

LITERATURE CITED

- Bilinsky, W. S., Stedman, R. L., *Tob. Sci.* **6**, 160 (1962).
Cheng, A. L. S., Tso, T. C., Chaplin, J. F., *Crop Sci.* **11**, 580 (1971).
Chortyk, O. T., Severson, R. F., Higman, H. C., *Beitr. Tabakforsch.* **8**, 204 (1975).
Chortyk, O. T., Schlotzhauer, W. S., *Beitr. Tabakforsch.* **7**, 165, (1973).
Chu, H., Tso, T. C., Chaplin, J. F., *Agron. J.* **64**, 280 (1972).
Chu, H., Tso, T. C., *Plant Physiol.* **43**, 428 (1968).
Davis, D. L., Legg, P. D., Collins, G. B., *Crop Sci.* **10**, 545 (1970).
Ellington, J. J., Schlotzhauer, P. F., Schepartz, A. I., *J. Chromatogr. Sci.* **15**, 295 (1977).
Grunwald, C., *Phytochemistry* **14**, 70 (1975).
Grunwald, C., *Tob. Health Workshop Conf., Proc. 4th*, 679-705 (1973).
Grunwald, C., *Plant Physiol.* **45**, 663 (1970).
Hoffmann, D., Woziwodzki, H., *Beitr. Tabakforsch.* **4**, 167 (1968).
Irvine, W. J., Woolen, B. H., Jones, D. H., *Phytochemistry* **11**, 467 (1972).
Schlotzhauer, W. S., Severson, R. F., Chortyk, O. T., Arrendale, R. F., Higman, H. C., *J. Agric. Food Chem.* **24**, 992 (1976).
Schlotzhauer, W. S., Schmeltz, I., *Beitr. Tabakforsch.* **5**, 5 (1969).
Schmeltz, I., dePaolis, A., Hoffmann, D., *Beitr. Tabakforsch.* **8**, 211 (1975).
Schmeltz, I., Hoffmann, D., "Carcinogenesis Vol. I, Polynuclear Aromatic Hydrocarbons: Chemistry, Metabolism and Carcinogenesis", Raven Press, New York, N.Y., 1973, pp 225-239.
Severson, R. F., unpublished data (1976).
Severson, R. F., Ellington, J. J., Schlotzhauer, P. F., Arrendale, R. F., Schepartz, A. I., *J. Chromatogr.* **139**, 269 (1977).
Severson, R. F., Shook, M. E., Arrendale, R. F., Chortyk, O. T., *Anal. Chem.* **48**, 1866 (1976).
Stedman, R. L., Rusaniewski, W., *Tob. Sci.* **3**, 167 (1959).
Tso, T. C., Chu, H., *Argon. J.* **62**, 512 (1970).
Tso, T. C., Lowe, R., DeJong, D. W., *Beitr. Tabakforsch.* **8**, 44 (1975).

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COMMUNICATIONS

Stability of Carboxymyoglobin in Refrigerated Ground Beef

This paper describes the uptake of carbon monoxide by myoglobin in beef patties exposed to a 1% CO atmosphere and the subsequent loss of carbon monoxide when samples are placed in an air atmosphere under fluorescent illumination. The half-life for the loss of carbon monoxide from such samples is about 2 days.

Modified atmospheres for storage of fresh meats containing high concentrations of carbon dioxide (CO₂) and low concentrations of carbon monoxide (CO) are currently being studied. Atmospheres containing 1% CO and 50% CO₂ have been shown to extend both the microbiological shelf life as well as the color shelf life of refrigerated ground beef (Gee and Brown, 1978). With the incorporation of CO in gas mixtures, there is formed carboxymyoglobin (MbCO), a pigment that appears to be more resistant to oxidation than is oxymyoglobin (MbO₂) in the presence of high concentrations of CO₂ (Wolfe et al., 1976). This report summarizes a study of the stability of MbCO formed in a 1% CO atmosphere in refrigerated ground beef stored in air.

METHODS AND MATERIALS

Lean ground beef was purchased at a local retail chain store. Circular ground beef disks of uniform weight (50 ± 0.1 g), diameter (8.0 cm), and thickness (1 cm) were formed using a die and piston. The meat was stored on stainless steel mesh shelves in a 10-L desiccator jar in a 1% v/v carbon monoxide atmosphere at 2 °C for approximately 3 days. This atmosphere was chosen because of its potential of color shelf life extension without hiding bacterial spoilage or producing overly bright-red meat color (Gee and Brown, 1978). The meat was then held under a normal air atmosphere with continuous fluorescent illumination (15 W at 50 cm). The average temperature at the surface of the meat samples (Mettler TM 15 thermometer) was 1.7 °C, with or without illumination.

Extractions of myoglobin were performed by blending each ground beef patty at high speed for 60 s in a total of 100 mL of 0.2 M phosphate buffer, pH 5.92. The extract was centrifuged for 30 min at 2 °C and 14 000g. The supernatant was filtered through a glass wool plug and the volume measured. A 20-mL aliquot of the supernatant was recentrifuged for 15 min at 2 °C and 37 000g. Aliquots of this extract were scanned from 700 to 450 nm vs. water in a Cary Model 15 spectrophotometer. The sample was rescanned following saturation with CO. Using the method of Wolfe et al. (1978), data from the two scans were used to calculate the percentages of metmyoglobin (MetMb), MbCO, and MbO₂ plus deoxymyoglobin (Mb). At each sampling point, three sets of scans of extracts from each of four ground beef patties were made.

RESULTS AND DISCUSSION

A summary of the data is presented in Figure 1. Each point is the average of 12 scans. Initially, about 36% of the myoglobin in the beef patties was in the MetMb state with the remainder in the reduced forms (MbO₂ + Mb). Following a 3.4 day exposure to 1% CO, the level of MetMb fell to about 23% while the total reduced myoglobin percentage increased. The MbCO percentage initially following treatment was about 17%. Subsequent sampling of CO-treated patties exposed to air showed a steady decrease in the levels of MbCO and an increase in levels of MetMb. The half-life of MbCO in samples stored in air was found to be 2.1 days. Determination of the half-life of MbCO was important because levels of carbon

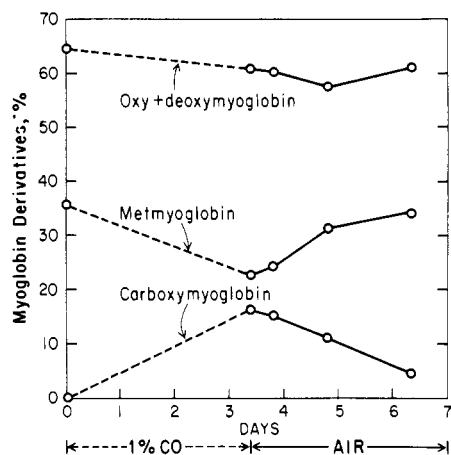


Figure 1. Relative amounts of myoglobin derivatives in ground beef samples exposed to carbon monoxide for 3.4 days, then stored in air: temperature, 2 °C; storage in air was under 15-W fluorescent illumination at 50 cm.

monoxide ingested as MbCO by the consumer can only be calculated if the rate at which CO dissociates from myoglobin occurs during the air storage period is known.

Since the rate of MbCO loss is approximately equal to

the rate in increase of MetMb, the loss of the CO ligand may lead to rapid oxidation of the resulting Mb. Sampling beyond 7 days was not useful due to extensive microbial growth as evidenced by slime formation.

LITERATURE CITED

- Gee, D. L., Brown, W. D., *J. Agric. Food Chem.* **26**, 000 (1978).
 Wolfe, S. K., Brown, W. D., Silliker, J. H., *Proc. Meat Ind. Res. Conf.*, Chicago, Ill., 1976, pp 137-148.
 Wolfe, S. K., Watts, D. A., Brown, W. D., *J. Agric. Food Chem.* **26**, 000 (1978).

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Extension of Shelf Life in Refrigerated Ground Beef Stored under an Atmosphere Containing Carbon Dioxide and Carbon Monoxide

The microbiological and color shelf lives of ground beef patties exposed to a 1% carbon monoxide, 50% carbon dioxide, plus 49% air atmosphere were significantly increased compared to similar samples exposed to air at 2 °C. Total microbial plate counts indicated nearly a 100-fold difference in bacteria per gram of meat after 6 days between samples exposed to the modified atmospheres and those exposed to air and would appear to extend the microbiological shelf life approximately 4.5 days. The fresh red color, measured with a Hunterlab Color/Difference Meter, was maintained for at least 6 days in samples exposed to modified atmospheres, while air-stored samples showed discoloration after 3 days.

In the modern day supermarket where virtually all fresh meats are prepackaged, the primary criterion used by today's consumer in meat selection is color. Although color is not a direct indicator of spoilage in meats, the formation of brown metmyoglobin (MetMb) may accompany extensive microbial growth. Spoilage in refrigerated meats is usually caused by the aerobic psychrophilic bacteria, primarily by species of *Pseudomonas*, *Flavobacterium*, and *Achromobacter* (Ayres, 1960; Jay, 1967; Haines, 1933). Spoilage has been shown to be retarded by use of atmospheres containing high concentrations of carbon dioxide (Haines, 1933; King and Nagel, 1967; Clark and Lentz, 1972; Huffman, 1974; Silliker et al., 1977). However, treatment of red meats, particularly beef, with high concentrations of carbon dioxide greatly accelerates the darkening of the meat's surface (Clark et al., 1969; Ledward et al., 1971; Silliker et al., 1977).

The use of carbon monoxide to extend the color life in fresh beef has been studied by El-Badawi et al. (1964). They showed that a mixture of 2% carbon monoxide and 98% air significantly preserved the color of fresh beef for 15 days. Under these conditions the pigment carboxymyoglobin (MbCO) is formed. MbCO, whose visual spectrum greatly resembles that of oxymyoglobin (MbO₂), is more resistant to oxidation than MbO₂. Clark et al.

(1976) studying atmospheres containing carbon monoxide and nitrogen determined that continuous exposure of beef to this atmosphere substantially extended both the color and odor shelf life. They determined that a 1% CO atmosphere was the minimum concentration required for optimum color. Besser and Kramer (1972) also reported on the beneficial effect of CO treatment on the color of beef patties.

This paper reports the effects of an atmosphere containing both carbon dioxide and carbon monoxide on color changes and microbiological counts in fresh ground beef.

METHODS AND MATERIALS

Meat and Storage. Freshly ground beef of the leanest grade was purchased from a local retail market. Patties of uniform weight (50 ± 0.1 g), diameter (8.0 cm), and thickness (1 cm) were formed. Seven control patties were stored in an air tight 10-L desiccator jar on stainless steel mesh shelves containing an air atmosphere. Seven other patties were stored in a desiccator containing 50% carbon dioxide and 1% carbon monoxide, the balance being air. Storage was at 2 °C.

Microbiological Counts. At each sampling time, a 50-g patty was placed in a sterile blender jar with 150 mL of sterile 50 mM Tris-HCl buffer, pH 6.8, and blended for