Effects of UV Irradiation in a Continuous Turbulent Flow UV Reactor on Microbiological and Sensory Characteristics of Cow's Milk

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ABSTRACT

The dairy industry under current pasteurization conditions (15 s at 72°C) and sanitary standards achieves a safe product with excellent quality. In an ever-competitive market there is still a need to improve product quality and extend shelf life of dairy products to increase competitiveness and open up new markets. In an attempt to test the effect of UV irradiation on microbiota of fluid milk, a continuous flow UV system at 254 nm was used to treat 3.5 and 2% fat milk at two UV doses (880 and 1,760 J liter^{-1}). Milk was obtained from three processors, and two lots from each processor were assessed. To assess the impact on the most descriptive native microbiota in pasteurized milk after UV illumination, the product was held at two storage temperatures (4 and 7°C) and tested weekly for 5 weeks for aerobic plate counts (psychrotrophic and mesophilic bacteria), laboratory pasteurization counts, aerobic sporeformers, coliform organisms, and titratable acidity. Microbial counts for all tested microorganisms were lower in UV-treated milk when compared with control throughout storage at 4 and 7°C in both 3.5 and 2% fat milk. Sensory analysis indicated that there is a sensory defect associated with UV treatment at the wavelength used.

Pasteurization of fluid milk at 161°F (71.7°C) for 15 s, or high-temperature short-time, as outlined in the Grade "A" Pasteurized Milk Ordinance *(6),* is the most common milk sanitation practice in the United States. Pasteurization coupled with dairy processing plant sanitary standards, which usually are more rigorous than the Pasteurized Milk Ordinance guidelines for Grade "A" for fluid milk, typically produces a product with a shelf life of 14 to 18 days. During refrigerated storage of or high-temperature short-time pasteurization of milk, growth of thermoduric organisms that survive pasteurization, or of bacteria that enter product after pasteurization, can cause spoilage of product. Thermoduric microbiota associated with milk spoilage that can impact shelf life include gram-negative and gram-positive organisms, including sporeformers *(21, 37, 43). Pseudomonas* spp. have been identified as a significant gram-negative milk spoilage organism *(16),* while the gram-positive *Paenibacillus amylolyticus* is a predominant organism present at the end of shelf life *(18).*

Fluid milk that has a longer shelf life is referred to as extended shelf life milk. Accepted technologies that are approved for making extended shelf life milk include increased heat, bactofugation, and microfiltration. The shelf life of extended shelf life milk varies depending on the treatment and ranges from 30 days for bactofugation to 180 days for ultrahigh-temperature–treated milk *(40).* UV light treatment of food is used for microbial inactivation and is currently approved for surface treatment of foods and for fruit juices *(2)* but is not approved for use in milk. UV is also currently accepted for the treatment of municipal water supplies *(4).* UV light at 254 nm dimerizes DNA *(33),* and when sufficient energy has been applied, the resulting DNA damage is great enough to overwhelm DNA repair mechanisms and cause irreversible damage to bacteria and cell death *(38).*

Use of UV at 254 nm in an opaque substance, such as milk, poses additional challenges in that low transmittance limits UV penetration. Pulsed UV from an excimer laser at 248 nm was used to treat raw milk spiked with *Serratia marcescens* in a collimated system; at an energy level above 12.6 J/cm^2, bacteria were inactivated *(42).* A continuous-flow laboratory-scale pulsed UV system was able to inactivate 7 log of *Staphylococcus aureus (27);* in a continuous-flow system with goat's milk, Matak et al. *(31)* achieved a 5-log reduction for *Listeria monocytogenes* with an energy level of 15 mJ/cm^2 *(31).* A UV reactor design using a continuous turbulent flow system to overcome the turbidity of milk that interferes with UV penetration was able to attain a 3-log reduction of mesophilic aerobes in milk with a dose of 1.5 kJ liter^{-1} *(39).* The turbulent flow system causes efficient mixing of

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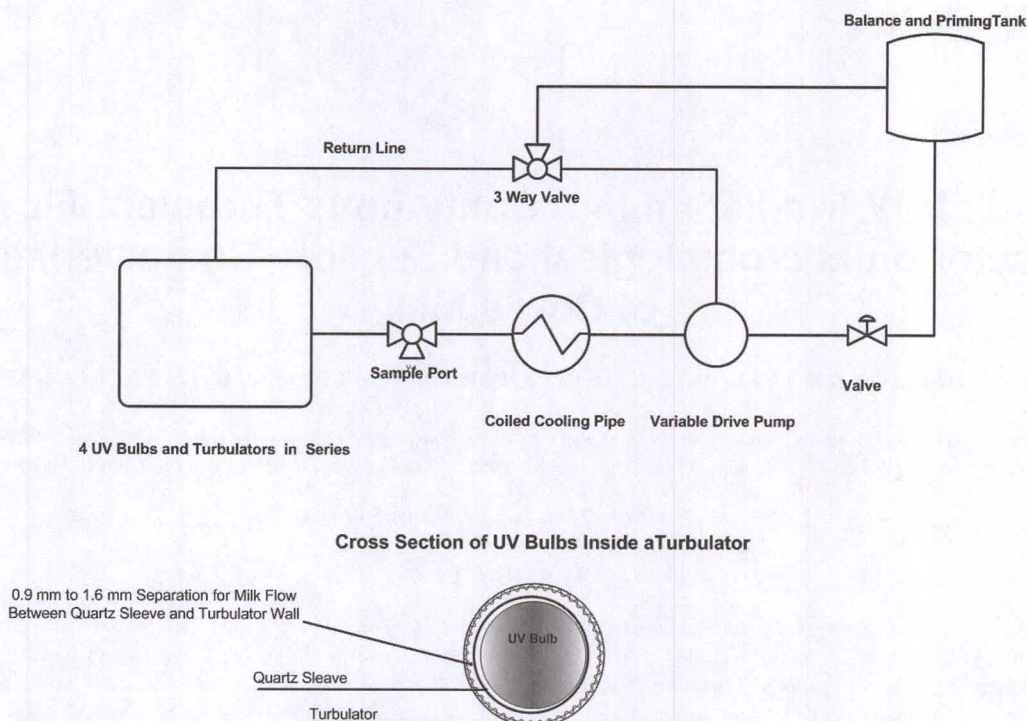
FIGURE 1. *Product flow for UV illumination and sampling.*

the milk, thereby facilitating a more homogenous distribution of UV dose.

Recently, the advantage of UV treatment of milk compared with microfiltration has been addressed. The polymeric or ceramic membranes used in microfiltration need to be cleaned and eventually replaced, with additional costs and quality controls. Fouling is the most important problem associated with the membrane-based milk sterilization process, mostly due to the deposition of bacteria, fats, proteins, and minerals on the membrane surfaces *(30)*. A further issue with microfiltration is that breakage of the filter can go undetected, thereby necessitating a postthermal treatment to inactive bacteria that are not removed by the process *(41)*.

In an attempt to investigate the feasibility of using UV to treat commercial high-temperature short-time pasteurized bovine milk, this study evaluated the effect of a continuous turbulent flow low-power UV reactor at 254 nm (two doses: 880 and 1,760 J liter^{-1}) at commercially practical flow rates (4,000 liters/h) on the shelf life and sensory characteristics of milk.

MATERIALS AND METHODS

Milk. Twelve lots of pasteurized milk (three processors, two fat contents [2 and 3.5%], and two lots each) were used in the experiment. Whole milk (3.5% fat) and reduced fat milk (2% fat), was purchased in 1-gal (3.785-liter) plastic containers from local retailers in Tulare County, California. Each retailer was polled for milk delivery dates, and milk was picked up on a scheduled delivery date to ensure maximum possible freshness from the processors. Three different fluid milk processors were enrolled by purchasing retail milk and confirming the uniqueness of the processor by checking bottle codes on the U.S. Food and Drug Administration (FDA) Web site's Interstate Milk Shipper List

(Sanitation Compliance and Enforcement Ratings of Interstate Milk Shippers) *(9)*.

UV decimal reduction in spiked milk. To determine the log reduction capability by UV treatment of ultrahigh-temperature–treated milk, milk samples (3.5% fat) were spiked with the following bacteria at an initial concentration of 10^7 CFU ml^{-1}: *E. coli* O157:H7 (ATCC 43888), *Salmonella enterica* subsp. *enterica* serovar Senftenberg (ATCC 43845), *Yersinia enterocolitica* (ATCC 9610), *Staphylococcus aureus* (ATCC 29213), *Campylobacter jejuni* (ATCC 33560), *Serratia marcescens* (ATCC 13880), *Aeromonas hydrophila* (ATCC 7966), *Listeria monocytogenes* (ATCC 43256), and the sporeformers *Bacillus cereus* (ATCC 4342), *B. licheniformis* (ATCC 14580), *B. pumilus* (ATCC 72), *B. subtilis* (ATCC 6051), and *Paenibacillus lautus* (ATCC 43898). Using the same UV reactor design being tested in this work, six dose-response trials for each target microorganism, from 0 through 8,800 J liter^{-1}, were conducted by standard methods *(26, 41)*. Target UV doses were achieved by recirculating milk and were kept below 6°C during UV treatment with the use of a heat exchanger: a coiled section of 1½-in. (38-mm) stainless steel tubing was placed in an ice and water bath (Fig. 1).

UV treatment and milk storage. UV treatment of fluid milk was performed in a research-scale low-power UV unit designated SP-4 (SurePure, Milnerton, South Africa). The unit contained four 22-W 254-nm UV bulbs in series. An optically pure quartz sleeve separated milk from the UV bulb. Milk was processed in a 0.9- to 1.6-mm channel over the quartz sleeve at 4,000 liters/h. Target UV doses were achieved by recirculating milk and were kept below 6°C during UV treatment with the use of a heat exchanger: a coiled section of 1½-in. stainless steel tubing was placed in an ice-water bath (Fig. 1). An electronic temperature sensor placed in line confirmed product temperature during the entire run. According to results of UV decimal reduction testing, milk received three treatments in the SP-4 unit: 0, 880, and 1,760 J liter^{-1}. After

treatment, milk was drained from the SP-4 unit and was placed into 0.5-liter sterile polyethylene bottles filled to the top. Milk sample bottles were then stored at 4°C (proper storage) and 7°C (temperature abuse) for subsequent testing at 0, 7, 14, 21, 28, and 35 days after UV treatment. The SP-4 unit was cleaned after every use with a clean-in-place process.

Cleaning of pilot-scale UV unit. All milk in the system was flushed with cold water and then treated with caustic wash with a conductivity of 6 to 13 mS at 70°C for 30 min, followed by a 70°C rinse, then by an acid wash at 23°C at 2 to 4 mS for 10 min, and finally by sanitization with hypochlorous acid at 3,300 ppm for 30 min at 23°C. In particular, the UV unit to treat milk was cleaned after every run by first draining the system of all milk, then flushing with 37°C water until it ran clear. Flow rate was changed to 5,000 liters/h for the caustic wash at 70°C with 1% alkaline solution for 30 min, followed by rinsing. Acid wash followed at 70°C for 10 min with 4 oz (0.118 liter) of three-way acid per 112 liters of water (Anfo Manufacturing, Hayward, CA), followed by rinsing. A final sanitizing step at 23°C for 30 min with 3 liters of 12.5% bleach per 112 liters was applied, followed by draining. The unit remained dry between uses, and a sanitizing run followed by a rinse was performed before each UV treatment. Proper levels of all reagents were confirmed with an inline conductivity meter (Cole-Parmer, Vernon Hills, IL).

UV dosage calculations. UV output was confirmed by measuring power with a UV radiometer (UVP, Upland, CA) on the milk side of the quartz sleeve, using the formula: power = UV dose mJ/cm^2. The length of the quartz sleeve was 0.86 m with a surface area of 661 cm^2. For working with milk, UV power was expressed in joules per liter. Conversion of power to joules per liter was described by Keyser et al. (25). UV output was used instead of direct measurements of dose delivered to the milk, for the sake of consistency and for data comparison with current researches carried out elsewhere (13, 15, 17, 29).

Microbiological analysis. Samples from each milk fat percentage (3.5 and 2%), storage temperature (4 and 7°C), and analysis time point (0, 7, 14, 21, 28, and 35 days) were subjected to microbiological analysis. Milk samples were diluted in Butterfield's phosphate buffer (Hardy Diagnostics, Santa Maria, CA) (11) before pour plating or spread plating using sterile plastic sticks (Life Science Products, Frederick, CO). Aerobic plate counts were conducted on tryptic soy agar (TSA; Hardy Diagnostics) for mesophilic bacteria at 30°C for 24 to 48 h (22) and on plate count milk agar (Veterinary Medicine Media Services, University of California, Davis) for psychrotrophic bacteria at 6.5°C for 7 days (24). Laboratory pasteurization counts (LPC) were conducted by heating 5 ml of milk in a glass tube for 30 min at 63°C, pour plating in standard methods agar (Hardy Diagnostics), and incubating at 33°C for 48 h. For aerobic sporeformers, 5 ml of milk was placed in a glass tube (16 by 125 mm) and treated at 80°C for 10 min in a shaking water bath and then immediately chilled on ice, before spread plating on TSA and culturing at 35°C for 18 to 22 h. Coliform bacterial enumeration was based on lactose-positive colony growth on MacConkey agar (Hardy Diagnostics) cultured at 35°C for 24 h.

Sensitivity for spread plate was 10 CFU ml^{-1} and for pour plate was 1 CFU ml^{-1}, and the 95% confidence limit, as given by the classic formula $2s = 2\sqrt{x}$ (1), ranged between ±37% and ±12% (i.e., plates with CFU ranging from 30 to 300). Therefore, no plates with fewer than 30 CFU were used for data analysis, and when this was applied to the lowest dilution, the results were recorded as <30 for pour plate and <300 for spread plate.

Chemical analysis. Milk components were checked for consistency of protein and fat content. Milk component analysis was performed with a MilkoScan 203 (DK-3400, Foss Electric, Hillerød, Denmark) to determine the concentrations of fat and proteins. Determination of titratable acidity was performed according to standard methods (44), and results are reported as percent of lactic acid.

Sensory analysis. The University of California at Davis, Institutional Review Board approved the sensory analysis of UV light–illuminated milk under exempt status. Milk samples were assessed for differences in odor and taste. Comparisons included (for both fat percentages of 2 and 3.5%) retail milk, retail milk exposed to UV light at 0 J liter^{-1}, retail milk exposed to UV light at 880 J liter^{-1}, and retail milk exposed to UV light at 1,760 J liter^{-1}. Each comparison was evaluated using a triangle test (23). Samples (~25 ml) were poured the day of testing into semi-opaque plastic cups with plastic lids, assigned a random three-digit code, and stored and maintained at 4°C until sensory testing. Samples were allowed to stand at least 20 min before testing. The samples for sensory analysis were held for the length of the code date established by the fluid milk processor, and sensory tests occurred at 1, 8, and 15 days. All combinations of the two samples were presented within each sensory session an equal number of times. Two sets of three samples were presented to each panelist, representing a balanced order of presentation. Panelists were instructed to identify the sample that smelled and tasted different in each group of three. There was additional space for comments, with instructions to describe any odor or taste associated with the unique sample. Thirty-six volunteers (≥18 years old) were recruited from the Veterinary Medicine Teaching and Research Center in Tulare, CA, to serve on each panel session. Each panelist contributed one observation per testing session, for a total of 36 observations per triangle test. Testing was conducted in individual booths in the Veterinary Medicine Teaching and Research Center. Panelists were required to complete a consent form, approved by the University of California at Davis, Institutional Review Board, prior to testing. Each panelist was verbally reminded that all samples were retail milk samples.

Statistical analysis. For each trial, the log of the arithmetic means for all microbiological analysis was calculated, following which all log values were analyzed with InStat, ver. 3.0b for Mac OS X (GraphPad Software, San Diego, CA) for analysis of variance (ANOVA) followed by the Tukey-Kramer multiple comparisons test.

The data for each triangle test were analyzed by the number of correct responses versus the total number of responses. Parameters were defined at $n = 36$, $\alpha = 0.01$, $\beta = 0.05$, and $\rho d = 50\%$; the critical number of correct responses for significance was 20 of 36 (24). One test was administered per repetition, and tests were not replicated. One-way ANOVA and Tukey-Kramer multiple comparisons test were used to analyze the data. A P value of <0.05 was considered to be significant. Data were analyzed using GraphPad InStat ver. 3.0b for Mac OS X.

Interpretive criteria. For a more perceptive interpretation of data, a series of cut-off criteria have been introduced, using as a reference the Grade "A" Pasteurized Milk Ordinance and the European Union regulations (3, 5, 6). These criteria have been set at levels below the food safety and spoilage limits and are used as process hygiene criteria. Indeed, spoiled milk actually has higher CFUs, and these cut-off levels were chosen as they represent bacteria in log-phase growth and anticipate sensory and physical defects (34). The following interpretive criteria have, therefore, been set up: psychrotrophic and mesophilic microbiota, log CFU

TABLE 1. *Decimal reduction for pathogens and sporeformers after UV treatment[a]*

	ATCC	D-value (J/liter)	SE
Escherichia coli O157:H7	ATCC 43888	334	27.98
Salmonella enterica serovar Senftenberg	ATCC 43845	365	22.91
Yersinia enterocolitica	ATCC 9610	311	30.01
Staphylococcus aureus	ATCC 29213	335	17.68
Campylobacter jejuni	ATCC 33560	354	24.41
Serratia marcescens	ATCC 13880	352	34.92
Aeromonas hydrophila	ATCC 7966	293	24.06
Listeria monocytogenes	ATCC 43256	350	31.69
Bacillus cereus	ATCC 4342	1,250	65.66
Bacillus licheniformis	ATCC 14580	294	28.49
Bacillus pumilus	ATCC 72	1,250	82.49
Bacillus subtilis	ATCC 6051	770	54.27
Paenibacillus lautus	ATCC 43898	1,430	137.49

[a] UV treatment at 254 nm (from 0 to 8,800); mean D-value at 6°C of six runs.

$ml^{-1} > 4$; LPC, log CFU $ml^{-1} > 2$; aerobic sporeformers, 5 CFU ml^{-1}; coliform organisms, first positive culture; titratable acidity, >0.2% lactic acid.

RESULTS

UV decimal reduction. Results are shown in Table 1. Targeted UV doses to be used in the experiment were based on the UV inactivation dose-response work with the major pathogens associated with foodborne milk outbreaks and spore-forming bacteria associated with spoilage of high-temperature short-time pasteurized milk. Regression analysis of UV doses from 0 through 8,800 J liter^{-1} in 3.5% fat milk indicated a decimal reduction that ranged from 293 to 365 J liter^{-1} for the eight different foodborne pathogen species and from 294 to 1,430 J liter^{-1} for viable spores. These trials, carried out in order to assess the best UV doses, indicated

TABLE 2. *Development of microbiota in 3.5% fat milk during storage at 4°C[a]*

Treatment	Day 0 Mean	SE	Day 7 Mean	SE	Day 14 Mean	SE	Day 21 Mean	SE	Day 28 Mean	SE	Day 35 Mean	SE
Psychrotrophic microbiota												
Control	0.10	0.07	3.87 A	0.93	6.98 A	0.69	7.58 A	0.29	6.57 A	0.93	7.05 A	0.29
0 J/liter	0.03	0.03	4.57 A	0.49	7.53 A	0.16	7.56 A	0.27	7.66 A	0.23	7.42 A	0.18
880 J/liter	LDL	LDL	LDL B	LDL	0.46 B	0.22	2.06 B	0.63	1.72 B	0.68	3.10 B	0.60
1,760 J/liter	LDL	LDL	LDL B	LDL	0.08 B	0.08	1.06 B	0.38	0.99 B	0.70	1.76 B	0.58
Mesophilic microbiota												
Control	0.67	0.17	4.62 A	0.83	7.23 A	0.28	7.63 A	0.24	7.52 A	0.07	7.40 A	0.26
0 J/liter	0.88	0.22	4.24 A	0.65	7.39 A	0.16	7.60 A	0.22	7.74 A	0.22	7.05 A	0.23
880 J/liter	0.52	0.19	0.33 B	0.20	0.94 B	0.25	2.25 B	0.64	0.80 B	0.42	3.69 B	0.44
1,760 J/liter	0.41	0.15	0.26 B	0.18	0.62 B	0.41	1.82 B	0.57	1.77 B	0.52	3.54 B	0.61
LPC												
Control	1.76	0.26	1.45	0.30	1.63	0.30	1.40	0.27	1.75	0.18	2.01	0.54
0 J/liter	1.64	0.34	1.64	0.25	1.63	0.26	1.68	0.17	1.82	0.18	1.67	0.24
880 J/liter	1.09	0.29	1.04	0.23	0.98	0.27	0.93	0.25	0.86	0.25	0.99	0.12
1,760 J/liter	0.88	0.28	0.68	0.28	0.62	0.19	0.84	0.13	0.86	0.26	0.82	0.18
Aerobic sporeformers												
Control	0.62	0.15	0.98	0.38	1.42	1.22	1.45	1.23	1.52	1.24	1.45	1.11
0 J/liter	0.58	0.16	1.29	0.44	1.51	1.16	1.57	1.17	1.52	1.16	1.76	1.24
880 J/liter	0.45	0.19	0.32	0.18	0.11	0.07	0.69	0.57	1.01	0.70	0.50	0.31
1,760 J/liter	0.18	0.15	0.06	0.06	LDL	LDL	LDL	LDL	0.62	0.25	0.17	0.11
Coliform organisms												
Control	LDL	LDL	0.22	0.22	1.84	0.92	1.28	0.71	2.04	2.04	0.69	0.38
0 J/liter	LDL	LDL	0.40	0.20	1.71	0.89	1.28	0.61	1.49	0.75	0.83	0.64
880 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL
1,760 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL

[a] Average values of six replicates, sampled in triplicate, and standard error (log CFU per milliliter). LDL, lower detection limit. Different letters within the same column indicate significantly (P < 0.05) different means.

TABLE 3. *Development of microbiota in 3.5% fat milk during storage at 7°C[a]*

Treatment	Day 0		Day 7		Day 14		Day 21		Day 28		Day 35	
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Psychrotrophic microbiota												
Control	0.10	0.07	5.78 A	0.62	7.36 A	0.24	7.64 A	0.25	6.88 A	0.65	7.15 A	0.22
0 J/liter	0.03	0.03	5.80 A	0.90	7.57 A	0.61	7.60 A	0.28	7.54 A	0.18	7.39 A	0.15
880 J/liter	LDL	LDL	0.39 B	0.19	1.51 B	0.64	3.48 B	0.94	3.40 B	1.34	4.31 AB	0.97
1,760 J/liter	LDL	LDL	0.35 B	0.21	1.33 B	0.59	2.76 B	0.81	2.84 B	0.84	2.64 B	0.95
Mesophilic microbiota												
Control	0.67	0.17	5.97 A	0.52	7.29 A	0.19	7.66 A	0.22	7.29	0.38	7.33	0.28
0 J/liter	0.88	0.22	6.10 A	0.66	7.63 A	0.36	7.80 A	0.21	7.44	0.17	7.37	0.17
880 J/liter	0.52	0.19	0.34 B	0.13	3.11 B	0.91	5.24 B	0.45	5.62	0.59	6.64	0.33
1,760 J/liter	0.41	0.15	0.39 B	0.24	2.87 B	0.87	3.30 B	0.79	5.14	0.44	5.10	0.88
LPC												
Control	1.76	0.26	1.77	0.18	1.89	0.20	1.97	0.17	2.95 A	0.85	1.85	LDL
0 J/liter	1.64	0.34	1.67	0.24	2.16	0.37	1.96	0.33	1.80 AB	0.37	LDL	LDL
880 J/liter	1.09	0.29	1.12	0.11	1.02	0.19	1.04	0.14	1.19 AB	0.26	1.36	0.42
1,760 J/liter	0.88	0.28	0.93	0.14	0.77	0.19	0.75	0.20	1.08 B	0.30	1.12	0.42
Aerobic sporeformers												
Control	0.62	0.15	0.81	0.53	1.46	1.13	1.37	1.30	1.57	1.19	1.61	1.11
0 J/liter	0.58	0.16	0.72	0.35	1.55	1.23	1.42	1.15	1.53	1.23	1.51	1.19
880 J/liter	0.45	0.19	0.26	0.17	0.25	0.11	0.73	0.67	0.22	0.11	0.56	0.35
1,760 J/liter	0.18	0.15	0.34	0.13	0.45	0.33	0.84	0.72	0.56	0.38	0.45	0.45
Coliform organisms												
Control	LDL	LDL	1.35	0.69	3.24	1.69	2.26	2.26	1.85	0.95	1.90	0.83
0 J/liter	LDL	LDL	2.12	1.06	2.98	0.45	2.29	1.22	1.98	1.98	2.09	2.09
880 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL
1,760 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL

[a] Average values of six replicates, sampled in triplicate, and standard error (log CFU per milliliter). LDL, lower detection limit. Different letters within the same column indicate significantly ($P < 0.05$) different means.

that doses below 880 J liter^{-1} have little effect (less than three decimal reductions), if any, on pathogens and spoilage microorganisms, while for doses higher than 1,760 J liter^{-1} there is a plateau and the decimal reduction does not increase.

Microbiological analysis. Results are shown in Tables 2, 3, 4, and 5. For all groups, on day 0, psychrotrophic and mesophilic microbiota and LPC were below 10^2 CFU ml^{-1}, aerobic sporeformers were always below 10^1 CFU ml^{-1}, and coliform organisms were always below the lower detection limit in all samples.

During storage (either at 4 and 7°C for both 3.5 and 2% milk), psychrotrophic and mesophilic microbiota reached values above 10^5 CFU ml^{-1} after 14 days for control and 0 J liter^{-1} groups, while in treated groups (880 and 1,760 J liter^{-1}) counts were always below 10^5 CFU ml^{-1}. LPC reached values up to 10^2 CFU ml^{-1}, for control and 0 J liter^{-1} groups, and were always below 10^5 CFU ml^{-1} in treated groups. Aerobic sporeformers were always below 10 CFU ml^{-1} in treated groups and below 10^2 CFU ml^{-1} in control groups. Coliform organisms reached levels up to 10^3 CFU ml^{-1} for control groups and were never detected from treated groups.

Chemical analysis. Results are shown in Table 6. Concentrations of fat for 3.5% milk ranged from 3.38 (plant

2) to 3.44 (plant 1), proteins were between 3.27% (plant 1) and 3.39% (plant 3), lactose was between 5.07% (plant 3) and 5.13% (plant 1), and solids between 8.85% (plant 1) and 8.91% (plant 3). Reduced fat milk (2%) analysis revealed that fat ranged from 1.96% (plant 3) to 1.99% (plant 1), proteins from 3.77% (plant 1) to 3.87% (plant 3), lactose from 5.71% (plant 3) to 5.84% (plant 2), and solids from 10.04% (plant 3) to 10.20% (plant 2).

Titratable acidity (Table 7) was below 0.2% lactic acid for all groups on day 0 and reached levels above 0.3% for control and 0 J liter^{-1} groups. Treated groups (880 and 1,760 J liter^{-1}) remained below 0.3%.

Sensory analysis. Panelists detected differences ($\alpha = 0.01$) between control (and 0 J liter^{-1} treatment) and 1,760 J liter^{-1} treatment throughout storage (Tables 8 and 9). Differences were also detected between 0 and 880 J liter^{-1} and 880 versus 1,760 J liter^{-1}. No differences were detected in the control versus 0 J liter^{-1} for 3.5% fat milk and control versus 0 J liter^{-1} and control versus 880 J liter^{-1} for 2% fat milk.

DISCUSSION

According to the decimal reduction trials conducted at the Veterinary Medicine Teaching and Research Center,

TABLE 4. *Development of microbiota in 2% fat milk during storage at 4°C[a]*

Treatment	Day 0 Mean	SE	Day 7 Mean	SE	Day 14 Mean	SE	Day 21 Mean	SE	Day 28 Mean	SE	Day 35 Mean	SE
Psychrotrophic microbiota												
Control	0.05	0.05	3.99	0.94	6.29 A	1.27	6.47 A	1.30	6.33 A	1.30	5.82 AB	1.28
0 J/liter	0.53	0.50	3.86	0.98	6.46 A	1.30	6.40 A	1.32	6.42 A	1.32	6.59 A	1.36
880 J/liter	LDL	LDL	LDL	LDL	0.27 B	0.27	1.68 AB	1.06	1.09 B	0.78	2.19 AB	0.86
1,760 J/liter	0.03	0.03	0.10	0.10	0.39 B	0.28	0.84 B	0.53	1.19 B	0.67	1.33 B	0.48
Mesophilic microbiota												
Control	0.94	0.09	4.39 A	0.74	6.62 A	0.67	7.20 A	0.56	7.20 A	0.44	7.51 A	0.36
0 J/liter	0.99	0.13	4.28 A	0.78	7.14 A	0.31	7.09 A	0.40	7.32 A	0.34	7.70 A	0.31
880 J/liter	0.42	0.18	0.34 B	0.11	0.22 B	0.14	1.30 B	1.10	2.65 B	0.88	2.99 B	0.81
1,760 J/liter	0.19	0.11	LDL B	LDL	0.20 B	0.13	0.91 B	0.43	3.84 B	0.50	2.49 B	0.63
LPC												
Control	1.75	0.25	2.01 A	0.29	1.76 AB	0.30	1.88	0.32	1.77 AB	0.23	1.93 A	0.28
0 J/liter	1.81	0.25	1.88 AB	0.32	1.90 A	0.28	2.08	0.21	2.01 A	0.40	1.32 AB	0.14
880 J/liter	1.13	0.42	1.04 AB	0.35	0.64 AB	0.36	0.90	0.30	0.47 B	0.21	0.68 AB	0.16
1,760 J/liter	0.53	0.25	0.55 B	0.16	0.44 B	0.16	0.92	0.18	0.44 B	0.16	0.46 B	0.14
Aerobic sporeformers												
Control	0.64	0.16	1.60	0.93	1.44	1.21	1.52	1.20	1.85	1.07	1.68	1.08
0 J/liter	0.70	0.17	1.44	0.84	1.70	1.19	1.76	1.18	2.08	1.19	1.68	1.26
880 J/liter	0.26	0.07	0.15	0.15	0.06	0.06	0.41	0.17	0.22	0.16	0.44	0.31
1,760 J/liter	0.13	0.07	0.17	0.07	0.08	0.08	0.14	0.09	0.11	0.07	0.27	0.15
Coliform organisms												
Control	LDL	LDL	0.75	0.75	0.47	0.47	1.14	0.66	0.34	0.34	0.33	0.19
0 J/liter	LDL	LDL	0.57	0.57	0.68	0.68	0.94	0.56	0.40	0.40	1.23	0.82
880 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL
1,760 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL

[a] Average values of six replicates, sampled in triplicate, and standard error (log CFU per milliliter). LDL, lower detection limit. Different letters within the same column indicate significantly ($P < 0.05$) different means.

School of Veterinary Medicine, University of California at Davis, which confirmed what is available in the literature *(17, 28, 31, 39)*, a control (0 J liter^{-1}) and two UV doses (880 and 1,760 J liter^{-1}) have been used throughout the experiment. The shape of the curve for microbial inactivation by UV light is known to be sigmoidal *(12)*. The initial plateau is due to an injury phase of the microorganism in response to UV exposure. After the initial plateau, the maximum amount of injury has been surpassed; thus, minimal additional UV exposure would be lethal for microorganisms, and survivor numbers rapidly decline. The end of the curve has a tailing phase due to UV resistance of the microorganisms and to experimental components, such as suspended solids that may block the UV irradiation. The UV decimal reduction in spiked milk trials demonstrated that for doses higher than 1,760 J liter^{-1} there is a plateau and the decimal reduction does not increase, while for doses lower than 880 J liter^{-1} the effect is negligible (less than three decimal reductions for pathogens and less than one decimal reduction for sporeformers).

UV irradiation of 3.5 and 2% fat milk before storage at 4 and 7°C greatly reduced the growth of psychrotrophic and mesophilic microbiota (Tables 2, 3, 4, and 5). From day 7 throughout storage, almost all comparisons produced significantly different means ($P < 0.05$) when control and 0 J liter^{-1} were compared with treatments (880 and 1,760 J liter^{-1}). Only for milk (both 3.5 and 2%) stored at 7°C did some analyses on day 28 and day 35 not result in significantly different means ($P < 0.05$), although bacterial counts were always lower (at least 2 log CFU ml^{-1}). Given the adoption of the ISO 4833:2003 procedure *(22)* with the incubation at 30°C for mesophilic microbiota, it is likely that the values recorded might also include psychrotrophic bacteria. On the other hand, the ISO 4833:2003 procedure is the method of choice for milk to determine the effect of two treatments, as stated by the European Union Community reference laboratory for milk and milk products (ANSES, Laboratoire de sécurité des aliments de Maisons-Alfort, Maisons-Alfort, France) *(8)*. Indeed, bacterial counts for mesophilic and psychrotrophic microbiota appear similar. This is due partly to the ability of certain psychrotrophic bacteria, such as certain strains of *Pseudomonas* spp., to grow at a wide range of temperatures: their average growth range is 0 to +40°C. In fact, it is mainly by their versatility that psychrotrophs differ from other classes, as illustrated by their predilection for habitats that undergo large thermal fluctuations *(19)*. In this study, after incubation of milk at 4 and 7°C, bacterial counts for mesophilic bacteria were performed at 30°C, according to ISO procedure *(22)* to best

TABLE 5. *Development of microbiota in 2% fat milk during storage at 7°C*[a]

Treatment	Day 0 Mean	Day 0 SE	Day 7 Mean	Day 7 SE	Day 14 Mean	Day 14 SE	Day 21 Mean	Day 21 SE	Day 28 Mean	Day 28 SE	Day 35 Mean	Day 35 SE
Psychrotrophic microbiota												
Control	0.05	0.05	4.57 AB	1.46	5.60 AB	1.38	5.49	1.45	5.96	1.33	5.26	1.41
0 J/liter	0.53	0.50	6.17 A	0.85	6.61 A	1.05	7.12	1.06	7.14	0.91	6.66	1.34
880 J/liter	LDL	LDL	0.28 B	0.28	1.78 AB	0.74	3.60	1.08	4.00	1.13	2.36	1.50
1,760 J/liter	0.03	0.03	0.21 B	0.16	0.99 B	0.32	2.81	0.88	2.71	0.96	2.07	0.58
Mesophilic microbiota												
Control	0.94	0.09	4.76 A	1.21	6.80 A	0.88	7.28 AB	0.65	6.55	0.83	7.27	0.39
0 J/liter	0.99	0.13	6.43 A	0.58	7.24 A	0.46	7.99 A	0.26	7.95	0.22	7.48	0.36
880 J/liter	0.42	0.18	0.47 B	0.32	2.41 B	0.53	4.94 B	0.34	6.20	0.51	6.28	0.67
1,760 J/liter	0.19	0.11	0.07 B	0.07	2.13 B	0.52	4.38 B	0.75	5.45	0.74	5.77	0.89
LPC												
Control	1.75	0.25	1.80	0.45	2.01	0.30	2.06	0.42	2.08	0.85	LDL	LDL
0 J/liter	1.81	0.25	1.56	0.43	2.01	0.27	1.70	0.32	1.97	0.67	LDL	LDL
880 J/liter	1.13	0.42	1.29	0.19	0.96	0.33	1.06	0.21	0.91	0.30	1.49	0.26
1,760 J/liter	0.53	0.25	1.01	0.34	0.84	0.17	0.88	0.24	0.91	0.20	1.49	0.45
Aerobic sporeformers												
Control	0.64	0.16	1.60	0.97	1.63	1.14	1.57	1.13	1.65	1.19	1.55	1.17
0 J/liter	0.70	0.17	1.43	1.05	1.62	1.12	1.61	1.25	1.80	1.28	1.76	1.26
880 J/liter	0.26	0.07	0.07	0.07	0.13	0.08	1.14	0.83	1.08	0.71	0.13	0.08
1,760 J/liter	0.13	0.07	0.18	0.18	0.44	0.25	0.87	0.75	1.06	0.99	0.38	0.38
Coliform organisms												
Control	LDL	LDL	0.68	0.68	0.75	0.75	0.21	0.21	LDL	LDL	0.31	0.31
0 J/liter	LDL	LDL	0.75	0.75	0.80	0.80	0.59	0.59	1.70	1.19	LDL	LDL
880 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	LDL	LDL	0.08	0.08	LDL	LDL
1,760 J/liter	LDL	LDL	LDL	LDL	LDL	LDL	0.10	0.10	LDL	LDL	LDL	LDL

[a] Average values of six replicates, sampled in triplicate, and standard error (log CFU per milliliter). LDL, lower detection limit. Different letters within the same column indicate significantly ($P < 0.05$) different means.

evaluate two different treatments (control versus UV-treated milk). For the abovementioned reasons, it is possible that raw data for mesophilic counts can overestimate true bacterial load due to growth of psychrotrophic microbiota also.

LPC, although always lower in treated groups (880 and 1,760 J liter^{-1}), produced significantly different means ($P < 0.05$) only for 3.5% fat milk stored at 7°C on day 28 and for 2% fat milk stored at 4°C on days 7, 14, 28, and 35.

Aerobic sporeformer and coliform organism counts did not produce significantly different means for the two fat percentages (3.5 and 2%) throughout both storage conditions (4 and 7°C), although counts were always remarkably lower in control groups. Coliform organisms were always below the lower detectable limit both in control groups and in UV-treated groups (880 and 1,760 J liter^{-1}). Thermoduric spores from psychrotrophic *Bacillus* spp. and *Paenibacillus* spp. that survive pasteurization have been implicated as major contributors to spoilage of fluid milk (20, 37). The spores present in the pasteurized milk did not cause an increase of sporeformer counts in control milk or in UV-treated groups, indicating that spore-producing strains that could cause spoilage were not present in the samples tested.

Means for titratable acidity were always significantly different between control and 0 J liter^{-1} and treated groups

(880 and 1,760 J liter^{-1}) from day 28, with the exception of 3.5% fat milk stored at 7°C, which produced significantly different means from day 21.

Using the interpretive criteria of log CFU ml^{-1} > 4 for psychrotrophic and mesophilic microbiota, UV-treated (880 and 1,760 J liter^{-1}) milk stored at 4°C was always below the interpretive criteria at the end of the observation period (day 35), as compared with the control group, which exceeded the interpretive criteria just at the first observation period (day 7) (Tables 10 and 11). For milk stored at 7°C, control and 0 J liter^{-1} groups exceeded the interpretive criteria on day 7 or day 14, while treated groups (880 and 1,760 J liter^{-1}) exceeded the interpretive criteria on day 21 and day 28 for mesophilic microbiota and on day 35 for psychrotrophic microbiota. Control milk, therefore, reached cut-off values in less than 1 week, with aerobic microbiota counts of 10^4 CFU ml^{-1} when held at 4°C and 10^6 CFU ml^{-1} at 7°C. This level of microbial growth in control milk is consistent with data from Ranieri et al. (36), who reported that the aerobic plate count of 2% fat fluid milk pasteurized at 72.9°C increased from 10^2 CFU ml^{-1} on day 1 postpasteurization to 10^6 CFU ml^{-1} on day 21 when stored at 6°C, and Petrus et al. (35), who reported a 10^2 CFU ml^{-1} increase in 4 days at 4°C. Spoiled milk actually has higher CFUs, and this cut-off level was chosen as it represents

TABLE 6. *Chemical parameters of milk[a]*

| | Plant 1 | | | | Plant 2 | | | | Plant 3 | | | |
| | 3.5% fat | | 2% fat | | 3.5% fat | | 2% fat | | 3.5% fat | | 2% fat | |
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
Fat	3.44	0.01	1.99	0.04	3.38	0.02	1.89	0.01	3.42	0.01	1.96	0.03
Proteins	3.27	0.03	3.77	0.04	3.35	0.02	3.84	0.01	3.39	0.03	3.87	0.01
Lactose	5.13	0.01	5.83	0.03	5.12	0.02	5.84	0.07	5.07	0.03	5.71	0.03
Solids nonfat	8.85	0.04	10.09	0.01	8.90	0.01	10.20	0.09	8.91	0.03	10.04	0.02

[a] Average values of two replicates per plant, sampled in triplicate, and standard error (wt%).

bacteria in log-phase growth and anticipates sensory and physical defects *(34)*. Titratable acidity generally correlated with this observation, but sometimes lagged by a week, with criteria of 0.20% lactic acid. Some samples did not reach the cut-off criteria, confirming that psychrotrophic and mesophilic microbiota counts are a better predictor of looming microbial spoilage than titratable acidity *(14)*. In fact, the interpretive criteria have been set using as a reference the Grade "A" Pasteurized Milk Ordinance and the European Union regulations *(3, 5, 6)* at levels below the food safety and spoilage limits to be used as process hygiene criteria.

Coliform organisms and aerobic sporeformers never reached the cut-off values in treated groups (880 and 1,760 J liter^{-1}), except for sporeformers in the 880 J liter^{-1} group (cut-off reached on day 21, both at 4 and 7°C storage) and in the 1,760 J liter^{-1} group (cut-off reached on day 21 at 7°C storage). LPC never reached the cut-off in treated

groups, when compared with control and 0 J liter^{-1} groups (cut-off reached on day 7 to 28).

Sensory analysis using the triangle test to confirm that a difference existed clearly indicates that panelists differentiated treated milk from control milk at both doses (880 and 1,760 J liter^{-1}); however, the method is not quantitative and does not assess the degree of difference or type of difference. Comments volunteered by panelists clearly indicate that there is a sensory defect associated with UV treatment. The defect was described with comments of "burnt," "off," "strong," and "stale," although the most common comment panelists made was that they could not taste a difference. A sensory defect attributed to UV light exposure was associated with lipid oxidation (as evidenced by an increase in thiobarbituric reactive substances) but not rancidity (as measured by acid degree value), which was similar to controls *(32)*. The trials carried out in order to

TABLE 7. *Development of titratable acidity in milk during storage[a]*

| Treatment | Day 0 | | Day 7 | | Day 14 | | Day 21 | | Day 28 | | Day 35 | |
	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
3.5% fat milk stored at 4°C												
Control	0.15	0.00	0.16	0.01	0.18	0.02	0.22	0.04	0.32 A	0.05	0.33 A	0.05
0 J/liter	0.16	0.00	0.16	0.01	0.18	0.02	0.24	0.06	0.29 AB	0.05	0.33 A	0.08
880 J/liter	0.15	0.00	0.15	0.01	0.16	0.01	0.17	0.01	0.16 B	0.01	0.15 B	0.01
1,760 J/liter	0.16	0.00	0.16	0.01	0.17	0.01	0.16	0.01	0.16 B	0.00	0.18 B	0.01
3.5% fat milk stored at 7°C												
Control	0.15	0.00	0.16	0.01	0.24	0.02	0.32 AB	0.05	0.39 A	0.05	0.46 A	0.05
0 J/liter	0.16	0.00	0.16	0.01	0.24	0.04	0.34 A	0.05	0.43 A	0.06	0.47 A	0.05
880 J/liter	0.15	0.00	0.16	0.01	0.15	0.00	0.18 AB	0.01	0.31 AB	0.05	0.26 B	0.03
1,760 J/liter	0.16	0.00	0.15	0.01	0.15	0.00	0.16 B	0.01	0.19 B	0.03	0.26 B	0.03
2% fat milk stored at 4°C												
Control	0.17	0.00	0.17	0.01	0.18	0.00	0.23	0.02	0.28 AB	0.04	0.30 AB	0.03
0 J/liter	0.16	0.00	0.17	0.01	0.19	0.01	0.23	0.03	0.31 A	0.06	0.34 AC	0.03
880 J/liter	0.18	0.00	0.16	0.01	0.16	0.00	0.18	0.01	0.19 B	0.01	0.20 B	0.02
1,760 J/liter	0.18	0.01	0.17	0.01	0.17	0.00	0.17	0.01	0.18 B	0.00	0.20 C	0.02
2% fat milk stored at 7°C												
Control	0.17	0.00	0.17	0.01	0.23	0.02	0.33	0.05	0.43 A	0.05	0.48 A	0.05
0 J/liter	0.16	0.00	0.17	0.01	0.25	0.03	0.34	0.04	0.45 A	0.03	0.54 A	0.07
880 J/liter	0.18	0.00	0.16	0.01	0.17	0.00	0.20	0.00	0.26 B	0.02	0.31 B	0.02
1,760 J/liter	0.18	0.01	0.17	0.01	0.17	0.00	0.20	0.00	0.23 B	0.02	0.32 B	0.03

[a] Average values of six replicates, sampled in triplicate, and standard error (% lactic acid). Different letters within the same column indicate significantly ($P < 0.05$) different means.

TABLE 8. *Sensory analysis, triangle test for 3.5% fat milk[a]*

	Taste			Aroma		
	Day 1	Day 8	Day 15	Day 1	Day 8	Day 15
Control vs 0 J/L	NS	NS	NS	NS	NS	NS
Control vs 880 J/L	NS	S	NS	NS	NS	NS
Control vs 1,760 J/L	S	S	S	NS	NS	NS
0 J/L vs 880 J/L	NS	NS	S	NS	NS	NS
0 J/L vs 1,760 J/L	S	S	S	S	NS	NS
880 J/L vs 1,760 J/L	NS	S	NS	NS	NS	NS

[a] Parameters were defined at $n = 36$, $\alpha = 0.01$, $\beta = 0.05$, and $\rho d = 50\%$. S, statistically significant difference ($P < 0.05$); NS, not statistically significant difference ($P > 0.05$).

assess the best UV doses clearly indicated that doses below 880 J liter^{-1} have little effect, if any, on pathogens and spoilage microorganisms, while for doses higher than 1,760 J liter^{-1} there is a plateau and the decimal reduction does not increase *(7)*. Currently, research in this area is aimed at studying the effect of UV doses in the range 800 to 1,600 J liter^{-1} *(17, 28, 31, 39)*.

The attempt to find alternative processing technologies to replace or augment traditional thermal methods should begin with an assessment of safety parameters. However, in developing novel technologies, sensory properties and consumer acceptance must also be given serious consideration. Even though this study has shown that UV treatment is effective for reduction of the bacterial load, UV irradiation at the wavelength 254 nm also had sensory consequences. Other studies, however, found no significant differences between pasteurized milk and UV-treated milk in terms of flavor *(17)*.

The basic design of the project was to use UV light as an additional hurdle for certain processes, such as the treatment of milk destined for further processing (e.g., fermentation, cheesemaking). Replacement of thermal pasteurization with UV would take years in the regulatory process and is thus not a practical consideration at this time. From a regulatory perspective, using UV as an add-on has a better prospect for being approved, although this has also taken years and is still in progress. The cost of the SP4 research unit is US$80,000. The biggest economic impact of UV technology to enhance the shelf life of milk would come in savings due to milk discarded for going beyond the

TABLE 9. *Sensory analysis, triangle test for 2% fat milk[a]*

	Taste			Aroma		
	Day 1	Day 8	Day 15	Day 1	Day 8	Day 15
Control vs 0 J/L	NS	NS	NS	NS	NS	NS
Control vs 880 J/L	NS	NS	NS	NS	NS	NS
Control vs 1,760 J/L	S	S	S	S	NS	NS
0 J/L vs 880 J/L	NS	S	S	NS	NS	NS
0 J/L vs 1,760 J/L	NS	NS	S	NS	NS	NS
880 J/L vs 1,760 J/L	NS	S	NS	NS	NS	NS

[a] Parameters were defined at $n = 36$, $\alpha = 0.01$, $\beta = 0.05$, and $\rho d = 50\%$. S, statistically significant difference ($P < 0.05$); NS, not statistically significant difference ($P > 0.05$).

TABLE 10. *Time in days to exceed minimum acceptable levels of psychrotrophic and mesophilic microbiota, LPC, aerobic sporeformers, coliform organisms, and titratable acidity for 3.5% milk stored at 4 and 7°C*

Treatment	Psychrotrophic (log CFU/ml > 4)		Mesophilic (log CFU/ml > 4)		LPC (log CFU/ml > 2)		Sporeformers (CFU/ml > 5)		Coliforms[a]		TA[b]	
	4°C	7°C	4°C	7°C	4°C	7°C	4°C	7°C	4°C	7°C	4°C	7°C
Control	14	7	7	7	35	28	7	7	7	7	21	14
0 J/L	7	7	7	7	>35	14	7	7	7	7	21	14
880 J/L	>35	35	>35	21	>35	>35	21	21	>35	>35	>35	28
1,760 J/L	>35	>35	>35	28	>35	>35	>35	21	>35	>35	>35	35

[a] First positive culture.

[b] TA, titratable acidity (>0.2% lactic acid).

TABLE 11. *Time in days to exceed minimum acceptable levels of psychrotrophic and mesophilic microbiota, LPC, aerobic sporeformers, coliform organisms, and titratable acidity for 2% milk stored at 4 and 7°C*

Treatment	Psychrotrophic (log CFU/ml > 4) 4°C	7°C	Mesophilic (log CFU/ml > 4) 4°C	7°C	LPC (log CFU/ml > 2) 4°C	7°C	Sporeformers (CFU/ml > 5) 4°C	7°C	Coliforms[a] 4°C	7°C	TA[b] 4°C	7°C
Control	14	7	7	7	7	14	7	7	7	7	21	14
0 J/L	14	7	7	7	21	14	>35	7	>35	7	21	14
880 J/L	>35	>35	>35	14	>35	>35	>35	>35	>35	28	>35	28
1,760 J/L	>35	>35	>35	21	>35	>35	>35	>35	>35	21	>35	28

[a] First positive culture.
[b] TA, titratable acidity (>0.2% lactic acid).

"code date" before sale in retail outlets. According to California Department of Food and Agriculture statistics for 2011 (10), 745,000,000 gallons of fluid milk was sold in stores for all classes of product. Of this, ca. 1.5% was discarded, as it could not be sold before the expiration date (shrink percentage from the California Milk Advisory Board).

Before recommendation of such nonthermal technologies as a complement or even as an alternative to heat treatment, however, more studies on the effect of UV exposure on sensory properties and on ways of reducing sensory defect associated with UV treatment are needed. In fact, UV doses applied to retain sensory characteristics have a negligible impact on enhancing the microbiological stability of milk. This, in turn, would limit the utility of UV as an alternative pasteurization method. At this point, research should concentrate on addressing the issue of negative sensory impact. This can be accomplished in two ways: reduction of UV doses and limitation of negative effect on sensory properties. In principle, the bacterial content of milk can be adequately controlled by exposure to pulsed UV light, while, in practice, the application of UV treatment to milk is challenging for two main reasons: the solids content of milk limits the penetration of UV light into the liquid, thereby reducing its efficacy, and excessive UV exposure can lead to oxidation and sensory defects in milk (42). Critical design elements in the application of UV treatment to milk have been identified as UV wavelength, intensity and dose rate, thickness of the radiation path, and flow turbulence (15, 17, 42); therefore, future research in this area should focus on these parameters.

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