



Effect of ultraviolet light on water- and fat-soluble vitamins in cow and goat milk

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ABSTRACT

The objective of this study was to investigate and compare the effects of UV light and heat treatment on vitamins A, B₂, C, and E in cow and goat milk. Vitamins were analyzed by reverse-phase high-pressure liquid chromatography. Ultraviolet and pasteurization treatments caused loss in vitamin C in milk. Pasteurization did not have any significant effect on vitamin B₂. However, UV light treatment decreased the amount of vitamin B₂ after several passes of milk through the UV system. In addition, UV light treatment decreased the amount of vitamins A and E. Vitamins C and E are more sensitive to UV light. UV light sensitivities of vitamins were C > E > A > B₂. These results show that UV light treatment decreases the vitamin content in milk. Also, the number of passes through the UV system and the initial amount of vitamins in milk are important factors affecting vitamin levels.

Key words: ultraviolet light, fat- and water-soluble vitamins, cow milk, goat milk

INTRODUCTION

For the production of milk and dairy products, heat treatment is the most common process to inhibit the growth of pathogens and spoilage microorganisms and to inactivate enzymes in milk. Three types of heat treatment to milk include thermization, pasteurization, and sterilization in the dairy industry (Raynal-Ljutovac et al., 2007). Pasteurization is widely used as a thermal process for milk. In pasteurization, milk is heated to 63°C for 30 min (low temperature, long time) or to 72°C for 15 s (HTST; Fox and McSweeney, 1998). Physical, chemical, and sensorial changes may occur during heat treatment of milk. Although some changes are favorable, others are considered undesirable, because they

cause adverse effects on sensory characteristics, nutritional values, and technological properties of milk (Fox and McSweeney, 1998; Lewis, 2003).

A loss of water- and fat-soluble vitamins in milk during the heating process has been reported (Holmes et al., 1945; Lavigne et al., 1989; Oamen et al., 1989; Sierra and Vidal-Valverde, 2001; Bendicho et al., 2002; Asadullah et al., 2010; Moltó-Puigmartí et al., 2011). Holmes et al. (1945) reported that HTST pasteurization (71–83°C, 22 s) of raw milk results in 18.7 and 3% of losses of vitamin C and B₁ (thiamin), respectively. No change to vitamin B₂ (riboflavin) content was observed. Lavigne et al. (1989) evaluated different pasteurization and sterilization processes in terms of changes to vitamins B₁, B₂, and C content in goat milk. Low-temperature, long-time (LTLT; 63.5°C for 30 min), HTST (76°C for 16 s), UHT (135°C for 4 s), and sterilization (121°C for 15 min) decreased the vitamin C level in goat milk at the rate of 40, 25, 30, and 70%, respectively. No significant decrease in vitamin B₂ content by HTST and UHT processing was observed. However, LTLT and sterilization decreased vitamin B₂ content by 20 and 30%, respectively. Asadullah et al. (2010) reported that household heating of milk (boiling about 100°C for 15 min) causes 27, 27, 29, 24, and 36.2% loss in vitamins B₁, B₂, B₃, B₆, and folic acid, respectively. Moltó-Puigmartí et al. (2011) investigated effects of LTLT pasteurization and high-pressure processing on ascorbic acid (vitamin C) and α - and δ -tocopherol isomers (vitamin E) in human milk. Low-temperature, long-time processing reduced vitamin C and α - and δ -tocopherol isomers in human milk at rates of 16.2, 0.97, and 2.08%, respectively. High-pressure processing did not significantly affect ascorbic acid and tocopherol levels.

Today, increasing consumer preference for high nutritional and quality food products have led to the development of novel techniques to process dairy products. Ultraviolet light treatment has commercial potential among nonthermal food technologies as an alternative to heat treatment (pasteurization) of liquid foods (Bintsis et al., 2000; Lopez-Malo and Palou, 2005; Koutchma, 2009). Ultraviolet light has been used for

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disinfection of surfaces, air, water, and equipment for over 60 yr. The UV light spectrum ranges from 100 to 400 nm. The range of 200 to 280 nm UV light spectrum (UV-C) has a germicidal effect on microorganisms, including bacteria, yeasts, molds, and viruses. Microbial inactivation in liquid foods by UV-C depends on optical and flow properties of the product, microbial load of the product, geometric configuration of the reactor, power, wavelength and physical arrangement of the UV source(s), and radiation path length (Koutchma et al., 2009). These critical factors of UV treatments for liquid food processing are discussed by Koutchma et al. (2009).

Several studies have been conducted on determining the effects of UV treatment on microbiological, physicochemical and sensory properties of clear liquid foods, such as fruit juices (Gachovska et al., 2008; Keyser et al., 2008; Franz et al., 2009; Guerrero-Beltrán et al., 2009; Fredericks et al., 2011; Uysal Pala and Kirca Toklucu, 2011; Caminiti et al., 2012). However, only a limited number of studies have been done on UV light applications in opaque liquid foods such as milk and liquid dairy products. Although UV light influence on pathogens and indicator microorganisms in milk has been a particular focus in these studies (Smith et al., 2002; Matak et al., 2005; Reinemann et al., 2006; Altic et al., 2007; Donaghy et al., 2009; Engin et al., 2009), its effects on nutritional properties of milk have not been assessed. Therefore, the aims of this research were to determine the effects of UV light and pasteurization on vitamins C, B₂, A, and E and to compare the vitamin losses by pasteurization and UV light treatment in milk from cows and goats.

MATERIALS AND METHODS

Milk

Goat milk samples ($n = 3$) were obtained from the Turkish Saanen breed in the late-lactation period (October 2010 to April 2011) at Canakkale Onsekiz Mart University-Technological and Agricultural Research Center (Canakkale, Turkey). Cow milk samples ($n = 4$) were obtained from local producers (May–July 2011) in Canakkale. One of the cow milk samples (C4) was used for the manipulation experiment. Milk samples were carried in thermo bag with an ice pack at 10°C and analyzed immediately in our laboratory.

Pasteurization

Milk samples were pasteurized in a 1-L glass jar in a thermostatic water bath [Gesellschaft für Labortechnik

(GFL) mbH, Großburgwedel, Germany] at 65°C for 30 min.

UV Light Treatment

A custom-made UV light system equipped with 9 UV-C type lamps (Gentra Stock Joint Co., Istanbul, Turkey) was used for UV treatment of milk (Engin and Karagul Yuceer, 2012). The UV reactor was designed by Yusuf Köprülü (Gentra Insaat ve Ticaret Ltd. Sti., Istanbul, Turkey). The UV unit consists of a stainless steel reflector, a corrugated Teflon tube coiled around quartz sleeve and 9 UV lamps (254 nm; 28 W UV-C output; the length of each lamp was 842.4 mm and the diameter was 16 mm). The UV light intensity per single pass through the reactor was calculated based on the flow rate of milk through the reactor and total output wattage of UV lamps (Engin and Karagul Yuceer, 2012) by using Equation 1, suggested by Geveke (2008) and Keyser et al. (2008):

$$\text{UV intensity (J/mL)} = \frac{\text{total UV-C output power (W)}}{\text{flow rate (mL/s)}} \quad [1]$$

Seven passes were applied to milk in the reactor to evaluate changes to vitamin content under UV light. Vitamin analysis was conducted after 1, 3, 5, and 7 passes. The beginning temperature of the milk was 18°C and was raised to 25°C after 7 passes through the system. The UV light intensities applied to milk samples are shown in Table 1.

Composition of Milk

The titratable acidity (lactic acid, %), pH, and DM (%), total protein (%), and ash (%) contents of the milk samples were determined according to procedures described by Bradley et al. (1992). The fat content of the milk samples was determined using a method by Gerber-Van Gulik (NEN, 1969).

Table 1. Ultraviolet light intensity applied to cow and goat milk

UV treatment	UV intensity (J/mL of milk)	
	Cow milk	Goat milk
First pass	12.6	11.72
Third pass	37.8	35.16
Fifth pass	63.0	58.60
Seventh pass	88.2	82.04

Water-Soluble Vitamins

Determination of Vitamin C Content. The ascorbic acid content of milk samples was determined by using the HPLC method of Romeu-Nadal et al. (2006) with minor modifications. Five milliliters of milk and 5 mL of metaphosphoric acid (5.6%) were poured into a 15-mL Falcon tube. The mixture was vortexed for 1 min in a dark room and centrifuged (2-12 K; Sigma, Göttingen, Germany) for 15 min at $1,000 \times g$ at 10°C . Then, the clear liquid phase was filtered through 0.45- μm polytetrafluoroethylene (PTFE) filter and 5 μL of filtrate was directly injected into the HPLC system. Separation of ascorbic acid in milk was conducted using the Agilent 1200 series HPLC system and Agilent LiCrospher RP 60 Select B (5- μm) column (Agilent Technologies Inc., Folsom, CA). The mobile phases were acetic acid (0.1%, vol/vol) in water and methanol (95:5). The eluent flow rate was isocratic 0.7 mL/min and the column temperature was 25°C . Vitamin C was identified by comparison of retention time of pure vitamin C standard at 254 nm. Quantification of vitamin C in cow milk was conducted using the external standard method, whereas that in goat milk was achieved by the addition method due to peak interactions with other substances in the chromatogram. The repeatability of the method was 0.56 relative standard deviation (RSD%). For measuring vitamin C in cow milk, the limit of detection (LOD) and limit of quantification (LOQ) were 0.27/ μg mL and 0.82 $\mu\text{g}/\text{mL}$, respectively. The recovery was 107.2% for vitamin C. Standard and sample chromatograms for vitamin C are shown in Figure 1.

Determination of Vitamin B₂ Content. The vitamin B₂ content of milk samples was determined using the HPLC method of Muñoz et al., (1994), with minor modifications. For sample preparation, 25 mL of milk was poured into a 50-mL Falcon tube; 2.5 mL of lead acetate solution (10%, pH adjusted to 3.2 using glacial acetic acid) was added, and the mixture was vortexed for 3 min and centrifuged (2-12 K; Sigma) for 15 min at $8,600 \times g$ at 4°C . The clear liquid phase was filtered through a 0.22- μm PTFE filter, and 10 μL of filtrate was directly injected into the HPLC system. All samples were prepared in a dark room to eliminate the effects of light. The mobile phase contained water-acetic acid (1.5 mL of acetic acid in 1 L of water) and methanol (60:40). The flow rate was 0.6 mL/min. Ultraviolet detection was at 270 nm, and the column temperature was 20°C . Riboflavin was identified by comparing the retention time of riboflavin standard provided and quantified using external standard methods. The repeatability of the method was 0.39 (RSD%). The LOD and LOQ were 0.01 $\mu\text{g}/\text{mL}$ and 0.18 $\mu\text{g}/\text{mL}$,

respectively. The recovery was 102.84% for vitamin B₂. Standard and sample chromatograms for vitamin B₂ are shown in Figure 1.

Fat-Soluble Vitamins

Determination of Vitamin A and E Content. Vitamins A and E in milk were extracted using the liquid-liquid extraction method suggested by Zahar and Smith (1990) and modified by Kondyli et al., (2007). Two milliliters of milk was poured into a 50-mL Falcon tube, and 5 mL of absolute ethanol (include 0.1% ascorbic acid) and 2 mL of 50% (wt/vol) KOH were added. Following a 2-min vortexing, the tube was placed in a water bath (GFL GmbH) for saponification at 80°C for 20 min. Then, the tube was cooled. Ten milliliters of petroleum ether:diethyl ether mixture (1:1) containing 0.01% butylated hydroxytoluene was added. The tube was vortexed for 1 min and allowed to stand for 2 min. Then, 10 mL of petroleum ether:diethyl ether mixture was added. The tube was vortexed for 2 min and allowed to stand for 5 min. Fifteen milliliters of cold water ($+4^{\circ}\text{C}$) was added to the tube, which was then shaken to disperse the content. The tube was centrifuged at $2,000 \times g$ for 15 min. The upper phase was transferred into a 25-mL rotary evaporating flask and the solvent was removed under vacuum at 40°C using a rotary evaporator (Heidolph Instruments GmbH & Co. KG, Schwabach, Germany). The residue was dissolved in 1 mL of methanol. The extract was filtered through a 0.45- μm PTFE filter and collected in an amber vial (Zahar and Smith, 1990; Kondyli et al., 2007). Injection volumes of the extracts were 25 and 50 μL for vitamin E and A, respectively. Separation of vitamins E and A in milk was conducted using the Agilent 1200 series HPLC system and Agilent Zorbax Eclipse XDB C18 column (5 μ , 4.6 mm \times 150 mm; Agilent Technologies Inc.) at 30°C . Methanol (100%) at a flow rate of 1.0 mL/min was used as a mobile phase for the separation of vitamin E. Ultraviolet detection was performed at 292 nm. For vitamin A, the mobile phases were water and methanol (90:10) and the flow rate was 1.2 mL/min. Ultraviolet detection was performed at 323 nm. Vitamins A and E were identified by comparing retention times of their standards and quantified using the external standards methods. Repeatability for vitamin A method was 0.18 (RSD%). The LOD and LOQ were 0.76 and 2.31 $\mu\text{g}/\text{mL}$, respectively. The repeatability for the vitamin E method was 0.18 (RSD%) and the LOD and LOQ were 0.88 and 2.67 $\mu\text{g}/\text{mL}$, respectively. The recovery was 82.07% for vitamin A and 81.86% for vitamin E. Standard and sample chromatograms of vitamins A and E are shown in Figures 2a and 2b, respectively.

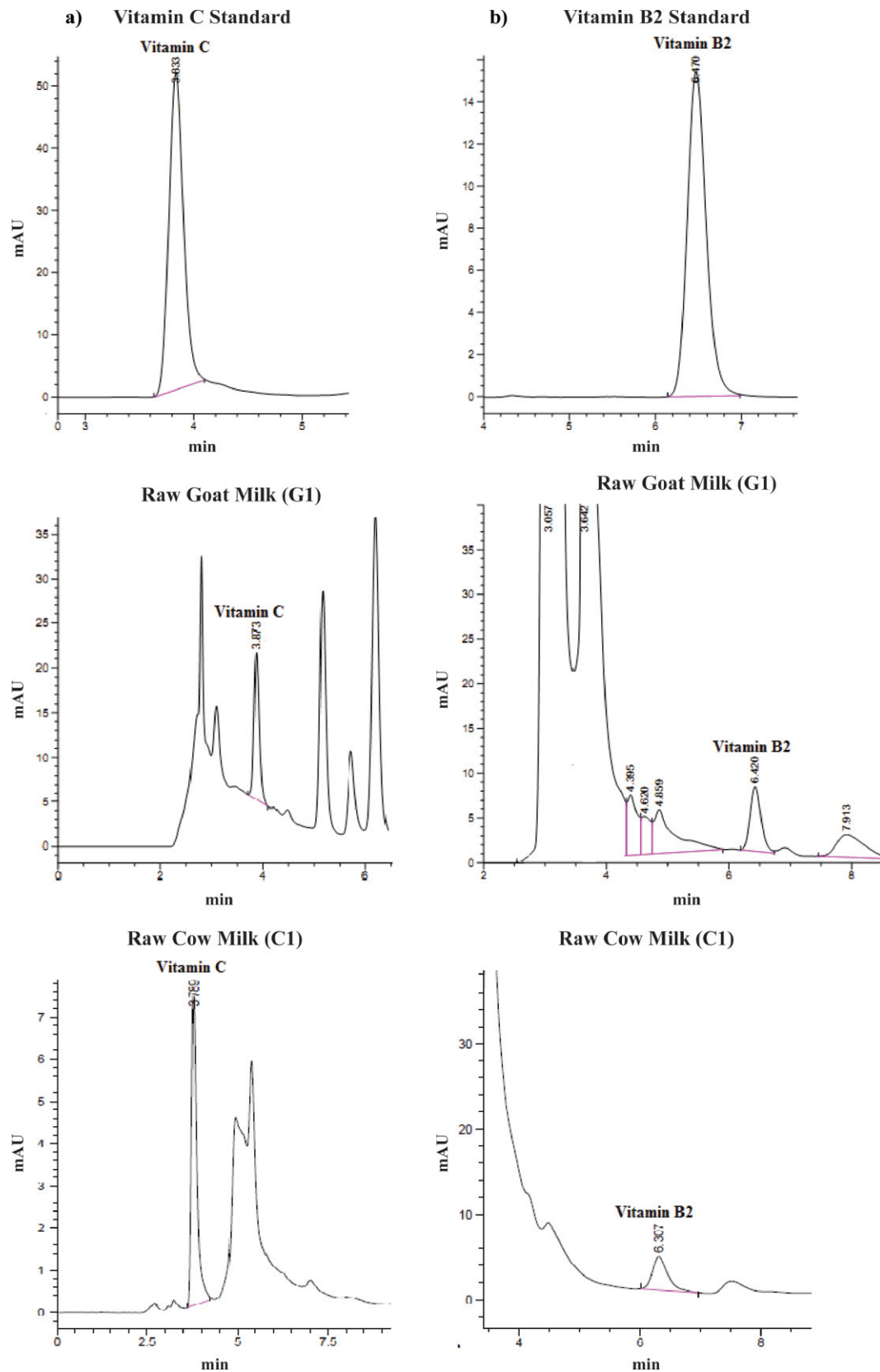


Figure 1. Chromatograms of (a) vitamin C standard and raw milk samples; and (b) vitamin B₂ standard and raw milk samples. Color version available in the online PDF.

Manipulation Experiment

The effect of UV light was also investigated by adding vitamin standards at certain levels to milk. Vitamins A, B₂, C, and E were added to milk at 4, 3, 40, and 3.5 mg/L, respectively. The same procedures were followed to determine vitamin contents in cow and goat milk samples.

Chemicals and Reagents

All chemicals, reagents, and vitamin standards used in the HPLC analysis were HPLC grade and obtained from Aldrich Chemical Co. (St. Louis, MO) and Merck KGaA (Darmstadt, Germany).

Statistical Analysis

Analysis of variance was conducted to determine the differences in vitamin content among milk samples that were applied UV treatment and pasteurization. The ANOVA model is shown in Equation 2:

$$Y_{ij} = \mu + \alpha_i + e_{ij}, \quad [2]$$

where Y_{ij} is the j th observation value in the i th milk, μ is the general population mean, α_i is the effect of the i th milk, and e_{ij} represents the random error term (Sheskin, 2004). The Tukey honestly significant differences (HSD) test was used for separating means; SPSS for Windows (version 15.0) was used for all statistical analyses (SPSS Inc., 2006).

RESULTS AND DISCUSSION

Composition of Milk

The compositions of cow and goat milk are shown in Table 2. Significant differences were observed in cow milk samples in terms of lactic acid, DM, and fat content ($P < 0.05$). However, no significant differences were detected in protein and ash contents of the samples ($P > 0.05$).

The highest DM and fat contents were observed in sample C1. Sample C3 had the highest lactic acid content, whereas the lowest lactic acid content was observed in sample C2. The fat, DM, and lactic acid contents of cow milk samples were 3.05 to 4.20%, 11.71 to 13.21%, and 0.14 to 0.23%, respectively (Table 2). These results are consistent with the findings of Amenu et al. (2006) and Ozrenk and Inci (2008). The differences in the composition of cow milk that was obtained at different lactation times may be related to the lactation period, feeding strategy of the cow, and feed

composition (Fox and McSweeney, 1998). In a study by Ozrenk and Inci (2008), significant changes in fat, total DM, and protein content of cow milk that was obtained from 12 different locations in Van, Turkey were determined during the lactation period. As expected, they observed that the DM, protein, and fat contents in milk collected in the winter season (January–March) were higher than in milk collected in the summer (June–August). Lindmark-Månsson et al. (2003) investigated seasonal changes in the composition of cow milk obtained from 9 different milk plants in Sweden. They found that the pH and the protein, lactose, ash, water, and fat-soluble vitamin (except biotin) contents of milk changed significantly through seasons, except that no significant changes in urea, fat, and DM content of milk were observed.

Significant differences were observed in goat milk samples obtained on different days ($P < 0.05$), except in fat content. The highest DM, total protein, ash, and lactic acid contents were observed in sample G2. Sample G3 had the lowest DM and protein content (Table 2). The total DM, lactic acid, and fat content of goat milk samples were 11.38 to 13.79%, 0.14 to 0.19%, and 4.00 to 5.50%, respectively. Güneşer et al. (2010) investigated the chemical composition and FA profiles of 4 different goat breeds in Canakkale. They determined that the lactic acid, DM, and fat contents of Saanen milk were 0.12, 10.29, and 3.25%, respectively. The differences in lactic acid, DM, total protein, and ash contents of goat milk samples can be attributed to changes in milk composition during the lactation period of goats. Thus, changes in goat milk composition based on factors such as breed, feeding, and lactation time, among others, were reported in many studies (Borges et al., 2004; Bhosale et al., 2009; Strzałkowska et al., 2009; Güzeler et al., 2010). Güzeler et al. (2010) identified significant changes in the DM, fat, total protein, and lactose content, energy values, acidity, pH, specific gravity, and sodium content of milk from Saanen × Kilis goat breeds during the lactation period. Strzałkowska et al. (2009) found a significant increase in density, acidity, and total protein, DM, urea, and FFA content of Polish white improved goat milk during the late-lactation period of goats.

Water-Soluble Vitamins

Table 3 shows the treatment effect on vitamins B₂ and C for each milk sample. The effect of UV light treatment was significant on vitamins B₂ and C in cow milk samples. The reduction in vitamin C content of milk samples upon UV light treatment is dependent on initial vitamin C content. No significant difference was observed in the vitamin C content of raw and pasteur-

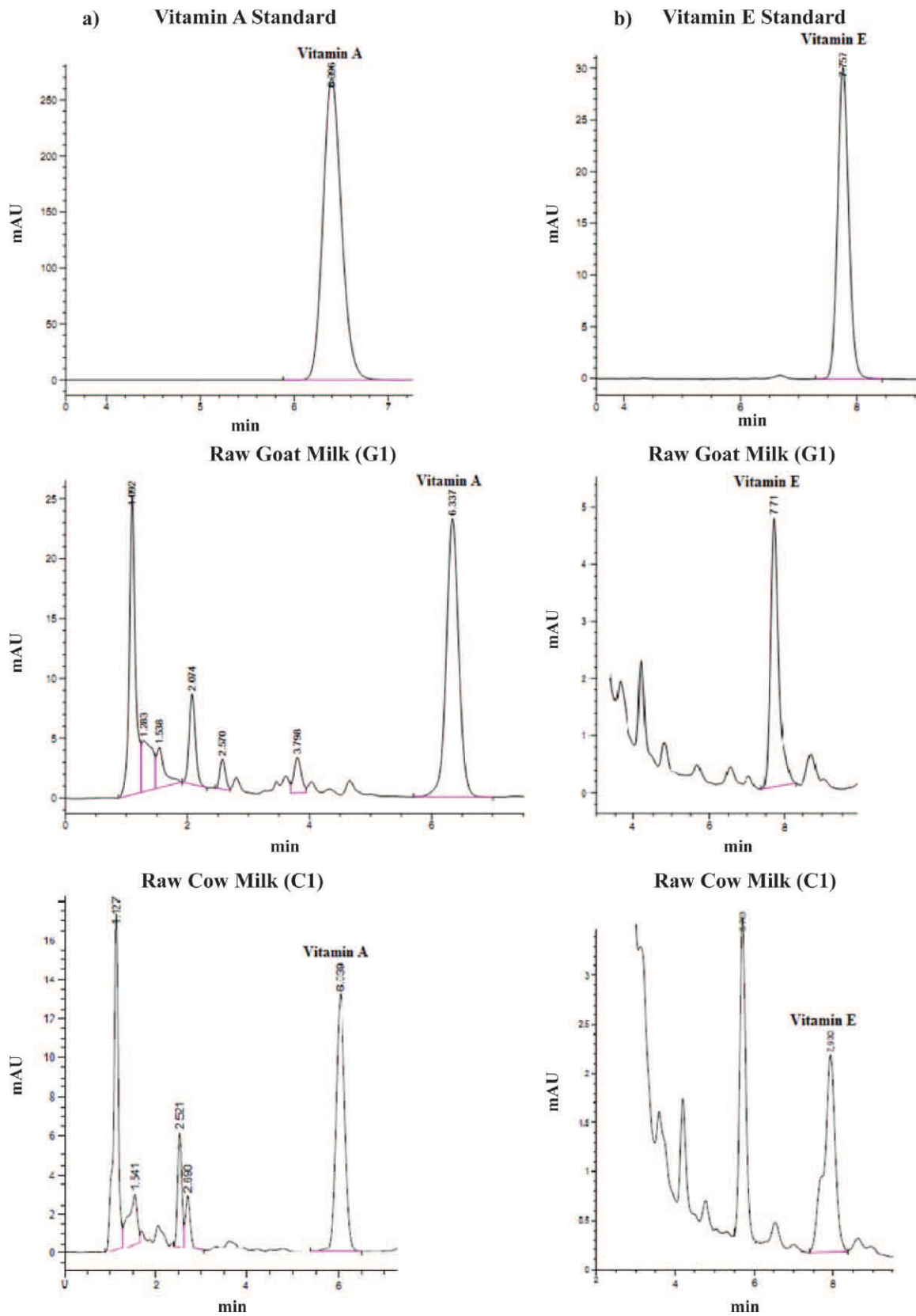


Figure 2. Chromatograms of (a) vitamin A standard and raw milk samples; and (b) vitamin E standard and raw milk samples. Color version available in the online PDF.

Table 2. Composition of cow and goat milk

Milk sample ¹	Chemical property				
	Lactic acid (%)	Total DM (%)	Fat (%)	Total protein (%)	Ash (%)
Cow milk					
C1	0.18 ± 0.01 ^b	13.21 ± 0.03 ^a	4.20 ± 0.01 ^a	1.91 ± 0.08	0.67 ± 0.01
C2	0.14 ± 0.01 ^c	11.71 ± 0.01 ^c	3.05 ± 0.05 ^b	2.13 ± 0.13	0.64 ± 0.01
C3	0.23 ± 0.01 ^a	11.89 ± 0.01 ^c	3.40 ± 0.01 ^b	1.89 ± 0.05	0.64 ± 0.02
C4	0.21 ± 0.01 ^{ab}	12.38 ± 0.08 ^b	3.37 ± 0.12 ^b	2.17 ± 0.09	0.64 ± 0.01
Goat milk					
G1	0.16 ± 0.01 ^b	12.46 ± 0.10 ^b	4.60 ± 0.40	2.73 ± 0.02 ^b	0.85 ± 0.04 ^{ab}
G2	0.19 ± 0.01 ^a	13.79 ± 0.16 ^a	5.50 ± 0.50	2.88 ± 0.02 ^a	1.00 ± 0.01 ^a
G3	0.14 ± 0.01 ^b	11.38 ± 0.01 ^c	4.00 ± 0.01	2.52 ± 0.02 ^c	0.80 ± 0.01 ^b

^{a-c}Means followed by different superscript letters represent significant differences within species for each chemical property ($P < 0.05$).

¹C1, C2, and C3 = cow milk (C1: May 2011; C2: June 2011, C3 and C4: July 2011); G1, G2, and G3 = goat milk (G1: September 2010, G2: October 2010, G3: April 2011).

ized cow milk samples. Three passes of milk in the UV system caused a loss of 78 and 91% of vitamin C in the C2 and C3 samples, respectively. One-pass UV treatment caused a loss of 74% of vitamin C in C1 milk. Vitamin C was not detected in C1 milk after 3, 5, and 7 passes from the system. No significant differences were observed in 3-, 5-, and 7-pass-treated C2 milk samples in terms of vitamin C. The vitamin B₂ content of cow milk was decreased by UV light treatment. No significant changes were observed in the content of vitamin B₂ in C1, C2, and C3 samples due to pasteurization. Moreover, no significant differences were observed in the vitamin B₂ content of raw, pasteurized, and 1-, 3-, and 5-pass UV light-treated C1 and C2 milk samples ($P < 0.05$). Seven-pass UV treatment significantly decreased the vitamin B₂ content of C1, C2, and C3 milk samples by 29, 20, and 31%, respectively (Table 3).

Significant differences were observed in the vitamin C content of raw, pasteurized and UV light-treated goat milk samples ($P < 0.05$). Pasteurization decreased the vitamin C content at the rate of 35, 35, and 47% in G1, G2, and G3 milk samples respectively, whereas 75% (G1), 91% (G2), and 87% (G3) vitamin C losses in goat milk samples were observed upon treatment with 1-pass UV light. Vitamin C was not detected in the G3 sample after 3-pass UV light treatment. Similar to cow milk, UV light treatment was more effective in decreasing the vitamin C content in goat milk when compared with pasteurization. No significant differences were observed in 3-, 5-, and 7-pass UV light treatments of both G1 and G2 for vitamin C. Pasteurization did not show a significant effect on vitamin B₂ in goat milk; however, the vitamin B₂ content in goat milk was decreased by UV light treatment. No loss of vitamin B₂ content in G1, G2, and G3 milk samples due to pasteurization was observed. The loss of 11 and 7% of vitamin B₂ was observed after 3 passes of G1 and G2 milk samples in the UV system, respectively. No significant differ-

ences were observed in the vitamin B₂ content of raw, pasteurized and 1-, 3-, and 5-pass UV light-treated G3 milk samples. Seven passes from the system decreased the vitamin B₂ content of sample G3 by 23% (Table 3).

Vitamins C, B₁₂, B₆, B₂, and folic acid are light-sensitive, water-soluble vitamins. Vitamin C highly absorbs UV light at 254 nm; therefore, degradation of vitamin C by UV light is dependent on the absorption coefficient of foods (Koutchma et al., 2009). The degradation rate of vitamin C was 8 times faster in clarified apple juice than orange juice due to the greater level of absorbed energy (Koutchma et al., 2009). Koutchma (2010) evaluated the degradation of vitamin C in milk exposed to 2 different UV light sources. The researcher found a 35% decrease in vitamin C content caused by a low-pressure mercury UV lamp, whereas 26% (PUV-1, 31 J), 35% (PUV-2, 344 J), and 24% (PUV-3, 644 J) reductions in vitamin C in milk were observed after treatments with 3 pulsed UV lamps with various energies (PUV-1, 2, and 3). We found a high vitamin C degradation rate in goat and cow milk. Differences between findings of that study and our results may be attributed to the type of UV light source and UV reactor design, as well as UV procedure. Cakmakçı and Turgut (2005) investigated the effects of various light sources and illumination intensities on degradation of vitamin C in pasteurized milk during storage. It was found that a fluorescent light source was more effective on loss of vitamin C in cow milk when compared with a tungsten light source. More vitamin C was lost in milk upon increasing light intensity. The effects of artificial light on ascorbic acid and oxidized flavor in homogenized and unhomogenized cow milk during 4-d storage were investigated by Smith and Macleod (1955). They found that fluorescent light with 193.75 lx light intensity decreased the initial vitamin C content of homogenized milk by 97% after 4 d of cold storage. However, the vitamin C content of milk was decreased

Table 3. Changes in water-soluble vitamins (means \pm SE) in cow and goat milk¹

Treatment	Vitamin C (mg/L)									Vitamin B ₂ (mg/L)								
	Cow milk sample			Goat milk sample			Cow milk sample			Goat milk sample			Cow milk sample			Goat milk sample		
	C1	C2	C3	G1	G2	G3	C1	C2	C3	G1	G2	G3	C1	C2	C3	G1	G2	G3
Raw milk	23.12 \pm 0.46 ^A	19.41 \pm 1.25 ^A	33.91 \pm 0.95 ^A	21.40 \pm 0.69 ^A	17.77 \pm 1.59 ^A	12.36 \pm 0.82 ^A	0.87 \pm 0.04 ^A	0.64 \pm 0.01 ^A	0.93 \pm 0.03 ^A	1.18 \pm 0.01 ^A	0.99 \pm 0.01 ^A	0.53 \pm 0.01 ^A	0.87 \pm 0.04 ^A	0.64 \pm 0.01 ^A	0.93 \pm 0.03 ^A	1.18 \pm 0.01 ^A	0.99 \pm 0.01 ^A	0.53 \pm 0.01 ^A
Pasteurized milk	12.94 \pm 3.04 ^{AB}	17.63 \pm 2.75 ^A	27.08 \pm 1.82 ^A	13.91 \pm 0.54 ^B	11.44 \pm 0.49 ^B	6.52 \pm 0.63 ^B	0.78 \pm 0.01 ^{ab}	0.64 \pm 0.01 ^A	0.92 \pm 0.02 ^A	1.14 \pm 0.01 ^A	0.95 \pm 0.01 ^{ab}	0.52 \pm 0.01 ^A	0.78 \pm 0.01 ^{ab}	0.64 \pm 0.01 ^A	0.92 \pm 0.02 ^A	1.14 \pm 0.01 ^A	0.95 \pm 0.01 ^{ab}	0.52 \pm 0.01 ^A
UV, 1 pass	5.90 \pm 0.61 ^B	10.58 \pm 3.08 ^{AB}	13.71 \pm 2.26 ^B	5.37 \pm 0.03 ^C	1.53 \pm 0.22 ^C	1.62 \pm 0.83 ^C	0.78 \pm 0.06 ^{ab}	0.62 \pm 0.01 ^A	0.84 \pm 0.02 ^B	1.17 \pm 0.03 ^A	0.95 \pm 0.01 ^{ab}	0.52 \pm 0.01 ^A	0.78 \pm 0.06 ^{ab}	0.62 \pm 0.01 ^A	0.84 \pm 0.02 ^B	1.17 \pm 0.03 ^A	0.95 \pm 0.01 ^{ab}	0.52 \pm 0.01 ^A
UV, 3 passes	ND	4.20 \pm 0.04 ^B	2.99 \pm 0.06 ^C	0.77 \pm 0.74 ^D	0.60 \pm 0.01 ^C	ND	0.73 \pm 0.04 ^{ab}	0.57 \pm 0.03 ^{ab}	0.77 \pm 0.77 ^C	1.05 \pm 0.01 ^B	0.92 \pm 0.01 ^B	0.47 \pm 0.02 ^{ab}	0.73 \pm 0.04 ^{ab}	0.57 \pm 0.03 ^{ab}	0.77 \pm 0.77 ^C	1.05 \pm 0.01 ^B	0.92 \pm 0.01 ^B	0.47 \pm 0.02 ^{ab}
UV, 5 passes	ND	1.80 \pm 0.01 ^B	ND	0.73 \pm 0.01 ^D	0.47 \pm 0.02 ^C	ND	0.66 \pm 0.03 ^{ab}	0.55 \pm 0.02 ^{ab}	0.68 \pm 0.11 ^d	1.04 \pm 0.01 ^B	0.85 \pm 0.01 ^C	0.45 \pm 0.02 ^{ab}	0.66 \pm 0.03 ^{ab}	0.55 \pm 0.02 ^{ab}	0.68 \pm 0.11 ^d	1.04 \pm 0.01 ^B	0.85 \pm 0.01 ^C	0.45 \pm 0.02 ^{ab}
UV, 7 passes	ND	1.66 \pm 0.01 ^B	ND	0.80 \pm 0.01 ^D	0.48 \pm 0.07 ^C	ND	0.62 \pm 0.04 ^B	0.51 \pm 0.01 ^B	0.64 \pm 0.14 ^c	0.97 \pm 0.01 ^B	0.81 \pm 0.01 ^D	0.41 \pm 0.01 ^B	0.62 \pm 0.04 ^B	0.51 \pm 0.01 ^B	0.64 \pm 0.14 ^c	0.97 \pm 0.01 ^B	0.81 \pm 0.01 ^D	0.41 \pm 0.01 ^B

^{A-D}Means followed by different superscripts represent significant differences within the same sample for vitamin C content ($P < 0.05$).

^{a-c}Means followed by different superscripts represent significant differences within the same sample for vitamin B₂ content ($P < 0.05$).

¹C1, C2, and C3 = cow milk; G1, G2, and G3 = goat milk; ND = not detected.

by 87% under incandescent (tungsten) light with the same light intensity. In another study, a loss of 87% of vitamin C in goat milk in a white glass bottle exposed to sunlight for 5 h was reported (Jandal, 1996).

Vitamin B₂ is another major vitamin in milk and dairy products. Although vitamin B₂ is heat stable, it is more sensitive to light. The degradation of B₂ depends on the intensity and wavelength of light (Muñoz et al., 1994; Choe et al., 2005). Vitamin B₂ acts as a photosensitizer compound in milk. Photosensitization of riboflavin results in the formation of superoxide anion radicals, singlet oxygen, hydroxy radical, and hydrogen peroxide. These compounds may cause nutrient losses in foods, including degradation of vitamin A in milk (Choe et al., 2005). Many studies were conducted to determine the degradation rate of vitamin B₂ in milk under different light conditions (Maniere and Dimick, 1976; Allen and Parks, 1979; Gaylord et al., 1986). Allen and Parks (1979) investigated the degradation kinetics of vitamin B₂ in milk exposed to fluorescent light with 2,690 lx light intensity. The same researchers found that the degradation kinetics of vitamin B₂ in whole and skim milk were followed the first-order kinetics (Allen and Parks, 1979). It was also found that the vitamin B₂ contents of whole and skim milk were decreased by 87.9 and 76.12%, respectively, when 32 h of fluorescent light was applied. Maniere and Dimick (1976) reported 90.4% loss in the vitamin B₂ amount in homogenized milk that was exposed to fluorescent light at 2,150 lx intensity for 48 h at 7°C. Gaylord et al. (1986) evaluated the effect of milk fat, milk solid, and light intensity on the stability of B₂. It was found that the stability of vitamin B₂ increased by increasing the fat content of milk. They found that the loss of vitamin B₂ in whole milk, 2% fat milk, and skim milk was 55, 64, and 70%, respectively, after 48 h of fluorescent light exposure at 1,614 lx. Saffert et al. (2006) reported 33% loss in vitamin B₂ in pasteurized whole milk (3% fat) that was stored under fluorescent light with 1,700 lx intensity at 8°C in clear 1-L polyethylene terephthalate bottles for 10 d.

Fat-Soluble Vitamins

Changes in vitamin A and E in cow and goat milk samples are shown in Table 4. The effect of UV light treatment was significant on vitamin A and E in cow milk ($P < 0.05$). It was found that several passes of milk in the UV light unit decreased the vitamin A content of cow milk. However, pasteurization did not show a significant effect on vitamin A in cow milk.

No significant differences were observed in vitamin A content of raw, pasteurized, and 1- and 3-pass UV light-treated C1, C2, and C3 milk samples. Therefore,

pasteurization and UV light treatment up to 3 passes had the same effect on degradation of vitamin A in cow milk. Although no loss in vitamin A content in C1, C2, and C3 milk samples due to pasteurization was observed, a 30, 32, and 32% decrease in vitamin A in C1, C2, and C3 milk samples, respectively, was observed after 7 passes. Ultraviolet light treatment decreased the vitamin E content in cow milk, but pasteurization did not show a significant effect on vitamin E in cow milk. No significant differences were observed in the vitamin E content of raw, pasteurized, 1-pass UV-treated C1, C2, and C3 milk samples. Seven-pass UV treatment decreased the vitamin E content of samples C1, C2, and C3 by 66, 69, and 72%, respectively.

The effect of UV light treatment was significant on vitamin A and E in goat milk ($P < 0.05$; Table 4). It was found that 7-pass UV light treatment had an adverse effect on the vitamin A content in goat milk, but pasteurization did not have a significant effect on vitamin A in goat milk. No significant differences were observed in the vitamin A content of raw, pasteurized, and 1-pass and 3-pass UV light-treated G1, G2, and G3 milk samples. It was found that 7-pass UV treatment decreased the vitamin A content of G1 and G2 samples by 19 and 30%, respectively, and 29% of vitamin A in G3 milk was lost by 7 passes. Similar to cow milk, the extent to which the vitamin E content of goat milk was affected by UV light treatment depended on the initial vitamin E content of the goat milk. Pasteurization did not cause any significant decrease in vitamin E content of G2 and G3 samples. The vitamin E content of G1 was decreased by pasteurization by 22%. It was found that the vitamin E content of G1 and G3 milk samples was reduced by 24 and 66%, respectively, after 7 passes in the system (Table 4).

Vitamin A is a derived form of carotenoids (principally β -carotene) and found in milk naturally as retinyl esters. Vitamin A, as well as riboflavin, is photosensitive in milk. Therefore, when milk is exposed to light, degradation in vitamin A occurs by photoisomerization (deMan, 1981; Miller et al., 2007). Degradation of vitamin A in milk due to light exposure was also investigated in several studies (deMan, 1981; Gaylord et al., 1986; Chapman et al., 1998; Whited et al., 2002). deMan (1981) investigated the loss of vitamin A in whole, 2% fat milk and skim milk exposed to fluorescent light with 2,200 lx intensity for 48 h at refrigerated temperatures. The researcher reported the loss of 32.3, 23.6, and 95.8% of vitamin A in whole milk, 2% fat milk, and skim milk, respectively, after 30 h of exposure. In another study (Gaylord et al., 1986), the losses by 43, 47, and 55% in vitamin A in whole milk, 2% fat milk, and skim milk after 48-h exposure to fluorescent light with 1,614 lx intensity were reported. Chapman et al.

Table 4. Changes in fat-soluble vitamins (means \pm SE) in cow and goat milk¹

Treatment	Vitamin A (mg/L)									Vitamin E (mg/L)									
	Cow milk sample			Goat milk sample			Cow milk sample			Goat milk sample			Cow milk sample			Goat milk sample			
	C1	C2	C3	G1	G2	G3	C1	C2	C3	G1	G2	G3	C1	C2	C3	G1	G2	G3	
Raw milk	0.40 $\pm 0.02^A$	0.22 $\pm 0.01^A$	0.25 $\pm 0.01^A$	0.70 $\pm 0.01^A$	0.46 $\pm 0.01^A$	0.45 $\pm 0.04^A$	1.66 $\pm 0.02^A$	1.14 $\pm 0.01^A$	1.55 $\pm 0.01^A$	1.65 $\pm 0.05^A$	1.73 $\pm 0.05^A$	0.96 $\pm 0.04^A$	0.40 $\pm 0.02^A$	0.22 $\pm 0.01^A$	0.25 $\pm 0.01^A$	0.70 $\pm 0.01^A$	0.46 $\pm 0.01^A$	0.45 $\pm 0.04^A$	1.66 $\pm 0.02^A$
Pasteurized milk	0.33 $\pm 0.03^A$	0.22 $\pm 0.01^A$	0.26 $\pm 0.01^A$	0.73 $\pm 0.13^A$	0.46 $\pm 0.01^A$	0.41 $\pm 0.01^{AB}$	1.36 $\pm 0.04^A$	0.98 $\pm 0.05^{ab}$	1.52 $\pm 0.06^A$	1.28 $\pm 0.01^b$	1.86 $\pm 0.01^a$	0.76 $\pm 0.08^{ab}$	0.33 $\pm 0.01^{AB}$	0.22 $\pm 0.01^B$	0.26 $\pm 0.01^{AB}$	0.73 $\pm 0.13^A$	0.46 $\pm 0.01^B$	0.41 $\pm 0.01^{AB}$	1.36 $\pm 0.04^A$
UV, 1 pass	0.35 $\pm 0.01^{AB}$	0.20 $\pm 0.01^{AB}$	0.23 $\pm 0.02^{AB}$	0.69 $\pm 0.01^A$	0.44 $\pm 0.01^A$	0.41 $\pm 0.01^{AB}$	1.12 $\pm 0.25^{ab}$	0.93 $\pm 0.07^{abc}$	1.27 $\pm 0.13^{ab}$	1.71 $\pm 0.01^a$	1.71 $\pm 0.07^a$	0.50 $\pm 0.07^{bc}$	0.35 $\pm 0.01^{AB}$	0.20 $\pm 0.01^B$	0.17 $\pm 0.01^B$	0.73 $\pm 0.13^{bc}$	0.46 $\pm 0.10^{ab}$	0.33 $\pm 0.01^b$	1.12 $\pm 0.25^{ab}$
UV, 3 passes	0.32 $\pm 0.01^{AB}$	0.18 $\pm 0.01^{AB}$	0.22 $\pm 0.01^{AB}$	0.68 $\pm 0.01^A$	0.44 $\pm 0.01^A$	0.36 $\pm 0.01^{ABC}$	0.70 $\pm 0.03^{bc}$	0.58 $\pm 0.13^{bcd}$	0.88 $\pm 0.13^{bc}$	1.70 $\pm 0.01^a$	1.67 $\pm 0.01^a$	0.55 $\pm 0.01^{bc}$	0.29 $\pm 0.01^{AB}$	0.16 $\pm 0.01^B$	0.20 $\pm 0.01^{AB}$	0.61 $\pm 0.11^c$	0.37 $\pm 0.08^{cd}$	0.32 $\pm 0.10^{cd}$	0.70 $\pm 0.01^a$
UV, 5 passes	0.29 $\pm 0.01^{AB}$	0.16 $\pm 0.01^B$	0.20 $\pm 0.01^{AB}$	0.61 $\pm 0.01^B$	0.37 $\pm 0.01^{AB}$	0.32 $\pm 0.01^{BC}$	0.54 $\pm 0.11^c$	0.53 $\pm 0.08^{cd}$	0.73 $\pm 0.03^c$	1.46 $\pm 0.10^{ab}$	1.32 $\pm 0.02^b$	0.37 $\pm 0.05^c$	0.28 $\pm 0.01^B$	0.15 $\pm 0.01^B$	0.17 $\pm 0.01^B$	0.57 $\pm 0.02^B$	0.32 $\pm 0.04^B$	0.26 $\pm 0.03^C$	1.46 $\pm 0.10^{ab}$
UV, 7 passes	0.28 $\pm 0.01^B$	0.15 $\pm 0.01^B$	0.17 $\pm 0.01^B$	0.57 $\pm 0.02^B$	0.32 $\pm 0.04^B$	0.26 $\pm 0.03^C$	0.56 $\pm 0.01^c$	0.35 $\pm 0.03^d$	0.46 $\pm 0.03^c$	1.25 $\pm 0.10^b$	1.10 $\pm 0.01^b$	0.33 $\pm 0.01^c$	0.28 $\pm 0.01^B$	0.15 $\pm 0.01^B$	0.17 $\pm 0.01^B$	0.57 $\pm 0.02^B$	0.32 $\pm 0.04^B$	0.26 $\pm 0.03^C$	1.25 $\pm 0.10^b$

^{A-C}Means followed by different superscripts represent significant differences within the same sample for vitamin A content ($P < 0.05$).

^{a-d}Means followed by different superscripts represent significant differences within the same sample for vitamin E content ($P < 0.05$).

¹C1, C2, and C3 = cow milk; G1, G2, and G3 = goat milk.

Table 5. Changes in water- and fat-soluble vitamins (means \pm SE) in manipulated milk (C4; mg/L of milk)

Milk sample	Vitamin A	Vitamin B ₂	Vitamin C	Vitamin E
Raw milk (C4)	0.15 \pm 0.01 ^f	1.76 \pm 0.05 ^c	16.63 \pm 0.04 ^d	0.80 \pm 0.01 ^g
Vitamin-added raw milk	3.41 \pm 0.01 ^a	4.90 \pm 0.01 ^a	61.15 \pm 0.71 ^a	3.52 \pm 0.06 ^a
Vitamin-added pasteurized milk	2.94 \pm 0.01 ^b	5.02 \pm 0.27 ^a	55.92 \pm 0.97 ^b	3.27 \pm 0.02 ^b
UV, 1 pass	2.93 \pm 0.01 ^b	4.85 \pm 0.29 ^a	49.99 \pm 0.89 ^c	2.87 \pm 0.01 ^c
UV, 3 passes	2.89 \pm 0.01 ^c	4.70 \pm 0.19 ^a	8.96 \pm 0.01 ^e	2.70 \pm 0.02 ^d
UV, 5 passes	2.86 \pm 0.01 ^d	4.57 \pm 0.22 ^{ab}	1.65 \pm 0.01 ^f	2.41 \pm 0.04 ^e
UV, 7 passes	2.57 \pm 0.01 ^e	3.58 \pm 0.06 ^b	1.48 \pm 0.01 ^f	1.57 \pm 0.01 ^f

^{a-f}Means followed by different superscripts represent significant differences in the same vitamin ($P < 0.05$).

(1998) evaluated vitamin A degradation and light oxidized flavor development in chocolate milk exposed to fluorescent light for 14 h at 4°C. They found 1.40% of vitamin A losses in chocolate colored and flavored milk in contrast to 24.24% loss in unflavored 2% fat milk. The degradation of vitamin A and light oxidized flavor defect in milk at different fluorescent light intensities were investigated in another study by Whited et al. (2002). They reported that vitamin A content was reduced by 29% in reduced fat milk and by 49% in nonfat milk after 16-h exposure to 2,000 lx fluorescent light. They also reported that the vitamin A content of whole milk, reduced-fat milk, and nonfat milk was reduced by 0, 24, and 32%, respectively, after 16-h exposure to 1,000 lx. Saffert et al. (2008) determined the effect of package light transmittance and fluorescent light intensity on vitamin A, B₂, and D₃ content in UHT milk. They reported a 66, 79, and 88% loss in vitamin A in milk stored in clear-transparent polyethylene terephthalate bottles after 12 wk of storage at 700, 1,700, and 3,000 lx at 23°C, respectively. When the results of previous studies were examined, the vitamin A degradation rate in milk varied, depending on the duration and intensity of light exposure. Relatively lower vitamin A degradation rate in milk exposed to 1-pass UV light (12.6 J/mL doses for cow milk; 11.72 J/mL doses for goat milk) at 254 nm was observed in the present study. It was observed that 1-pass UV light treatment had the same degradation effect as pasteurization did on vitamin A in both cow and goat milk. However, further detailed studies are essential to determine the effects of UV light on fat-soluble vitamins in milk and dairy products.

Manipulation Experiment

Changes in vitamins A, B₂, C, and E in the manipulated cow milk sample (C4) are shown in Table 5. The effect of UV light treatment on vitamin C in milk was found to be significant ($P < 0.05$). No significant differences were observed among vitamin-added raw, pasteurized, and 1-pass, 3-pass, and 5-pass UV light-treated milk samples for vitamin B₂ ($P > 0.05$).

Seven-pass UV treatment significantly decreased the vitamin B₂ content of milk; 27% of vitamin B₂ loss in milk was observed after 7-pass UV treatment. On the other hand, it was determined that UV treatment decreased vitamin C content of milk more than the pasteurization process. Nine percent of vitamin C in milk was lost upon pasteurization; however, 19% of vitamin C was lost by 1-pass UV light treatment. A significant decrease in vitamin C content of milk was also observed by increasing the application dose of UV treatment ($P < 0.05$). Five-pass UV treatment decreased the content of vitamin C in milk by approximately 98%.

The effects of UV and heat treatments were significant on vitamins A and E in milk. A significant decrease in vitamin A content of milk was also observed by increasing the application dose of UV ($P < 0.05$). Although pasteurization decreased the content of vitamin A by 14%, 3-, 5-, and 7-pass UV treatments decreased the content of vitamin A by 15, 16, and 25%, respectively (Table 5). In addition, significant differences were observed between pasteurized and UV-treated milk samples in terms of vitamin E content. The 1-pass UV-treated milk sample had a lower vitamin E content than the vitamin-added raw and pasteurized milk samples ($P < 0.05$). Pasteurization decreased vitamin E by 7%. The 1-, 3-, 5-, and 7-pass UV treatments decreased vitamin E in milk by 18, 23, 32, and 55%, respectively. This result shows that UV treatment was more effective than the pasteurization process at degrading vitamin E. Hence, a significant decrease in vitamin E content of milk was also observed by increasing the application dose of UV treatment ($P < 0.05$; Table 5).

CONCLUSIONS

The amounts of water- and fat-soluble vitamins in milk were affected by pasteurization and UV treatment. The ranges of vitamin losses by heat and UV light treatment of cow and goat milk samples are summarized in Table 6. One-pass UV treatment decreased vitamin A, B₂, C, and E contents of cow milk by 8 to 13%, 3 to 10%, 45 to 74%, and 16 to 33%, respectively. Vitamin

Table 6. Ranges for vitamin losses in milk samples¹ by heat and UV light treatments

Treatment	Loss of vitamin in milk (%)							
	A		B ₂		C		E	
	Cow milk	Goat milk	Cow milk	Goat milk	Cow milk	Goat milk	Cow milk	Goat milk
Pasteurization	0–17	0–8.8	1–10	1.8–4.0	9.0–44	35–47	1.9–14	0–22.4
UV, 1 pass	8.0–13	1.0–9.0	3.0–10	1.0–2.0	45–74	75–91	16–33	1.0–48
UV, 3 passes	12–20	2.85–20	10.9–17.2	11–11.3	78.4–91.2	91.2–96.6	43.2–57.8	3.5–42.7
UV, 5 passes	20–27.5	12.8–28.9	14.1–26.9	11.9–15.1	90.7–100	96.3–100	52.9–67.5	11.5–61.5
UV, 7 passes	30–32	18.6–42.2	20.3–31.2	17.8–22.6	91.4–100	96.3–100	66.3–70.3	24.2–65.6

¹The milk samples are cow milk (C1, C2, and C3) and goat milk (G1, G2, and G3) samples.

A, B₂, C, and E contents in goat milk were decreased by 1-pass UV treatment by 1 to 9%, 1 to 2%, 75 to 91%, and 1 to 48%, respectively. Specifically, vitamins C and E are more sensitive to UV light. Ultraviolet light sensitivities for cow and goat milk samples were in the following order: vitamin C > vitamin E > vitamin A > vitamin B₂. The amount of vitamin losses in milk depends on the number of passes of milk through the UV system, initial amount of vitamin in milk, and the intensity of UV light application. Before recommending this nonthermal technology as an alternative to heat treatment, more studies are needed on the effects of UV light on the nutritional value of milk and dairy products.

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