutanal	NB	Malty/chocolate	<1 (1.5)	<1 (2.5)	2 (1.75)	ND ⁷	664		RI, ⁸ odor
2	Acetic acid	AC	Vinegar/sour	ND	1 (1.5)	1 (1)	2 (2)		1510	RI, MS, odor
3	Diacetyl	NB	Buttery/green	ND	ND	2 (3)	1 (1.5)	704	954	RI, odor
4	Pentanal	NB	Peanut/roasted/sweet	1 (2.5)	<1 (2)	2 (2)	ND	731	937	RI, MS, odor
5	Dimethyl disulfide	NB	Rubbery/garlic/sweet	3 (1)	1 (1)	2 (2.5)	<1 (2.25)	732	1073	RI, MS, odor
6	Ethyl butanoate	NB	Fruity/catty	<1 (1.75)	ND	<1 (2.5)	<1 (1.5)	815	1192	RI, odor
7	Hexanal	NB	Grassy/plastic	ND	1 (1.5)	ND	1 (2)	821	1050	RI, MS, odor
8	Butanoic acid	AC	Cheesy/rancid/buttery	ND	<1 (1.75)	2 (1)	<1 (1.5)		1721	RI, MS, odor
9	2-Methylbutyl acetate	NB	Fruity	1 (2)	<1 (2.5)	ND	ND	886		RI, MS, odor
10	Z-4-Heptanal	NB	Plastic/green/fishy	5 (2.5)	ND	<1 (3)	4 (2.25)	909	1224	RI, MS, odor
11	Methional	NB	Potato/green/veg	3 (2.5)	<1 (1.5)	3 (2.5)	3 (1.5)	931	1434	RI, odor
12	2-Acetyl-1-pyrroline	NB	Popcorn/fatty	4 (3.5)	<1 (1.5)	<1 (2)	4 (2)	947	1319	RI, odor ⁸
13	Dimethyl trisulfide	NB	Garlic/sulfur/citrus	6 (2.5)	ND	3 (3)	1 (1.5)	965	1365	RI, odor
14	1-Octen-3-one	NB	Mushroom	5 (2)	3 (2)	3 (2.5)	1 (2.25)	1001	1285	RI, MS, odor
15	Octanal	NB	Citrus	1 (2)	ND	ND	1 (2.5)	1028	1277	RI, MS, odor
16	Unknown	NB	Cooked/cheesy/bread	1 (3)	<1 (2)	ND	ND	1054		Odor
17	Thenylthiol	NB	Garlic	6 (3)	2 (2)	2 (3.5)	ND	1087		RI, odor
18	2-Methoxy phenol	NB	Smokey/burnt/fatty	<1 (2)	3 (2.5)	ND	2 (2.5)	1119	1466	RI, MS, odor
19	2,3-Diethyl-5-methyl pyrazine	NB	Cabbage/potato	1 (1)	<1 (2)	<1 (2.5)	ND	1159	1540	RI, odor
20	2-Phenethanol	AC	Fruity	ND	<1 (1.5)	ND	<1 (1.5)	1167	1880	RI, odor
21	(E,Z)-2-6-Nonadienal	NB	Cucumbers/burnt	1 (2.5)	3 (2.5)	<1 (2.5)	2 (2.5)	1173	1565	RI, MS, odor
22	(E)-2-Nonenal	NB	Sweet/carpet/cucumber	<1 (2)	<1 (2)	ND	<1 (2.5)	1188	1580	RI, MS, odor
23	Dimethyltetrasulfide	NB	Garlic	4 (2)	<1 (2.5)	ND	ND	1202		RI, MS, odor
24	Phenylethyl acetate	AC	Cucumbers/rosy	ND	3 (2)	<1 (1.75)	<1 (3)	1292	1815	RI, odor
25	(E)-2-Undecenal	NB	Rubber	4 (2)	1 (1)	3 (1)	5 (2)	1377		RI, odor
26	Decanoic acid	AC	Fatty	ND	<1 (1.5)	ND	3 (2.5)	1418	2411	RI, MS, odor
27	2-Dodecenal	NB	Sweet/green/fat	<1 (2)	1 (1.5)	ND	1 (1.5)	1454		RI, odor
28	Tridecanal	NB	Sweet/must	4 (2.25)	3 (1)	3 (3)	2 (2)	1511		RI, odor
29	(E,E)-Farnesyl acetate	NB	Waxy	<1 (1.5)	ND	<1 (1)	ND	1925		RI, odor

¹NB = neutral/basic compounds; AC = acidic compounds.²Odor description at the gas chromatograph (GC) sniffing port.³Flavor dilution factors were determined on a DB-5MS column for NB compounds and on a DB-Wax column for AC compounds.⁴Aroma intensity at the GC sniffing port on a scale from 1 to 5.⁵Retention indices were calculated from gas chromatography/olfactory data.⁶Compounds were identified by comparison with the authentic standards on the following criteria: RI on DB-Wax and DB-5MS columns, odor property at the GC-sniffing port on both columns, and mass spectra (MS) in the electron impact mode. Positive identifications indicate that mass spectral data were compared with authentic standards.⁷ND = not detected.⁸Compound identified by comparing RI and aroma with literature (Avsar et al., 2004).

of differences in the temperature conditions used in the 2 processes. Specifically, 2/3-methylbutanal, dimethyl sulfide, and dimethyl trisulfide were more apparent in SD products compared with FD products. These differences may reflect the extreme heat to which products are exposed during spray drying. Thermal denaturation of the 2 main whey proteins, β -LG and α -LA, occurs between 50 and 75°C causing the protein to unfold and disulfide bonds to break and unmasking an SH group (Linden et al., 1999). The higher presence of sulfur compounds in the SD products is most likely caused by denaturation from the higher temperature exposure during drying (inlet temperature of 200°C, outlet temperature of 95°C). In contrast, hexanal and phenethanol were only detected by GC-O in FD products and 2-methoxyphenol and (E, Z)-2-6-nonadienal were present at higher flavor dilution factors, on average, in FD products compared with SD products. Hexanal and (E, Z)-2-6-nonadienal are lipid oxidation products, whereas 2-phenethanol and 2-methoxyphenol are formed by the Strecker degradation of aromatic amino acids, especially phenylalanine (Singh et al., 2003; Carunchia Whetstine et al., 2005a). Once again, the prolonged time required for freeze drying at 35°C may have promoted formation of these compounds.

Acetic acid, diacetyl, butanoic acid, and methional generally had higher flavor dilution factors in WPC or were not detected in SPC. All 4 of these compounds are potent odorants in Cheddar cheese and likely represent differences due to starter culture fermentation (Singh et al., 2003). 1-Octen-3-one, thenylthiol, dimethyl tetrasulfide, and 2-methylbutyl acetate were more prevalent in SPC. 1-Octen-3-one is a lipid oxidation compound previously reported in whey products (Carunchia Whetstine et al., 2005a), whereas thenylthiol, dimethyl tetrasulfide, and 2-methylbutyl acetate have not been previously identified in dried whey; however, they have been identified in other dairy products including ewe's milk cheeses (Larrayoz et al., 2001), Cheddar cheese (Carunchia Whetstine et al., 2005b; Avsar et al., 2004), Parmesan cheese (Qian and Reineccius, 2003), and Gorgonzola cheese (Moio et al., 2000).

CONCLUSIONS

The 34% protein SPC and WPC manufactured from the same milk were distinct in composition. The WPC had higher fat and calcium contents and lower pH than SPC. The WPC contained GMP and the SPC did not. Although color differences were not evident between the 2 spray-dried or 2 freeze-dried powders, SPC solutions were clear whereas WPC solutions were cloudy when rehydrated at 10% solids. Sensory differences between SPC and WPC produced from the same source of milk

were minor but distinct, as were volatile compound differences. Most differences seemed to be related to differences in fat content and compounds that would be derived from starter culture in whey. However, both pilot-plant products were bland in flavor compared with commercially manufactured WPC of similar composition. Lipid oxidation products were also generally lower in the products manufactured in this study compared with commercially manufactured WPC. Our results suggest that when SPC and WPC are manufactured under controlled conditions in a similar manner with same equipment, there are few sensory differences between them but distinct compositional differences and differences in physical properties, which may influence functionality. Further, flavor (sensory and instrumental) properties of both pilot-scale protein powders were different from commercial powders, and there were large differences in flavor among the commercial products, suggesting the role of other influencing factors (e.g., processing conditions, equipment sanitation, and milk source).

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