JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY

Reduction of Stale Flavor Development in Low-Heat Skim Milk Powder via Epicatechin Addition

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The influence of epicatechin (EC) on off-flavor development in low-heat skim milk powder samples during processing and storage was investigated. Milk powder samples were prepared from a concentrated skim milk (control) plus a concentrated skim milk with EC (treatment). Volatile extracts of the powders were analyzed by aroma extract dilution analysis (AEDA) at 0 days and after 17 months of storage in conjunction with sensory analysis of the flavor attributes. The treatment milk powders with EC added prior to drying reported a reduction in the formation of three main compounds, 4-hydroxy-2,5-dimethyl-3-(2H)-furanone, *o*-aminoacetophenone, and furfural, by 8-, 4-, and 4-fold for the 0 day old samples, while for the 17 month aged samples *o*-aminoacetophenone was the major compound reduced in formation by 8-fold, respectively, based on the flavor dilution factors reported. The sensory evaluations indicated that the treatment milk powders for 0 day old and 17 month aged samples were statistically lower ($\alpha = 0.05$) in stale flavor intensity in comparison to the respective control samples, while no differences were noted in bitterness intensity.

KEYWORDS: Skim milk powder; nonfat dried milk; AEDA; epicatechin; stale; aroma; flavor; reduction

INTRODUCTION

Milk powders are widely utilized as ingredients in the food industry. In comparison to fluid milk, milk powders have lower shipping expenditures, improved microbial stability, and unique functionality (*1*). Consequently, milk powder has become an important commodity of the U.S. dairy industry, and the production of skim milk powder (SMP) in 2005 is estimated to be 685,000 metric tons, largely due to the annually increasing exportation of this product (*2*). Although SMP has many advantages as a food ingredient, it can also impart negative attributes to the finished product, such as "stale" off-flavor characteristics. The development of stale or gluey off-flavors in milk powders historically has been characterized as a Maillard type reaction product (*3*−*6*).

Parks et al. (*3*), in 1964, first reported that *o*-aminoacetophenone contributed to the stale off-flavor of milk powder. Ferretti and Flanagan (*4*) tried to determine key aroma compounds, which contribute to the stale properties of a 2 year old SMP using low-temperature steam distillation. They suggested 12 compounds (consisting of alkylpyrazines and pyrroles) that were important in the stale off-flavor development in the aged samples. More recently, Karagul-Yuceer et al. (*5*, *6*) using aroma extract dilution analysis (AEDA) defined the overall aroma profile of the three different types of SMP ranging from 3 months to 25 years in age. These authors reported that Maillard reaction compounds like 4-hydroxy-2,5-dimethyl-3-(2H)-furanone (Furaneol), methional, *o*-aminoacetophenone, 2-acetyl-1-pyrroline, and 2-acetyl-2-thiazoline were important aroma-active compounds, which contribute to the stale flavor properties of these milk powders. Consequently, the ability to control the Maillard reaction in milk powders would be beneficial to limit off-flavor development and likewise improve the product quality.

Natural product polyphenol extracts (i.e., epicatechin, EC) have recently been reported to be inhibitors of the Maillard reaction in aqueous model systems and food products (*7*) and have been applied to control cooked flavor development during ultrahigh-temperature processing of fluid milk (*8*). The mechanism of how EC functions as an inhibitor of the Maillard reaction was furthermore reported by Totlani and Peterson (*9*) to be, in part, due to the direct quenching of key precursors or intermediate reaction products (C_2, C_3, and C_4 sugar fragments).

The objective of this study was to investigate the ability of EC to inhibit the generation of key off-flavor Maillard compounds during production (spray drying) and storage in low-heat SMP (low-moisture food matrix).

MATERIALS AND METHODS

The following chemicals were obtained from commercial suppliers for analysis: diethyl ether (DEE) 1× repurified was prepared by distilling high-purity 99.9% anhydrous DEE (Burdick & Jackson, Muskegon, MI) through packed column distillation (30 cm, 4 mm glass beads, distilled at 40 °C), 99% EC (Zhejiang Yixin Pharmaceutical Co., Ltd., China), sodium chloride (J. T. Baker, Philipsburg, NJ), sodium sulfate (EMD Chemicals, Inc., Gibbstown, NJ), sodium bicarbonate (EM Science, Gibbstown, NJ), hydrochloric acid (Fisher Scientific, Fair

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10.1021/jf0519764 CCC: $33.50
Published on Web 12/15/2005

Lawn, NJ), and caffeine (Sigma-Aldrich Chemical Co., St. Louis, MO). Sucrose (Domino Granulated Pure Cane Sugar, Domino Foods, Inc.) was obtained from the local supermarket.

Flavor chemicals no. 1, 2, 4−6, 8−10, 12, and 14−20 (see **Tables 1−3**) and the internal standard 2-methyl-3-heptanone for the neutral basic fraction were purchased from Sigma-Aldrich Chemical Co. Chemical no. 7 was purchased from Pyrazine Specialities, Inc. (Atlanta, GA). Internal standard 2-methylpentanoic acid for the acidic fraction was bought from TCI America (Portland, OR), and no. 13 was purchased from Silesia (Chicago, IL).

Chemical no. 3 was not available for purchase commercially. This compound was extracted from Pandan leaves (*Pandanus amaryllifolius* Roxb.), which were bought at a local market. An extract of the Pandan leaves was prepared according to the procedure outlined by Colahan-Sederstrom and Peterson (*8*).

Milk Powder Production and Storage Conditions. Two gallons of low-heat condensed skim milk (0.11% butterfat and 32% solids) was obtained from Land O' Lakes (Carlisle, PA) and spray dried over two different time periods. The concentrated milk was first spray dried for the accelerated shelf life (ASL) study and then second, <1 week prior to the end of the ASL storage time period to coordinate the analytical and sensory analysis of both the 0 day old or "fresh" (<1 week at −40 °C) and the aged milk powder, which was adopted from the experimental design previously reported by Hough et al. (*10*) and Driscoll et al. (*11*) for off-flavor sensory analysis of SMP. For each processing time period, the milk was divided into two 1 gallon portions by weight. EC was then added to 1 gallon of the concentrated milk at the 0.1% level (w/w) (treatment milk), while nothing was added to the remaining gallon of concentrated milk (control milk). Addition of EC at the 0.1% level to low-heat condensed skim milk was selected based on two key criteria: (i) This level was effective in a previous study by Colahan-Sederstrom and Peterson (*8*) to reduce off-flavor development during the UHT processing of fluid milk, and (ii) at this level, when hydrated to 10% solids, no noticeable bitterness was apparent by benchtop analysis.

Milk samples were spray dried using a vertical floor spray dryer (Niro Inc., Hudson, WI). The spray drying conditions were as follows: inlet temperature, 200 °C; outlet temperature, 80 °C. The milk was continuously agitated with an electric mixer (Cole Palmer, Vernon Hills, IL) while it was pumped at a flow rate of 25 mL/min to a two-fluid pneumatic atomizer using a peristaltic pump (Master Flex L/S, Cole Palmer). The atomizer was pressurized at 50 psi. The spray dried powder was collected and transferred to 1 L amber glass bottles with Teflon polyethylene-lined caps with septa (VWR International, West Chester, PA). For the estimated 0 day old samples, 200 g of the control and of the treatment powder was placed in amber glass bottles and was frozen in a blast freezer at −40 °C for <1 week prior to analytical and sensory analysis. For the ASL storage study, 200 g of control and treatment powder in amber glass bottles (referred to as 17 month powder) was stored in an oven (Napco, Winchester, VA) at 37 °C for 17 weeks (estimated 17 month shelf life) (*12*). The 17 week storage period for the ASL samples was selected based on when experienced milk tasters (benchtop analysis) considered a duplicate control SMP sample to have a very notable stale flavor property (evaluated every 3 weeks and until week 15 and then every week thereafter). Initially, the milk tasters described the initial ASL control sample (at 0 days) to be a very typical "fresh" SMP (low stale−similar to pasteurized skim milk).

Milk Powder Moisture Content, T_g Determination, and Oxygen Content. The moisture content was analyzed using a vacuum oven technique following AOAC method 927.05 (*13*). A differential scanning calorimeter (Perkin-Elmer DSC 7, Boston, MA) was used to measure the glass transition temperature (T_g) of the milk powders. The DSC program was as follows: It was held for 5 min at 20 °C, then it was cooled from 20 to −30 °C at 40 °C/min, and then it was heated from −30 to 70 °C at 5 °C/min. The oxygen content was monitored with a headspace analyzer probe (Mocon PAC Check, model 450, Minneapolis, MN). The moisture content of the 0 day old control and treatment powders was found to be 2.4% moisture d.b. ± 0.1, whereas the ASL milk powders were 3.5% moisture d.b. ± 0.1 before and 3.4% moisture d.b. ± 0.1 after the ASL study. The T_g temperature for the aged powders was found to be ≥57 °C both at 0 day old and after the ASL study, indicating that the powders were in the glassy state. The oxygen content of the headspace remained unchanged during the ASL study (21%).

Preparation of Extracts. Volatile extracts of the milk powders were prepared using a modified version of the methods found in Karagul-Yuceer et al. (*5*) and Colahan-Sederstrom and Peterson (*8*). Ninety grams of each powder was reconstituted using 500 mL of boiled deodorized water. The milks were placed in 1 L graduated flasks, and 6 μL of internal standards 2-methyl-3-heptanone (22 μL/5 mL in methanol for the neutral basic fraction) and 2-methylpentanoic acid (29 μL/5 mL in methanol for the acidic fraction) was added to each milk and gently shaken using an orbital shaking table (model 3540, Labline Instruments, Inc.) at 50% power for 30 min. Internal standards were used to ensure equivalent sample workup and detection [gas chromatography−olfactometry (GC-O)]. The milk was then divided equally into 3−250 mL wide mouth Teflon screw cap bottles (Nalgene, Rochester, NY). Sodium chloride (J. T. Baker) (68 g/190 g milk) was added to each bottle and stirred. A 150 mL amount of repurified 99% purity DEE (Burdick & Jackson) was used to extract the milk volatiles (4 × 150 mL). Bottles were shaken on an orbital shaking table for 20 min at 50% power and then centrifuged at 4 °C for 20 min at 5000*g* (IECCR 6000, Damon/IEC Division, Needham Heights, MA). The organic supernatant was decanted from the milk, pooled, and dried with anhydrous sodium sulfate (EM Science). Extracts were filtered and subsequently concentrated to 130 mL using packed column distillation (30 cm, 4 mm glass beads, distilled at 40 °C). The solvent-assisted flavor evaporation technique (*14*) was used to collect the volatile fraction of the milk extracts (distillate).

The distillates were then fractionated into neutral/basic (N/B) and acidic (A) fractions. Each distillate was extracted with 1 M $NaHCO_3$ (EM Science) (3 × 50 mL). The organic phase was then washed with a saturated NaCl solution (2 × 55 mL), and the organic phase was collected (N/B fraction). The aqueous phase from the $NaHCO_3$ extract was acidified to a pH of ∼2.0 using 12 N HCl and then extracted using 1 × repurified DEE (3 × 50 mL) collecting the organic phase (A fraction). The N/B and A fractions were then distilled to a final volume of 400 μL using spinning band distillation (B/R Instruments Corp., Easton, MD; spinning band at 60% power, 40 °C). The extracts were divided into two equal parts, one for gas chromatography−mass spectrometry (GC-MS) analysis (200 μL) and one for GC-O analysis (200 μL).

GC-MS. Each sample was injected on an Agilent 6890A GC system (Agilent Technologies, Inc., New Castle, DE) equipped with an Agilent 5973 mass selective detector (MSD) (Agilent Technologies, Inc.) and an EZ No-Vent Connector (Restek, Bellefonte, PA). All samples were run on two columns of different polarities: DB-5ms (60 m × 0.25 mm × 0.25 μm) and DB-Wax (60 m × 0.25 mm × 0.25 μm) or DB-FFAP (30 m × 0.25 mm × 0.25 μm) for the neutral/basic and acidic fractions, respectively. GC conditions for DB-5ms and DB-Wax were as follows: inlet temperature, 200 °C; 1 μL of sample was injected in pulsed splitless mode (17 psi; 0.5 min); hydrogen carrier gas at a constant flow of 0.8 mL/min. GC conditions for DB-FFAP were as follows: inlet temperature, 200 °C; 1 μL of sample was injected in splitless mode (1 min); hydrogen carrier gas at a constant flow of 0.8 mL/min. The oven conditions for the DB-5ms column were as follows: 30 °C for 2 min, ramped at 2 °C/min to 250 °C, held for 2 min at 250 °C. For the DB-Wax column, the oven conditions were as follows: 30 °C for 2 min, ramped at 3 °C/min to 230 °C, held for 4 min at 230 °C. For the DB-FFAP column, the oven conditions were as follows: 40 °C for 2 min; ramped at 5 °C/min to 230 °C; held for 4 min at 230 °C. The MSD operational parameters were as follows: capillary direct interface temperature at 250 °C, source temperature at 150 °C, mass range 35−250 amu at 6.35 cycles/min. Positive identifications by MS were determined by comparing the mass spectra fragmentation pattern of aroma compounds in the SMP extracts with mass spectra of known compounds from the Wiley Database. Linear retention index (LRI) values were calculated using an *n*-alkane ladder.

GC-O/AEDA. GC-O was performed using a Hewlett-Packard 5890 Series II GC equipped with a flame ionization detector (FID), autosampler (HP 7673), and sniffing port. The effluent was split (1:1) between the FID and sniff port with deactivated capillary tubing (1 m

length/0.15 mm i.d.; SGE Inc., Austin, TX). A stream of nitrogen at 40 mL/min was applied to sweep the effluent out of the sniffing port. A DB-5ms (30 m × 0.25 mm × 0.25 μm) and a DB-FFAP (30 m × 0.25 mm × 0.25 μm) column were used for analysis of the neutral/basic and acidic fractions, respectively. GC conditions were as follows: The inlet temperature was 200 °C, the detector temperature was 250 °C, and the sniff port temperature was 230 °C; 1 μL of sample was injected in splitless mode (1 min). The oven profile for the neutral/basic fraction (DB-5ms) and the acidic fraction (DB-FFAP) was the same as described for the GC-MS analysis for the respective columns. For AEDA, samples were serially diluted (1:1) with high-purity DEE. Three experienced panelists sniffed the GC effluent for each sample dilution. The dilutions were analyzed until no panelists reported any odorants, and flavor dilution (FD) factors were calculated similar to Kirchhoff and Schieberle (*15*). LRI values were calculated using an *n*-alkane ladder. All odors from GC-O analysis were positively (LRI, odor, MS, and reference compound) or tentatively identified (LRI, odor, and reference compound).

Semiquantitative Analysis of *o*-Aminoacetophenone [GC/MS-Selected Ion Monitoring (SIM)]. Quantification of *o*-aminoacetophenone was preformed using the same GC-MS conditions as outlined above with the following exception: The MSD was operated in SIM mode. Two specific ions were monitored for the analyses, *m*/*z* 85 for 2-methyl-3-heptanone and *m*/*z* 120 monitored for *o*-aminoacetophenone. All samples were run on DB-Wax (60 m × 0.25 mm × 0.25 μm). A standard curve was used to quantify 2-methyl-3-heptanone (10, 20, 60, 120, and 180 μg/mL DEE) and *o*-aminoacetophenone (0.005, 0.01, 0.05, 0.1, and 0.5 μg/mL DEE) in the aroma extracts. The peak area of the single ion for each compound was plotted vs concentration ($r^2 > 0.99$). The concentration of *o*-aminoacetophenone in the SMP samples was calculated as follows:

$$a = \frac{b}{c} \times d \times 5.6$$

where a = μg *o*-aminoacetophenone/kg SMP, b = μg/mL of *o*-aminoacetophenone in aroma extract (determined by calibration curve), c = μg/mL of 2-methyl-3-heptanone in aroma extract (determined by calibration curve), and d = μg of 2-methyl-3-heptanone spiked with internal standard in the 180 g SMP 1 L solution.

Sensory Evaluation and Statistical Analysis. Descriptive sensory analysis using the spectrum method was conducted on the reconstituted milk powders (*16*, *17*). The milk powder was prepared according to International Dairy Federation standards (*18*) using filtered water and an electric mixer (Oster, Shelton, CT) on medium for 30 s. Milk powders were prepared, poured into 3.5 oz plastic cups (Fabri-Kal, Kalamazoo, MI), coded with three digit numbers, and put into an Ambi-Hi-Low chamber (Labline Instruments, Inc., Melrose Park, IL) at 20 °C for 1 h prior to each panel to ensure that all milk samples were sampled at ambient conditions.

Twelve panelists ranging in age from 23 to 65 years were trained over 12 × 1 h sessions on the flavor properties of SMP. These sessions included taste sessions, discussions, and screening to ultimately determine the panelists that were capable of perceiving the attributes in the milk samples. The panel evaluated the milk powders for the attributes of bitter (caffeine solutions 0.05 and 0.055% w/w in water corresponding to a 2 and 4 reference value), cooked, and stale using a 15 cm line scale (0 = none, 15 = very high). The intensity of the cooked and stale attributes was measured using cross-modality of the sucrose solutions (sucrose solutions 2, 4, and 6% w/w in water corresponding to reference values 2, 4, and 6). To familiarize the panel with the concept of both a cooked and a stale dairy flavor (i) a cooked sample was made by microwaving (General Electric Co., Louisville, KY, 1.3 KW, VAC/HZ: 120/60) 500 mL of pasteurized skim milk from a local dairy on high for 4.5 min to 75−80 °C and (ii) a stale sample was prepared from a low-heat SMP, which was >3 months old. Panelists were also provided with a 2 week old commercial SMP sample as an example of a low stale milk powder.

Products were evaluated in duplicate over two sessions in 1 day using a complete balanced block design computed with Compusense five (version 4.2, Guelph, ON, Canada). Evaluations occurred in individual booths in a quiet, odor-free environment. The milk was presented to the panelists in random order. Panelists were instructed to swallow the milk samples and to rinse their palates between samples with filtered water. The data were analyzed by a general linear model analysis of variance test with sample, panelist, and their interaction as variation factors. The least significant difference values were found by running an all-pairwise comparison test using Statistix v. 8.0 (Tallahassee, FL).

Table 1. Aroma-Active Compounds in Estimated 0 Day Old SMP Detected during AEDA (FD Factor ≥ 4) – Neutral/Basic Fraction[a]

		LRI			FD factor	
no.	compound	DB-5ms	DB-Wax	odor[b]	control	treatment (0.1% EC)
1	furfural[c]	831	1462	sweet	16	4
2	methional[d]	905	1453	potato	16	8
3	2-acetyl-1-pyrroline[d]	920	1327	corn	16	8
4	phenylacetaldehyde[c]	1042	1642	rose/floral	16	16
5	4-hydroxy-2,5-dimethyl-3-(2H)-furanone[d]	1053	2033	sugar/caramel	16	2
6	acetophenone[c]	1063	1652	fruity	4	4
7	2-acetyl-2-thiazoline[c]	1102	1761	popcorn	4	2
8	*o*-aminoacetophenone[c]	1299	2225	grape	4	1
9	decanoic acid[c]	1371	2275	sour/soapy	8	8
10	δ-decalactone[c]	1490	2201	coconut	4	4

[a] The FD factor was ≥4 for either the control or the treatment. [b] Odor described at the GC sniffing port during GC-O. [c] Compound positively identified (LRI, MS, and reference compound). [d] Compound tentatively identified (LRI, odor, and reference compound).

Table 2. Aroma-Active Compounds in Estimated 17 Month Old SMP Detected during AEDA (FD Factor ≥ 4) – Neutral/Basic Fraction[a]

		LRI			FD factor	
no.	compound	DB-5ms	DB-Wax	odor[b]	control	treatment (0.1% EC)
2	methional[d]	905	1453	potato	8	8
3	2-acetyl-1-pyrroline[d]	918	1327	popcorn/rice	32	16
4	phenylacetaldehyde[c]	1039	1642	floral	128	64
5	4-hydroxy-2,5-dimethyl-3-(2H)-furanone [d]	1051	2033	sweet/sugar/candy	16	8
6	acetophenone[c]	1062	1652	grape/fruity	8	8
7	2-acetyl-2-thiazoline[c]	1098	1761	popcorn/corn	32	16
8	*o*-aminoacetophenone[c]	1296	2225	grape	16	2
10	δ-decalactone[c]	1487	2201	coconut	4	4
11	unknown	1356		sweet/coconut	4	2
12	γ-decalactone[c]	1465	2149	sweet	8	8
13	γ-6-(*Z*)-dodecenolactone[c]	1650	2405	floral/spicy/coconut	32	32
14	γ-dodecalactone[c]	1677	2381	sweet	4	4

[a] The FD factor was ≥4 for either the control or the treatment. [b] Odor described at the GC sniffing port during GC-O. [c] Compound positively identified (LRI, MS, and reference compound). [d] Compound tentatively identified (LRI, odor, and reference compound).

RESULTS AND DISCUSSION

The FD factors for the aroma-active compounds identified in the 0 day old (stored for < week 1 at −40 °C) and 17 month old (aged) SMP samples are reported in **Tables 1** and **2** (N/B fraction), respectively. The aroma actives identified in the 17 month SMP samples acidic (A) fractions are also reported in **Table 3**. Furthermore, any compounds that reported the FD factor of the control divided by treatment sample as ≥2 for the 0 day old and 17 month old SMP samples are illustrated in **Figures 1** and **2**, respectively. Overall, the addition of EC prior to spray drying had the greatest reduction on the formation of compounds furfural, phenylacetaldehyde, methional, *o*-aminoacetophenone, 4-hydroxy-2,5-dimethyl-3-(2H)-furanone, 2-acetyl-1-pyrroline, and 2-acetyl-2-thiazoline during processing and storage. These results were in agreement with those by Colahan-

Table 3. Aroma-Active Compounds in Estimated 17 Month Old SMP Detected during AEDA (FD Factor ≥ 4) – Acidic Fraction[a]

		LRI			FD factor	
no.	compound	DB-FFAP	DB-5ms	odor[b]	control	treatment (0.1% EC)
15	butanoic acid[c]	1613	822	cheesey/vomit	64	64
16	3-methylbutyric acid[c]	1664	882	stinky/acidic	8	8
17	hexanoic acid[c]	1850	1026	sour/cheesey	16	16
18	octanoic acid[c]	2068	1186	soapy	256	256
19	phenylacetic acid[c]	2569	1248	floral/perfume	64	64
20	3-phenylpropionic acid[d]	2633	1347	floral	16	16

[a] The FD factor was ≥4 for either the control or the treatment. [b] Odor described at the GC sniffing port during GC-O. [c] Compound positively identified (LRI, MS, and reference compound). [d] Compound tentatively identified (LRI, odor, and reference compound).

Sederstrom and Peterson (*8*) who reported that EC added to raw fluid milk prior to UHT processing reduced the thermal generation of aroma compounds (cooked flavor) and had the greatest impact on Maillard type reaction products.

The influence of spray drying (i.e., thermal processing step) on the aroma properties of SMP has not been, to our knowledge, previously defined. Direct comparison of the ratio of FD factors for the 0 day old (stored < week 1 at −40 °C) control and treatment powders (see **Figure 1**) for the indicated that spray drying did impact the overall aroma development of the SMP. Sensory evaluations of the 0 day old milk powders are also presented in **Table 4** and were in agreement with the data illustrated in **Figure 1**; the control was statistically higher in stale flavor intensity than the treatment powder (no difference was noted in "cooked" intensity). Albeit, the stale flavor intensity in both samples was relatively low, the panel was still able to detect a statistical difference between the two powders. The largest noted reduction in FD factors for the control powder in comparison to the treatment powder was for 4-hydroxy-2,5-dimethyl-3-(2H)-furanone (at 8-fold), while *o*-aminoacetophenone, furfural, methional, 2-acetyl-2-thiazoline, and 2-acetyl-1-pyrroline reported 4-, 4-, 2-, 2-,and 2-fold lower FD factors, respectively. 4-Hydroxy-2,5-dimethyl-3-(2H)-furanone, a known sugar degradation product, has been previously reported in SMPs (*5*, *6*). *o*-Aminoacetophenone has been reported by Arnold et al. (*19*) in a stale concentrated milk and also in SMP (*3*, *5*, *6*). It has been suggested that *o*-aminoacetophenone is the degradation product of the amino acid tryptophan. Moreaux and Birlouez-Aragon (*20*) found that tryptophan may be degraded during the advanced stages of the Maillard reaction in β-lactoglobulin-lactose mixtures, suggesting how *o*-aminoacetophenone can be formed in milk powders. The sugar degradation product, furfural, also has been previously reported in UHT milk (*8*, *21*−*25*), whereas 2-acetyl-2-thiazoline has been reported to be formed from sugar degradation products in the presence of cysteine (*26*). Methional, a commonly identified sulfur-containing compound found in thermally processed milk products, and phenylacetaldehyde are both known to be formed via Strecker degradation of the amino acid methionine (*27*) and phenylalanine (*28*), respectively. The last compound 2-acetyl-1-pyrroline, a known product of 2-oxopropanal and proline (*29*), has been reported in other dairy products such as fresh milk (*30*), UHT milk (*8*), liquid cheddar whey (*31*), sweet whey powder (*32*), and nonfat dry milk powder (*5*, *6*).

Historically, the development of stale flavor in milk powders has been primarily associated with storage time. As anticipated, both the AEDA and the sensory evaluations of the 17 month SMP (see **Tables 2** and **4**) suggested that the aroma development and stale flavor intensity of the milk powders increased during storage (in comparison to the 0 day old powders; **Table 1**). It should be noted that the 0 day old and 17 month old samples are different milk powders (spray dried at different time periods);

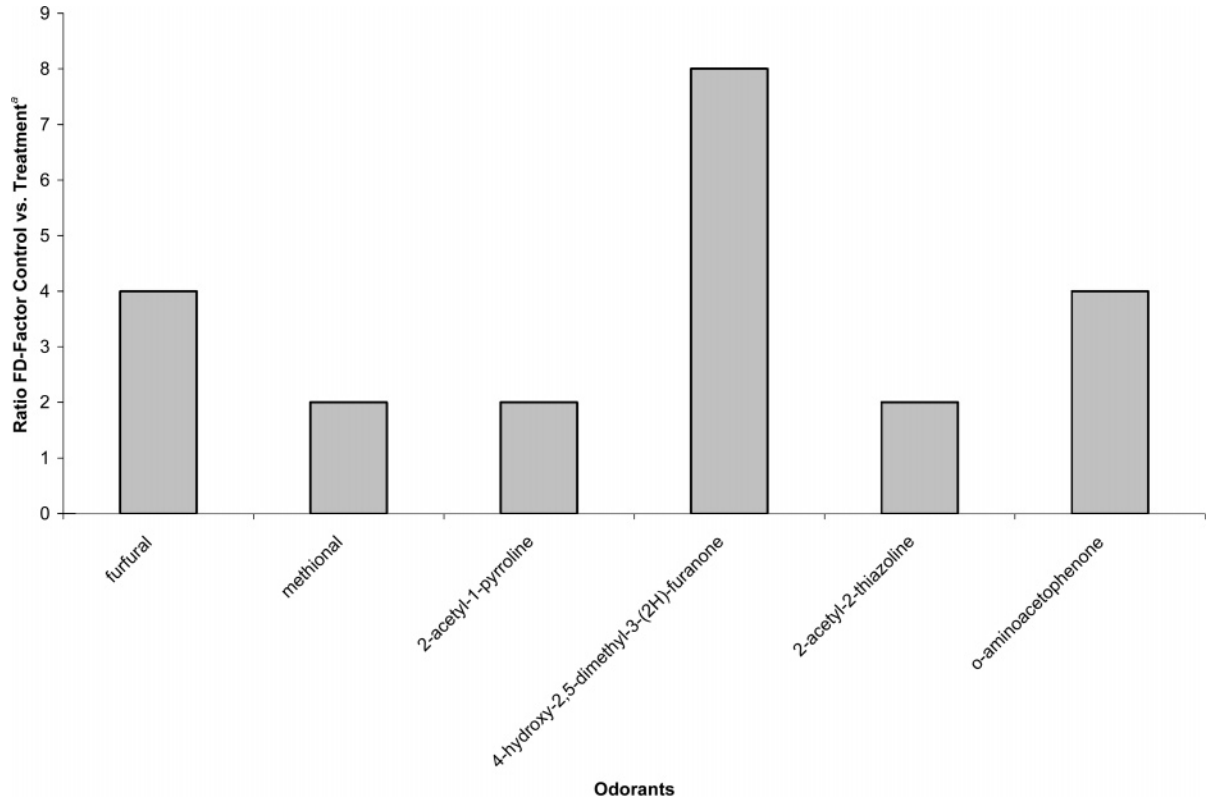

Figure 1. Ratio of FD factor for the control 0 day old milk powder vs treatment (0.1% EC) 0 day old milk powder; only odorants with a ratio of ≥2 FD factor are shown. [a] $2^{\text{FD factor control}}/2^{\text{FD factor treatment}}$.

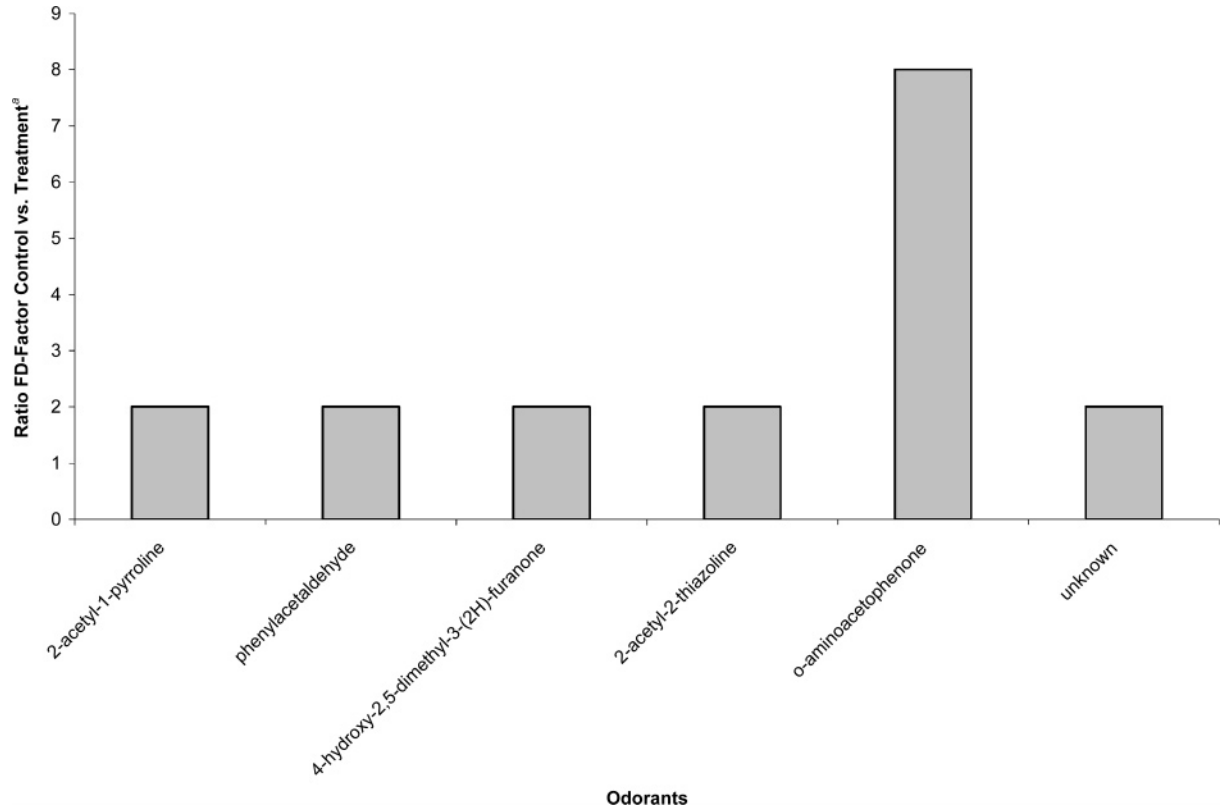

Figure 2. Ratio of FD factors for control estimated 17 month old milk powder vs treatment (0.1% EC) 17 month old milk powder; only odorants with a ratio of ≥2 FD factors are shown. [a] $2^{\text{FD factor control}}/2^{\text{FD factor treatment}}$.

Table 4. Mean Scores for Stale, Cooked, and Bitter Intensity of Milk Powders[a]

	flavor attributes[b,c]		
sample	stale (LSD = 0.40)	cooked (LSD = 0.36)	bitter (LSD = 0.19)
control, 17 months	3.82 A	1.68 AB	0.25 A
treatment (0.1% EC), 17 months	2.33 B	1.46 B	0.35 A
control, 0 day old	1.97 B	1.99 A	0.19 A
treatment (0.1% EC), 0 day old	1.56 C	1.72 AB	0.28 A

[a] $n = 12$. [b] A 15 cm line scale was used for evaluations (0 = none) and (15 = very high). [c] Different letters (A–C) indicate a statistically significant difference between samples ($\alpha = 0.05$).

thus, the statistical comparison of these samples could be confounded by any processing effects. However, both the 0 day old and the 17 month old SMP samples were found to be very similar in flavor properties directly after spray drying (benchtop analysis by experienced milk tasters—as compared to pasteurized skim milk; data not shown) and likewise were considered equivalent. Furthermore, SMP is a bulk commodity; therefore, a low variation in initial SMP flavor quality would be expected.

Direct comparison of the ratio of the FD factors for aged milk powders also indicated that EC reduced the formation of *o*-aminoacetophenone, 2-acetyl-2-thiazoline, phenylacetaldehyde, 4-hydroxy-2,5-dimethyl-3-(2H)-furanone, 2-acetyl-1-pyrroline, and an unknown compound by 8-, 2-, 2-, 2-, 2-, and 2-fold in the treatment sample in comparison to the control sample, respectively (see **Figure 2**). These noted differences in aroma development were also supported by the sensory data, which indicated that the aged treatment sample was statistically lower in perceived stale intensity vs the aged control sample.

Table 5. Semiquantitative Values of *o*-aminoacetophenone in SMP[a]

sample	concentration (μg/kg SMP)
control, 0 day old	0.01
treatment (0.1% EC), 0 day old	ND[b]
control, 17 months	0.36
treatment (0.1% EC), 17 months	0.04

[a] Relative to internal standard (2-methyl-3-heptanone). [b] Not detected.

Furthermore, the aged treatment sample was not statistically different in stale intensity but notably was reported to be statistically lower in cooked intensity in comparison to the control 0 day old powder. Therefore, the addition of EC to milk powder was able to produce a SMP that could be aged for 17 months and remain not statistically different in stale perception as a traditional 0 day old milk powder. Polyphenols are commonly associated with bitter taste attributes of foods and beverages; however, none of the treatment milk powders were found to be statistically higher in bitter intensities (**Table 4**).

The well-documented stale flavor compound in milk powder, *o*-aminoacetophenone, was also quantified in the SMP powder samples and is shown in **Table 5**. The relative ratios of quantities of *o*-aminoacetophenone reported in the milk powder samples were also in agreement with the FD factors reported in **Tables 1** and **2**; however, the absolute values should be considered semiquantitative (relative to the recovery of the internal standard 2-methyl-3-heptanone).

Various other aroma-active compounds were identified in the fresh and aged milk powders, such as lipid degradation products, but no differences between of the control and treatment milk powders were reported (EC had no apparent effect on compound formation, or these compounds were intrinsic to the concentrated milk samples). δ-Decalactone was found in both the 0 day old

and the 17 month old milk powders, while three other lactones, γ-decalactone, γ-6-(*Z*)-dodecenolactone, and γ-dodecalactone, were only reported for the 17 month old powders. Lactones have been linked to the milky odor of milk powders (*33*).

The acidic fraction for the estimated 17 month old powder similarly did not report any clear differences in FD factors between the control and the treatment extracts (see **Table 3**). The 0 day old powder acidic fraction was not evaluated by AEDA as the concentration of each acidic aroma-active compound was found to be equivalent in concentration (based on GC analysis; data not shown) between the control and the treatment samples.

In conclusion, a polyphenol (EC) extracted from a natural product (green tea) was found to reduce the development of stale aroma compounds in SMP samples during processing and storage. Control of Maillard type reactions in processed food systems via phenolic reactivity may also impart additional benefits associated with health and wellness. For example, in model sugar−casein systems (dairy foods), the Maillard reaction has been linked to the formation of mutagens (*34*). Furthermore, medical/nutritional researchers have also extensively studied the negative effects of Maillard reaction products on protein structure and function [glycation or advanced glycosylated end products (AGE)] on age-related pathologies [see symposium proceedings (*35*) for a current overview]. Dietary sources of AGE precursors (i.e., "carbonyl stress") as well as AGEs have been linked to plasma AGE concentration and furthermore related to chronic disease (*36*, *37*). On the basis of the reported reactivity of EC with C_2, C_3, and C_4 sugar fragments (or carbonyls) by Totlani and Peterson (*9*), investigation into the application of EC (polyphenolics) to reduce the carbonyl load in processed foods for improved health (e.g., diabetes) may provide an important link between food chemistry and health and wellness.

ACKNOWLEDGMENT

We extend our gratitude to Land O' Lakes for generously supplying us with low-heat condensed skim milk for spray drying and low-heat SMP used in training our descriptive sensory panel.

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Received for review August 11, 2005. Revised manuscript received November 6, 2005. Accepted November 13, 2005.

JF0519764