The Measurement of the Effects of Container Materials on Production of Light Induced Off-Flavors in Milk Using Solid Phase Microextraction (SPME) Gas Chromatography-Mass Spectroscopy

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Introduction

Exposure of milk to light will produce off-flavors (OFs) that reduces product quality. Ultra-violet (UV) light causes a free radical reaction with unsaturated fatty acids, forming hydroperoxides that readily convert mostly to pentanal and hexanal. Because of their low sensory threshold, these malodors are readily detected in milk products. Other UV induced OFs are sulfides formed from the degradation of sulfur-containing amino acids. Dimethyldisulfide (DMDS) and dimethyl sulfide are the primary products of this reaction. The presence of these products was readily observed with the transition from glass to plastic milk storage containers.

In this study, milk was placed in similar sized containers made from both glass and a variety of plastic materials. The containers were then exposed to fluorescent lighting for a fixed time. The flavors and off-flavors in the exposed milk were extracted using headspace SPME with a Carboxen-PDMS coated Nitinol fiber followed by analysis using GC-MS. This fiber coating retains low molecular weight analytes, and allowed detection in the samples at concentration levels less than 1 ng/mL (ppb).

Materials & Methods

Milk was purchased directly from a local dairy that stores the milk in 4 mm thick 1/2 gal. glass containers. The plastic cap was wrapped in aluminum foil and the milk was stored in the dark at 4°C.

Various types of plastic containers with similar dimensions were obtained and used during the study. The containers were filled to 93% ±1% of the internal volume. The plastic types and dimension are shown in Table 1.

A 500 mL volumetric flask was filled with cold milk and spiked with an internal standard, hexanal-d12 at 5 µg/L. The milk was immediately dispensed into the containers at the volume levels listed in Table 1. Caps were covered with aluminum foil to reduce UV permeation.

The containers were placed under Sylvania Octron 32w fluorescent lights at a distance of about 10 cm to expose the milk. The exposure time was 2 hours. Table 1 shows the specifics of the containers and volumes.

After the milk was exposed for 2 hr, the containers were placed in the refrigerator at 4°C for 1 hr to assure that the milk was cooled. A total of twelve 10-mL vials were placed in a metal vial tray and inserted into a peltier cooled vial tray holder set at 4°C on the MPS II.

Five-mL of milk was transferred in duplicate into the 10 cooled vials. Milk, not exposed to light, was spiked at 5 µg/L with hexanal-d12 and dispensed in the remaining 2 vials.

The following sampling parameters in Table 2 were used to analyze the milk components.

Table 2 - SPME sampling conditions

Auto sampler:	Gerstel MPS II with cooled tray holder
Sample:	5 mL milk
Fiber:	**Carboxen-PDMS on Nitinol core**
Incubation:	50°C for 1 min with agitation
Agitation:	Rotation speed of sample at 250 rpm
Extraction:	Headspace for 15 min at 50°C with agitation
Desorption:	3 min at 300°C
Post desorption:	2 min at 280°C in needle cleaner

The following conditions in Table 3 were used to analyze the extracted components.

Table 3 – Analytical conditions

GC	Agilent 7890
GC column:	VOCOL 30m x 0.25mm ID, 1.5µm Df
Oven program:	45°C (2 min) to 100°C at 8°C/min to 140°C at 12°C/min to 180°C at 16°C.min (0.2 min)
Carrier Gas:	Helium at 1 mL/min constant flow rate
Inlet:	300°C with 0.75 mm ID liner Splitless for 0.75 min then vent at 20 mL/min
Injection port:	
Transfer line:	250°C
Detector:	MSD quadrupole, m/z 40-150
Quantitation ions:	pentanal-44; hexanal-56; dimethyldisulfide-94; hexanal-d12 -64

Table 1– Container materials and dimensions used in light exposure study

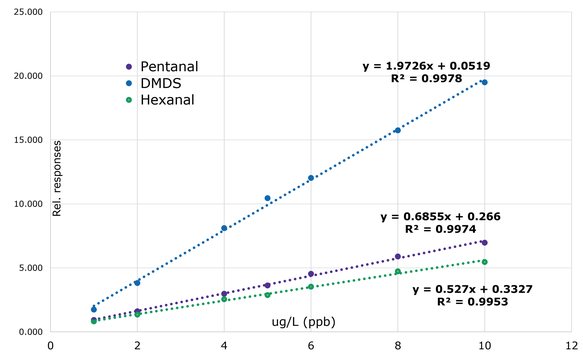
Container Material	Wall Thickness mm	Internal diameter or length mm	Internal radius or width mm	Base area mm^2	Height mm	Total surface area mm^2	Volume of milk in container mL	Internal volume of container mL	Percent of fluid volume
*PETE	0.60	26.8	13.4	563.8	115	10241	55	59	93%
**HDPE	0.80	35	17.5	961.6	81	9864	65	71	92%
Polypropylene	1.32	28.8	14.4	651.1	100	9694	50	54	93%
***white HDPE	1.50	40	40	1600.0	55	10400	67	72	93%
Glass bottle	2.00	47	23.5	1734.1	65	11327	75	80	94%

*PETE - polyethylene terephthalte ether
**HPDE – high densiy polyetheylene
***white HDPE – Opaque white filler in HPDE

Results

The Carboxen PDMS fiber on the Nitinol core is an excellent choice for this application due to the small micropores of Carboxen PDMS. The addition of salt did not increase recovery of the analytes in milk so no salt was added to the samples. Figure 1 shows he linearity of the relative response of the OFs in milk from 1-10 ug/L (ppb).

Fig. 1 – Calibration Curve of Relative Responses of OFs

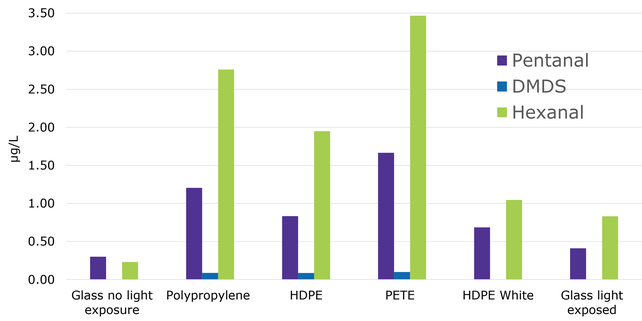


The responses from the 3 analytes have regression coefficient values in excess of 0.99 and low y intercept values.

Chromatograms of milk not exposed to light spiked with the IS (A) and milk exposed to light in a polypropylene container (B) are shown in Figure 2.

Figure 3 shows the concentration of each OF in the milk samples in µg/L after exposure to light for 2 hours stored in various types of container materials

Fig 3 – Concentration of OFs in milk after exposure to light



OF concentrations were calculated by multiplying relative response by the slope of the line (Fig. 1) minus the blank milk relative response times slope.

PETE and polypropylene appear to be more susceptible to UV light penetration. HDPE, especially impregnated with dye appears to be the UV best barrier next to glass. It was also determined that the thickness of the glass determines its effectiveness as a barrier to UV light.

Summary

The type of material used to store milk can be critical in the prevention of lipid peroxidation. This study shows that glass is still the best barrier to UV light but HDPE impregnated with a dye is a good option. In this case white dye helped to reduce OF formation, but studies have shown that yellow or pink dye may be better.

The Carboxen-PDMS fiber on the Nitinol core was capable to retain the small flavoring analytes. The micropores retain and release these analytes efficiently. The Nitinol core is highly inert and extremely durable. The new SPME coatings on the Nitinol core are highly reproducible

Fig. 2 – Chromatograms of Milk with IS not exposed to light (A) and milk exposed to light stored in a polypropylene container (B)

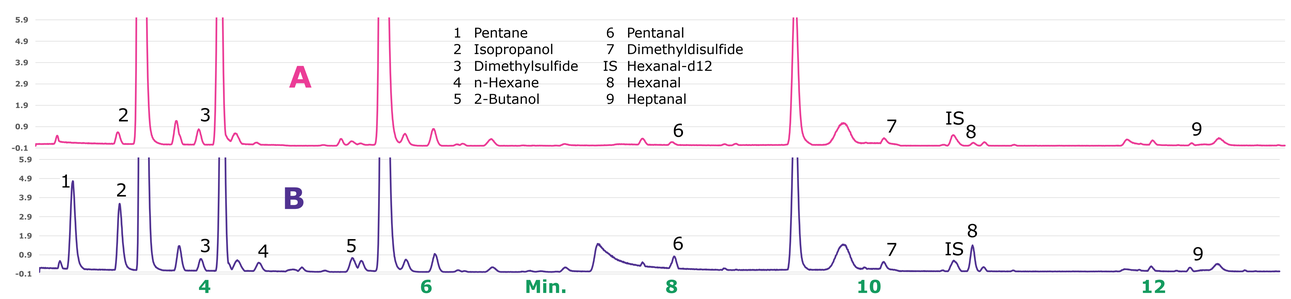


Table 4 shows the relative responses of 3 OFs in milk after exposure to light in the various storage containers. The relative responses are the average of duplicate samples. The conditions for the analysis are shown in Tables 2 and 3.

Table 4 – Relative responses of OFs in milk after exposure to light in various containers

	no light	Polypropylene	HDPE	PETE	HDPE white	Glass
Pentanal	0.206	0.826	0.572	1.142	0.470	0.282
DMDS	0.000	0.172	0.170	0.196	0.000	0.000
Hexanal	0.122	1.454	1.027	1.826	0.551	0.438

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