

Low-Oxygen Packaging of Fresh Meat with Carbon Monoxide: Meat Quality, Microbiology, and Safety

Daren P. Cornforth and Melvin C. Hunt

Introduction

Fresh retail meats in the United States commonly are packaged using either polyvinyl chloride (PVC) film or modified atmosphere packaging (MAP). Since 2002, carbon monoxide (CO) has been permitted as a MAP gas for use during distribution in the United States (Rulis, 2002). In 2004, CO was further permitted at low levels as a MAP gas in retail fresh meat packaging (Tarantino, 2004). Although use of CO in meat packaging applications is relatively new in the United States, meat products have been exposed to CO as a component of wood smoke for decades. CO also has been used in the United States since the 1970s as a modified atmosphere gas component for shelf life extension of iceberg lettuce during distribution (Mermelstein, 1977; Kader, 1983) and is recommended as a component of modified atmospheres to prolong shelf life of tomatoes, cauliflower, cantaloupe, citrus, and strawberries (Wolfe, 1980).

Comparison of Fresh Meat Packaging Methods

Polyvinyl chloride (PVC) was discovered in the 1920s by rubber scientist Waldo Semon, who was hired by B. F. Goodrich to develop a synthetic rubber. After World War II, PVC was used in a number of commercial applications, including packaging of fresh meats. PVC film has relatively high oxygen permeability (Landrook and Wallace, 1955), which is advantageous because meat surfaces in contact with oxygen develop an attractive bright red color due to reaction with meat myoglobin and residual blood hemoglobin to form oxymyoglobin and oxyhemoglobin, respectively. Another advantage of PVC packaging is that both the film and related equipment are relatively inexpensive and easy to use, allowing widespread use of this method in retail stores. PVC films

are thin and easily heat-sealable but are highly susceptible to punctures and tearing, leading to a significant frequency of "leaky" packages. The greater disadvantage of PVC-wrapped meats, however, is susceptibility to browning due to meat pigment oxidation and formation of metmyoglobin. Shelf life of PVC wrapped meats is only 5-7 days for steaks or roasts and less for ground meats. When surface browning due to metmyoglobin exceeds 40%, retail meats typically are discounted or discarded (Greene, Hsin & Zipsper, 1971). Brown discoloration can be avoided or minimized by vacuum packaging, which is an acceptable method for lightly pigmented cuts of pork and chicken. Vacuum packaged meats have been marketed successfully for years in many countries. However, the dark-purplish color of deoxymyoglobin in vacuum packaged retail beef has not been accepted by US consumers (Meischen, Huffman & Davis, 1987).

It is important to note that the relatively rapid browning of PVC-wrapped meats is primarily due to the increased rate of myoglobin oxidation that occurs at low oxygen concentrations (George and Stratmann, 1952; Sørheim, Grini, Nissen, Andersen & Lea, 1995), which occurs at the limit of oxygen diffusion into meat in PVC packaging and is visible as a brown sub-surface band (Cornforth, 1994). To prevent browning, meat package oxygen levels must be less than 0.15%. Oxygen levels of 0.15-2.0% predispose fresh beef products to browning (Mancini and Hunt, 2005). The elevated oxygen levels used in high-oxygen MAP will delay browning of fresh meats, compared with PVC packaging, because the depth of the bright red surface oxymyoglobin layer is increased by 3-5 mm (MacDougall and Taylor, 1975).

Faster bacterial growth is favored in PVC-wrapped meats compared with vacuum packaged meats (Seideman and Durland, 1983). Vacuum packaging lowers total plate count and favors lactobacilli, whereas pseudomonads usually dominate the spoilage microflora of PVC-wrapped meats (Pierson, Collins-Thompson & Ordal, 1970; Roth and Clark, 1972; Gill, 1983). It is recognized, however, that in PVC packaging, browning occurs due to oxygen stimulated metmyoglobin formation even while bacterial numbers are low. Dipping or injecting fresh meats with an antioxidant solution, typically containing 0.3% sodium tripolyphosphate and 500

Authors: Daren P. Cornforth, *Nutrition and Food Sciences, Utah State University, Logan, UT 84322, daren.cornforth@usu.edu*

Melvin C. Hunt, *Animal Sciences and Industry, Weber Hall, Kansas State University, Manhattan, KS 66506, bhunt@ksu.edu*

Reviewers:

James R. Claus, *University of Wisconsin-Madison*

Andrew L. Milkowski, *University of Wisconsin-Madison*

Joseph G. Sebranek, *Iowa State University*

ppm sodium ascorbate, reduces the rapid browning that occurs in meats that are still acceptable from a microbiological standpoint (Cheng, 1987; Manu-Tawiah, Ammann, Sebranek & Molins, 1991).

High-Oxygen Modified Atmosphere Packaging

To extend fresh meat shelf life, retailers have continued to search for an alternative to wrapping with PVC film. Increasingly, fresh meats are sold in high (70-80%) oxygen MAP, with 20-30% carbon dioxide (CO₂) (Eilert, 2005). Typically, co-extruded polyamide (nylon) - polyethylene films are used for high-oxygen MAP (Sørheim, Nissen & Nesbakken, 1999; John, Cornforth, Carpenter, Sørheim, Pettee & Whittier, 2004). The nylon provides strength, and the polyethylene provides gas and water vapor barrier properties and heat sealability. Meats packaged in high-oxygen MAP typically retain acceptable red color for 10-14 days of retail display, compared with 3-7 days for PVC packaged meats. The MAP film is more puncture-resistant than PVC film, but the primary economic advantage of MAP is the additional 7-10 days of red color stability, allowing retail meat packaging to occur in large volumes at central packaging facilities. Retail packages are shipped to stores in a "case-ready" format for retail display. This allows supermarkets to offer retail fresh meat products at lower cost because the expense of in-store retail meat packaging is avoided (Cornforth, 1994).

Air contains 78% nitrogen, 20.9% oxygen (O₂), 0.35% CO₂, water vapor, and traces of inert gases (Church, 1994). Compared with air, the elevated oxygen levels used in high-oxygen MAP saturate meat pigments with oxygen and slow surface metmyoglobin formation. Carbon dioxide is included in MAP systems for its antimicrobial properties. Bacterial inhibition occurs with > 20% CO₂ in MAP systems (Enfors, Molin & Ternstrom, 1979; Nissen, Sørheim & Dainty, 1996; Luño, Roncalás, Djenane & Beltrán, 2000). Australian processors were using CO₂ atmospheres to extend shelf life of fresh meat exports in the 1930s, but the process was replaced with freezing after World War II due to lower costs and longer shelf life (Bastian, McBean & Smith, 1979).

Disadvantages of high-oxygen MAP include accelerated lipid oxidation and off-flavor development (Jakobsen and Bertelsen, 2000; Jayasingh, Cornforth, Brennand, Carpenter & Whittier, 2002), bone darkening of bone-in cuts (Mancini, Hunt, Hackmeister, Kropf & Johnson, 2005), and premature browning during cooking (Tørngren, 2003; Seyfert, Mancini & Hunt, 2004a,b; John *et al.*, 2004; John, Cornforth, Carpenter, Sørheim, Pettee & Whittier, 2005). Ground beef from high-oxygen MAP developed objectionable oxidized flavors upon cooking after as few as 6 days in an 80% oxygen environment (Jayasingh *et al.*, 2002). Oxidized (rancid) flavor development can be slowed by injecting whole-muscle cuts with antioxidants, such as sodium tripolyphosphate (Seyfert *et al.*, 2004a) or rosemary extracts (Sandusky, Reynhout & Jones, 2006), but at additional expense. However, no satisfactory antioxidant treatment is currently available for ground beef in

high-oxygen MAP because non-meat ingredients are not permitted in product labeled "ground beef."

Important microbial safety concerns are involved with the premature browning that occurs during cooking of ground beef from high-oxygen MAP. High-oxygen treated ground beef appears fully cooked at temperatures as low as 57°C, which is well below the recommended internal temperature of 71°C for destruction of *Escherichia coli* O157:H7 (John *et al.*, 2004; Seyfert *et al.*, 2004b). Thus, consumers who rely on cooked color as the sole indicator of doneness may significantly undercook the product, resulting in increased food safety risk.

There are also health concerns regarding prolonged consumption of oxidized foodstuffs. The lipid peroxidation chain reaction (rancidity) yields a variety of mutagens, promoters, and carcinogens such as fatty acid peroxides, cholesterol hydroperoxide, and peroxy radicals (Ames, 1983). High-oxygen MAP beef has significantly higher levels of lipid oxidation products than beef packaged by CO-MAP or vacuum packaging (John *et al.*, 2004, 2005). However, the health risk from ingestion of oxidized food components in an occasional meal is probably low. Diets also contain antioxidant compounds, including vitamins E and C, and a variety of polyphenolic compounds in spices, fruits, and vegetables, and mammalian tissues also contain antioxidant enzymes such as glutathione peroxidase, catalase, and superoxide dismutase to combat the effects of oxygen radicals formed during metabolism (Ames, 1983). Given the quality issues associated with high-oxygen MAP meat, processors continue to search for alternative technologies to better maintain retail fresh meat acceptability, shelf life, and safety.

Low-Oxygen Packaging of Fresh Meat with Carbon Monoxide

The most recent meat packaging technology is anaerobic (essentially no oxygen) MAP with low levels (0.4%) of CO, 20 to 30% CO₂ and the remainder nitrogen (CO-MAP). This packaging method offers several advantages over aerobic packaging with PVC or high-oxygen MAP including:

1. Desirable red color stability (El-Badawi, Cain, Samuels & Anglemeier, 1964; Gee and Brown, 1978; Brewer, Wu, Field & Ray, 1994; Jayasingh, Cornforth, Carpenter & Whittier, 2001; Kusmider, Sebranek, Lonergan & Honeyman, 2002; Krause, Sebranek, Rust & Honeyman, 2003; Hunt, Mancini, Hackmeister, Kropf, Merriman & DelDuca, 2004) associated with microbial plate counts below spoilage levels for as long as 28 days for ground beef and 35 days for steaks or roasts (Hunt *et al.*, 2004; Jayasingh *et al.*, 2001).
2. Better flavor acceptability, no oxidized flavors (Jayasingh *et al.*, 2002).
3. No bone darkening (relative to high-oxygen MAP; Mancini *et al.*, 2005).

4. No premature browning during cooking (relative to high-oxygen MAP; Tørngren, 2003; Seyfert *et al.*, 2004a,b; John *et al.*, 2004, 2005).
5. Decreased growth of spoilage organisms and pathogenic bacteria in MAP compared with PVC due to combined effects of anaerobic conditions, refrigeration, and elevated CO₂ (Silliker, Woodruff, Lugg, Wolfe & Brown, 1977; Silliker and Wolfe, 1980; Sørheim *et al.*, 1999; Nissen, Alvseike, Bredholdt, Hoick & Nesbakken, 2000; Doyle and Ma, 2007; Brashears, 2007). High-oxygen MAP and CO-MAP both share this advantage compared with meats in PVC wrap.
6. Increased tenderness, due to less protein oxidation in an anaerobic environment (Lund, Lametsch, Hviid, Jensen & Skibsted, 2007), and also due to longer shelf life, allowing continuous action of endogenous tenderizing enzymes (Tørngren, 2003; Grobbel, Dikeman, Hunt & Milliken, 2007).

Disadvantages of CO-MAP are:

1. Negative image of CO by consumers because it is a potentially hazardous gas.
2. Concern that products might look fresh even though bacterial levels are high and the product is spoiled.

Studies on CO Use with Fresh Meats: 1900-1985

The increased red color stability of meats exposed to CO was recognized more than 100 years ago (Church, 1994). However, commercial application of CO in meat packaging was not then considered feasible because of possible environmental hazards for workers resulting from the dangerous ability of CO to displace oxygen from hemoglobin and reduce the oxygen-carrying capacity of blood cells.

Another widely cited concern with CO use in fresh meat packaging was that CO will remain tightly bound to meat pigments (myoglobin and residual blood hemoglobin), and the meat will remain red after bacterial numbers have reached spoilage levels. Thus, consumers could conceivably purchase meat that appeared fresh but was in fact spoiled. So, through most of the 20th century there was no commercial use of CO with fresh red meats. However, sporadic research continued on fresh meat applications of CO, ultimately leading to safe packaging processes and better understanding of time/temperature relationships affecting spoilage in MAP. El-Badawi *et al.* (1964) conducted one of the first experiments using CO in fresh meat MAP. They flushed packaged beef cuts with a mixture of 2% CO and 98% air before heat sealing and found that red color was stabilized and maintained for 15 days at 2-3°C. Clark, Lentz & Roth, (1976) reported that continuous exposure of beef to 0.5-10% CO in nitrogen extended both color and odor shelf life for more than 30 days. Gee and Brown (1978) conducted research using CO and CO₂ together in a MAP system. Beef patties exposed to 1% CO, 50% CO₂, and 49% air had 2 log

lower levels of bacteria per gram than controls after 6 days storage. The fresh red color was maintained, while samples stored in air exhibited discoloration after 3 days. Seideman, Carpenter, Smith, Dill & Vanderzant (1979) similarly reported that beef roasts stored in an atmosphere containing 1% CO had less surface discoloration and lower metmyoglobin values than roasts in modified atmospheres containing oxygen. Since metmyoglobin formation from myoglobin is an oxidative process (Greene *et al.*, 1971), CO could be considered an antioxidant compound by its ability to inhibit metmyoglobin formation and promote metmyoglobin reduction, even in presence of oxygen (Lanier, Carpenter, Toledo & Reagan, 1978). Watts, Wolfe & Brown (1978) found that CO-treated ground beef did not remain red indefinitely. When ground beef was treated with 1% CO and then exposed to air, CO was slowly lost ($T^{1/2} = 3$ days), and there was an 85% loss of CO after cooking.

Beginning in 1985, Norwegian meat processors were the first to commercially package fresh meats in MAP containing CO. Their system used 0.3-0.5% CO, 60-70% CO₂, and 30-40% nitrogen (Sørheim, Nissen, Aune & Nesbakken, 2001). This system was unique in that it combined very low CO levels ($\leq 0.4\%$), anaerobic conditions, and high levels of CO₂. With anaerobic or near anaerobic conditions, less CO was needed to maintain acceptable red color. Anaerobic conditions also inhibit aerobic bacterial growth and significantly increase microbiological shelf life of meat products (Pierson *et al.*, 1970; Roth and Clark, 1972; Gill, 1983; Seideman and Durland, 1983). Further bacterial inhibition occurs with > 20% CO₂ in MAP systems (Enfors *et al.*, 1979; Nissen *et al.*, 1996; Luño *et al.*, 2000).

Evaluation of CO Exposure Levels from CO-MAP to Workers and Consumers

The Norwegian CO-MAP system posed no significant risk of CO toxicity to workers in processing facilities or consumers (Sørheim, Aune & Nebaskken, 1997). The gas mixtures were pre-mixed to specifications and delivered in pressurized gas cylinders to packaging facilities. In the packaging process, headspace air is removed using vacuum, the modified gas mixture is flushed into the package at atmospheric pressure, and the package is rapidly heat sealed. Virtually no CO escapes into the working environment. Of course, facilities should be well ventilated and equipped with CO detectors, a common environmental safety protocol.

The quantity of CO in one or several packages also is too low to cause health concerns when packages are opened at home (Table 1) or when CO-treated meats are cooked and consumed (Table 2). Comparison of various environmental sources of CO is shown in Table 3. For modified atmosphere packages containing 0.4% CO with a headspace of 1.5 liter, opened in a typical room with a volume of 150 m³, opening of 216 packages would be required (Table 1) to exceed the EPA National Ambient Air Quality Standard of 9 ppm for 8 hr (EPA, 2007b), for a typical person inhaling 5 m³ air / 8 hr. If one consumed a large meat meal (8.8 ounces; 0.25 kg), and all the CO in a typical CO-MAP remained bound to

meat after cooking, one would consume only 3.5% of the EPA 8-hr maximum safe level (Table 2). Realistically, one would consume even less CO per meal because it is known that only 15% of bound CO remains with the meat after cooking (Watts *et al.*,

1978). Also, it is unlikely that all ingested CO is absorbed. Thus, CO exposure from the packaging process or from consumption of CO-packaged meat is well below EPA safety standards.

Table 1: Calculation of CO exposure level from opening CO-packaged meats.

The Environmental Protection Agency National Ambient Air Quality Standard for CO inhalation is 9 parts per million (ppm) per 8 hours (EPA, 2007b). The density of air at 20°C and atmospheric pressure is 1.20 kg/m³ (Anon, 2007). The density of CO at 0°C is 1.25 kg CO/m³. 9 ppm CO = 9 mg CO/kg air.

Step 1. Conversion of ppm to mg CO/m³ air.

$$\frac{9 \text{ mg CO}}{\text{kg air}} \times \frac{1.2 \text{ kg air}}{\text{m}^3} = \frac{10.8 \text{ mg CO}}{\text{m}^3 \text{ air}}$$

Step 2. Calculation of CO level in a typical chilled meat package (1.5 liter = 1,500 ml) headspace.

$$1,500 \text{ ml} \times 0.4\% \text{ CO} = \frac{6 \text{ ml CO}}{\text{package}}$$

$$\frac{6 \text{ ml CO}}{\text{package}} \times \frac{1.25 \text{ kg CO}}{\text{m}^3} \times \frac{1,000 \text{ mg}}{\text{kg}} = \frac{7.5 \text{ mg CO}}{\text{package}}$$

Step 3. Calculation of CO level after package opening and mixing with air in a typical room (7.07 meters x 7.07 meters x 3 meters high = 150 m³ air volume).

$$\frac{7.5 \text{ mg CO}}{150 \text{ m}^3} = \frac{0.05 \text{ mg CO}}{\text{m}^3}$$

Step 4. Calculation of the number of opened packages needed to exceed the EPA limit of 10.8 mg CO/m³ 8 hr (from step 1).

$$\frac{10.8 \text{ mg CO}}{\text{m}^3 \text{ 8hr}} \times \frac{\text{m}^3}{0.05 \text{ mg CO}} = \frac{216 \text{ packages}}{8 \text{ hr}}$$

Assuming a room air exchange rate of 100% per hour, one would need to open 216 bags per hour to exceed the EPA CO inhalation limit. To compare CO concentrations in terms of ppm units, a 0.4% CO-MAP package contains 4,000 ppm CO, equivalent to 7.5 mg CO per package. Upon dilution in 150 m³ air (average size room), the CO concentration is:

$$\frac{7.5 \text{ mg CO}}{150 \text{ m}^3} \times \frac{1 \text{ m}^3}{1.2 \text{ kg air}} = \frac{0.042 \text{ mg CO}}{\text{kg}} = 0.042 \text{ ppm CO}$$

This is well below the National Fire Protection Association threshold level of 35 ppm (Table 5).

Table 2: Calculation of CO exposure level from consumption of CO-packaged meats.

Step 1. Calculation of maximum safe CO exposure level.

The EPA National Ambient Air Quality Standard for CO inhalation is 9 parts per million (ppm) per 8 hours, equal to 10.8 mg CO/m³ (Table 1, step 1). The typical person breathes 5 m³ air/8 hrs. Thus the EPA maximum safe exposure level is:

$$\frac{5 \text{ m}^3 \text{ air}}{8 \text{ hr}} \times \frac{10.8 \text{ mg CO}}{\text{m}^3} = \frac{54 \text{ mg CO}}{8 \text{ hr}}$$

Step 2. Calculation of the amount of CO bound to meat.

A chilled package with 1.5 liters headspace and 0.4% CO will contain 7.5 mg CO/package (Table 1, Step 2). For a package containing 1 kg (2.2 lbs) meat, with all CO bound to meat pigments, the raw meat CO content = 7.5 mg CO/kg meat. If one consumed a large meat meal (0.25 kg; 8.8 oz), and all CO remained with the cooked meat, the consumption of CO per meal would be:

$$\frac{7.5 \text{ mg CO}}{\text{kg meat}} \times \frac{0.25 \text{ kg meat}}{\text{meal}} = \frac{1.88 \text{ mg CO}}{\text{meal}}$$

Step 3. Calculation of CO consumption per meal, relative to the EPA safe exposure level of 54 mg CO inhaled/8 hr (step 1).

$$\left[\frac{1.88 \text{ mg CO}}{\text{meal}} \div 54 \text{ mg CO} \right] \times 100 = 3.5\%$$

Table 3. Environmental Sources of Carbon Monoxide Exposure

Source	Level, ppm	Range, ppm	Reference
Cigarette Smoke (undiluted with air)	2,600	1,600-3,700	Jaffe and Chavasse, 1999
Cigarette Smoke	---	1,500-5,500	Godish, 1985
Sidestream (SS) smoke (moderate)	15	---	Willes, Fitzgerald, Permutt, Proud, Haley & Bascom, 1998
SS smoke	---	30-35	Leone, Mori, Bertanelli, Fabiano & Filippelli, 1991
Cigarette Smoke (side stream=SS)	Depends on room size	26.8-36.6 mg CO/cigarette	Adams, O'Mara-Adams & Hoffman, 1987
In home (smoking in bedroom)	37	33-45	Ott, Klepeis & Switzer, 2003
In restaurant (smoking)	5.1	0.5-9.9	Siegel, 1993
In bar (smoking)	11.6	3.1-17	Siegel, 1993
Van Exhaust	5,000	4,600-6,600	Chow, Wong & Fung, 1996
Car Exhaust	240	---	Jaffe and Chavasse, 1999
Highway Traffic-California	9.8	6.8-14.9	Ott, Switzer & Willits, 1994
Traffic Tunnel (waiting)	200	---	Chow, 1991
Parking Garage	55-85	10-165	Chow <i>et al.</i> , 1996
City Air – United States 1983	8.2	4-14	EPA, 2007a
City Air – United States 2002	2.6	1.8-4.4	EPA, 2007a
Natural Gas Fired Smokehouse	---	103-152	Cornforth <i>et al.</i> , 1998
0.4% CO-MAP	4000 ppm in package 0.042 ppm in room air after opening the package		Table 1.

Notes

ppm = parts per million. 1% CO = 10,000 ppm.

The EPA standards for maximum safe exposure level are 9 ppm CO/8 hr or 35 ppm CO/1 hr (EPA, 2007b).

For most commercial CO detectors in the United States, the audible alert is triggered above 35 ppm CO.

Effect of Packaging Method on Microbial Safety and Shelf Life

As discussed previously, fresh meats in high-oxygen MAP or CO-MAP both have longer shelf life than meats in PVC due to the inhibitory effects of CO₂ on bacterial growth. However, none of these packaging systems kill food pathogens. Meat packers, processors, and retailers have known for years that it is imperative to use good sanitation practices to minimize product contamination and to keep products cold during processing, distribution, and retail display to minimize bacterial growth. It is also imperative that consumers maintain adequate refrigeration at home and monitor internal temperature during cooking to assure that products reach 71°C, as recommended by the USDA and numerous public health authorities to assure destruction of vegetative food pathogens such as *Salmonella* or *E. coli* O157:H7. Electron beam irradiation is the only lethal process currently available for pasteurizing fresh meats without altering fresh meat properties; however, this process has limited current use. Low CO packaging also improves the color life of irradiated ground beef to complement the greatly improved microbial shelf life achieved by the irradiation process (Kusmider *et al.*, 2002).

At abuse temperatures of 10°C, CO-MAP was inhibitory to growth of *Yersinia enterocolitica*, *Listeria monocytogenes*, and *E. coli* O157:H7 but was not as inhibitory against *Salmonella* strains, indicating that temperature control is important during storage, regardless of packaging method (Nissen *et al.*, 2000). Doyle and Ma (2007) and Brashears (2007) have similarly demonstrated that growth of inoculated *E. coli* O157:H7 is inhibited in temperature-abused ground beef in CO-MAP, compared with aerobic PVC packaging. Silliker and Wolfe (1980) reported that *Clostridium botulinum* toxin production was delayed by 1 day at 27°C for pork cubes inoculated with a mixture of type A and B spores and held in modified atmospheres containing 60% CO₂ and 0.5% CO compared with samples held in air or CO₂ without CO. Growth of *Salmonella* in ground beef held at 10°C for 7 days was a thousand-fold less in modified atmospheres containing 60% CO₂ and 0.5% CO compared with samples held in air. Silliker *et al.* (1977) were the first to report a post-treatment, residual effect of CO₂-containing atmospheres to inhibit microbial growth even after fresh meat was placed in an air environment at a higher temperature. Aroma indicative of spoilage was noted, and microbial plate counts (log₁₀ colonies/cm²) exceeded 10⁸ for fresh pork loins held in air at 4°C for 4 days after 7 days storage at 1°C. However, loins stored initially in an atmosphere of 60% CO₂ + 25% O₂ + 15% nitrogen and then transferred to an air atmosphere at 4°C for 4 days had microbial plate counts of only 10⁴ per cm² and no off-odor. These studies indicate that if refrigeration was temporarily lost during distribution, causing a product temperature increase up to 10°C, fresh meats in MAP containing CO₂ or CO₂ + CO would have less growth of spoilage and pathogenic microorganisms than would fresh meats in PVC packaging. A major factor in the antimicrobial efficacy of CO₂ is its ability to penetrate the bacterial membrane, causing intracellular pH changes of greater magnitude

than would be found for similar external acidification (Wolfe, 1980).

Similar to PVC or high-oxygen MAP product, fresh meat in CO-MAP will eventually spoil. In CO-MAP, ground beef or steaks maintain bacterial numbers less than 10⁷/g (spoilage levels) for 4-5 weeks, but acceptable red appearance is maintained for at least 8 weeks (Hunt *et al.*, 2004; Jayasingh *et al.*, 2001). This emphasizes the importance of the “use or freeze by” dating system established by USDA for retail sale of meat in CO-MAP. In this regard, CO-MAP meats are similar to fresh milk. Both products have a “use-by” date on the label and will develop off-odor, indicating spoilage, if temperature abused. The extent of consumer reliance on color as an indicator of meat spoilage has probably been over-stated. Consumers will routinely purchase brown-colored meats at discount, indicating their knowledge that browning alone does not indicate spoilage. A study by Carpenter, Cornforth & Whittier (2001) showed that for regular buyers of beef products, eating satisfaction of cooked beef was unaffected by color of the raw meat. This group of consumers recognized that brown color of raw beef was not a definitive indicator of spoilage.

Recent Studies on CO Use with Fresh Meats: 1985 - Present

In Norway, the low CO packaging process grew to 60% of the retail red meat market (Sørheim *et al.*, 2001). The commercial success and safety record of the Norwegian process was a factor in the renewed interest in fresh meat packaging using CO in the United States. Pre-treatment of beef steaks with 5-100% CO followed by vacuum packaging resulted in variable red color stability, depending on CO exposure time and concentration (Brewer *et al.*, 1994; Jayasingh *et al.*, 2001). Vacuum packaged steaks pretreated with 5% CO for 24 hr retained redness for 5 weeks, while steaks and ground beef both maintained redness for the duration of the study (8 weeks) in 0.5% CO-MAP (Jayasingh *et al.*, 2001). These and previous studies (Gee and Brown, 1978; Watts *et al.*, 1978) demonstrated that process conditions affect the duration of red color stability. The red color of CO-treated meats gradually changed to a brownish discoloration when cuts were stored under aerobic conditions (Hunt *et al.*, 2004). In contrast, meats continuously held in the presence of CO in a MAP environment maintain red color for extended periods.

In response to a petition from Pactiv Corporation (Lake Forest, IL), on February 21, 2002, the U.S. Food and Drug Administration (FDA) and the USDA Food Safety and Inspection Service (USDA-FSIS) “concluded that the MAP system---described in Pactiv’s notice, and used under the conditions stated in Pactiv’s notice, would be acceptable for packaging red meat cuts and ground meat” (Rulis, 2002). The Pactiv MAP system consisted of retail fresh meat products individually wrapped in PVC film, and distributed in a “master bag”. At the retail store, the master bag was opened, and individually wrapped cuts were placed in retail display in air, and without further contact with CO. The FDA response further stated “...this MAP system complies with FDA’s

definition of a processing aid...” There is no lasting functional effect in the food and there is an insignificant amount of carbon monoxide present in the finished product under the proposed conditions of use”. Thus, 0.4% CO as a component of the Pactiv MAP system was accepted as GRAS (generally recognized as safe) with no labeling issues for fresh meats during distribution and storage prior to retail display. In addition, the approval was for meat from all livestock species.

In January 2004, Precept Foods, LLC submitted a petition to the USDA for GRAS approval of their fresh meat MAP system, containing 0.4% CO, 20 – 100% CO₂, and 0-80% nitrogen. This system differed from the Pactiv system, in that the retail package, not just the bulk pack during distribution, contained 0.4% CO. On July 29, 2004, the FDA, after consultation with USDA-FSIS, “concluded that the MAP system as described in Precept’s notice, and used under the conditions stated in Precept’s notice, would be acceptable for packaging red meat cuts and ground meat” (Tarranto, 2004). Also in July 2004, after 19 years of successful application, the use of CO for meat was discontinued in Norway to comply with regulations of European trading partners (Wilkinson, Janz, Morel, Purchas & Hendriks, 2006).

CO Use in Seafood

Hawaii International Seafood, Inc. (Honolulu) petitioned the FDA in March 1999 for approval of tasteless smoke containing 7-30% CO for preservation of tuna and other seafood (GRN 000015, as cited in Rulis, 2002). The tasteless smoke was produced by burning wood or sawdust in a smoke generator followed by filtration of the conventional smoke to remove particulate matter and taste components. The filtered smoke was collected for later use. Seafood was placed in a cold chamber, flooded with tasteless smoke, and held for a sufficient time to impart a preservative effect (Kowalski, 1999). Their petition contended that filtered smoke had been used on raw fish at cold temperatures for more than 70 years. Fish has been both hot and cold smoked for generations, and a filtered smoke has been used to cold smoke salmon in Europe and North America for decades. Salmon was treated with the filtered smoke to preserve its color and texture and to impart a light smoke taste. Tasteless smoke is a super-filtered version of the same smoke that has been used in salmon smoke houses for decades. In response to Hawaii International Seafood’s petition for use of tasteless smoke on raw tuna to preserve taste, aroma, texture, and color, the FDA had “no questions regarding the notifier’s conclusion that tasteless smoke is GRAS under the intended conditions of use” (Rulis, 2002). Clear Smoke (Anova Food Inc., Tampa, FL), filtered, but with phenolics for anti-microbial effects, is also approved for seafood and some red meat applications (Olson and Brinsmade, 2004). It is commercially available for approved food applications in pressurized cylinders containing up to 30% CO. Wood smoke, which includes CO as a component, is permitted by regulation as an ingredient in meat and poultry products under USDA regulations 9 CFR 318.7(c)(4), 381.147(c) (4) and 424.21(c) (as cited in Rulis, 2002). Combustion product gases,

which include CO as a component at a maximum level of 4.5% by volume, are approved for use in the production of beverages and other foods (except fresh meat) under FDA’s regulation 21 CFR 173.350 (CFR, 1992).

Regulatory Status of MAP Gases

Modified atmosphere packaging gases are considered from a regulatory standpoint to be processing aids, not food additives (Rulis, 2002). As such, their use is not listed on the label ingredient statement. Currently, consumers are not informed by the package itself regarding use of CO, CO₂, or elevated O₂ levels in the headspace of MAP meats. Several options are available to better inform consumers regarding food processes and treatments, including mandatory or voluntary listing of compounds used as processing aids and general consumer education by industry, media, and retail outlets. These issues currently are being discussed by both food regulatory agencies and consumers because ramifications of changing labeling policies reach beyond meat packaging.

Modified Atmosphere Packaging of Fruits and Vegetables with CO

Carbon monoxide is a fungistatic gas. It inhibits brown rot on peaches and *Botrytis* rot on tomatoes, strawberries, and grapes (Kader, 1983), presumably by virtue of its ability to interact with some cytochromes, thereby blocking oxidative decay processes (Wolfe, 1980). Carbon monoxide use (5-10%) should be combined with reduced oxygen levels of 2-4% to maximize fungistatic effects (Kader, 1983). Carbon monoxide also has been shown to inhibit brown discoloration of cut surfaces and mechanically damaged vegetable tissue. Carbon monoxide at 2-3% of modified atmospheres has been used in commercial shipments of lettuce since the 1970s (Mermelstein, 1977; Kader, 1983). Carbon monoxide has been licensed for use in the United States to prevent browning in packed lettuce (Mullan and McDowell, 2003). Carbon monoxide also has been included in the modified atmospheres of marine transport containers for some commodities (Kader, 1983), in part to kill mites and other insects (Kader, 2000). For minimally processed fruits and vegetables, packaging under slight vacuum with some addition of CO is recommended to retard discoloration (Cantwell, 2005).

Biological Effects of CO

The deleterious effects of CO occur because CO binds more strongly than oxygen to hemoglobin in red blood cells, impairing oxygen transport to tissues. Relationships between blood carboxyhemoglobin (COHb) levels (% of total hemoglobin) and human health effects are shown in Table 4. At a COHb concentration of about 2.5%, individuals with cardiovascular disease display changes in cardiac function and might report chest pain. In healthy adults, no adverse health effects have been described at CO concentrations that result in 5% or less COHb (Sørheim *et al.*, 1997). A small amount of CO is formed naturally in the body,

from breakdown of hemoproteins by the heme oxidase enzyme (Durante and Schafer, 1998). Carbon monoxide regulates blood flow and blood fluidity by inhibiting vasomotor tone, smooth muscle cell proliferation, and platelet aggregation (Durante and Schafer, 1998). Normal bodily CO production leads to a COHb concentration of about 0.5%. The average COHb level in non-smokers is 1.2-1.5% (from both endogenous and environmental CO) and 3-4% in smokers (Sørheim *et al.*, 1997). The association between CO concentration in air and health effects is shown in Table 5. No adverse effects are observed for CO exposure at levels below 35 ppm for 1 hr (Table 5).

Table 4. Association between blood carboxyhemoglobin (COHb) levels and health effects (Sørheim *et al.*, 1997).

COHb, % of total	Observed Health Effects
Above 50	Unconsciousness, lethal if not treated
Above 30	Headache, nausea, vomiting, dizziness
Above 10	Life threatening for heart and lung patients. Headache in others
Above 5	Reduced maximum oxygen consumption during exercise in healthy people. Reduced visual perception, learning ability, and fine motor performance. Also, CO exposure of pregnant women at this level may affect the fetus
Above 2.9	Angina patients endure less physical stress before an attack
Above 2.3	Reduced physical work capacity and endurance
Above 2.0	Possible reduction in attention and ability to concentrate. Signs of local oxygen deficit and onset of chest pain in heart patients

Table 5. Association between CO levels in air (parts per million, ppm) and health effects (National Fire Protection Association, 2007).

CO (ppm)	Symptoms
35	No adverse effects within 8 hr
200	Mild headache after 2-3 hr exposure
400	Headache and nausea after 1-2 hr
800	Headache, nausea, and dizziness after 45 min; collapse after 2 hr
1000	Loss of consciousness after 1 hr
1600	Headache, nausea, and dizziness after 20 min; unconsciousness after 30 min
3200	Headache, nausea, and dizziness after 5-10 min; unconsciousness after 30 min
12,800	Immediate physiological effects; unconsciousness and danger of death after 1-3 min

Conclusions

Low-oxygen packaging of fresh meat with CO improves red color stability compared with meats in high-oxygen MAP or PVC. Flavor is improved (less oxidized flavor) for meat in low CO compared to high-oxygen MAP. Premature browning (a food safety issue) during cooking occurs nearly 100% of the time in meat packaged in high-oxygen MAP and to a lesser extent in PVC-wrapped meats at longer storage times. This phenomenon does not occur in meats packaged in CO-MAP or vacuum. Risk of CO-toxicity from the packaging process or from consumption of CO-treated meats is negligible. Red color can be maintained in low-CO treated meats that have spoiled, emphasizing the need for adherence to label instructions for product shelf life and the use of odor and overall appearance as spoilage indicators. Both MAP methods (high-oxygen and low-CO) are inhibitory to growth of spoilage and pathogenic bacteria during refrigerated storage compared with meats wrapped in PVC. Due to the residual effect of CO₂ treatment to inhibit bacterial growth even after removal from packaging or when storage temperature is raised (Silliker *et al.*, 1977), fresh meats in MAP containing CO₂ or CO₂ + CO would have less growth of spoilage and pathogenic microorganisms than meats in PVC packaging, if temperature control was temporarily lost during distribution. Overall, inclusion of CO as a component of MAP systems has both advantages and disadvantages that must be thoroughly considered to develop a packaging technology benefiting both consumers and the meat industry.

References

- Adams, J.D., O'Mara-Adams, K.J., & Hoffman, D. (1987). Toxic and carcinogenic agents in undiluted mainstream smoke and sidestream smoke of different types of cigarettes. *Carcinogenesis*, 8, 729-731.
- Ames, B.N. (1983). Dietary carcinogens and anticarcinogens – Oxygen radicals and degenerative diseases. *Science*, 221, 1256-1263.
- Anonymous (2007). Physical properties online calculator. <http://members.aol.com/engware/calcl1.htm>. Accessed Dec. 2, 2007.
- Bastian, J., McBean, M.D., & Smith, M.B. (1979). *50 Years of Food Research* (pp. 20-23). Melbourne: CSIRO.
- Brashears, M. (2007). Personal communication.
- Brewer, M.S., Wu, S., Field, R.A., & Ray, B. (1994). Carbon monoxide effects on color and microbial counts of vacuum packaged beef steaks in refrigerated storage. *Journal of Food Quality*, 17, 231-236.
- Cantwell, M. (2005). Postharvest handling systems: minimally processed fruits and vegetables. University of California Cooperative Extension Vegetable Research and Information Center. <http://vric.ucdavis.edu/selectnewtopic.minproc.htm>. Accessed Dec. 2, 2007.
- Carpenter, C.E., Cornforth, D.P., & Whittier, D. (2001). Consumer preferences for beef color and packaging did not affect eating satisfaction. *Meat Science*, 57, 359-363.
- Code of Federal Regulations (1992). Combustion product gas. 21 C FR 173.350. pp. 125-126. U.S. Government Printing Office, Washington, D.C.
- Cheng, C. (1987). Process for prepackaging fresh meat. U.S. patent no. 4,683,139.

- Chow, W.K. (1991). Prediction of CO level near vehicular tunnel with waiting queue. *Journal of Environmental Engineering*, 117, 116-125.
- Chow, W.K., Wong, L.T., & Fung W.Y. (1996). Field study on the indoor thermal environment and carbon monoxide levels in a large underground car park. *Tunneling and Underground Space Technology*, 11, 333-343.
- Church, N. (1994). Developments in modified-atmosphere packaging and related technologies. *Trends in Food Science and Technology*, 5, 345-352.
- Clark, D.S., Lentz, C.P., & Roth, L.A. (1976). Use of carbon monoxide for extending shelf-life of prepackaged fresh beef. *Canadian Institute of Food Science and Technology Journal*, 9, 114-117.
- Cornforth, D.P. (1994). Color: Its basis and importance. In A.M. Pearson, & T.R. Dutson, *Advances in meat research* (Vol. 9) (pp. 34-78). London: Blackie Academic & Professional.
- Cornforth, D.P., Rabovitsch, J.K., Ahuja, S., Wagner, J.C., Hanson, R., Cummings, B., & Chudnovsky, Y. (1998). Carbon monoxide, nitric oxide, and nitrogen dioxide levels in gas ovens related to surface pinking of cooked beef and turkey. *Journal of Agricultural and Food Chemistry*, 46, 255-261.
- Doyle, M. & Ma, L. (2007). Personal communication.
- Durante, W. & Schafer, A.I. (1998). Carbon monoxide and vascular cell function (review). *International Journal of Molecular Medicine*, 2, 255-262.
- Eilert, S.J. (2005). New packaging technologies for the 21st century. *Meat Science*, 71, 122-127.
- El-Badawi, A.A., Cain, R.F., Samuels, C.E., & Anglemeier A.F. (1964). Color and pigment stability of packaged refrigerated beef. *Food Technology*, 18, 159-163.
- Enfors, S.O., Molin, G., & Ternstrom, A. (1979). Effect of packaging under carbon dioxide, nitrogen, or air on the microflora of pork stored at 4°C. *Journal of Applied Bacteriology*, 47,197-208.
- EPA (2007)a. Air Trends - carbon monoxide - National trends in CO levels. <http://www.epa.gov/airtrends/carbon.htm>. Accessed Dec. 7, 2007.
- EPA (2007)b. National ambient air quality standards (NAAQS). <http://www.epa.gov/air/criteria.html>. Accessed Dec. 9, 2007.
- Gee, D.L. & Brown, W.D. (1978). Extension of shelf life in refrigerated ground beef stored under an atmosphere containing carbon dioxide and carbon monoxide. *Journal of Agricultural and Food Chemistry*, 26, 274-276.
- George, P. & Stratton, C.J. (1952). The oxidation of myoglobin to metmyoglobin by oxygen. 2. The relation between the first order rate constant and the partial pressure of oxygen. *Biochemical Journal*, 51, 418-425.
- Gill, C.O. (1983). Meat spoilage and evaluation of the potential storage life of fresh meat. *Journal of Food Protection*, 46, 444-452.
- Godish, T. (1985). *Air quality*. Chelsea, Michigan: Lewis Publishers, Inc.
- Greene, B.H., Hsin, I., & Zipser M.W. (1971). Retardation of oxidative color changes in raw ground beef. *Journal of Food Science*, 36, 940-942.
- Grobbel, J.P., Dikeman, M.E., Hunt, M.C., & Milliken, G.A. (2007). Different packaging environments alter tenderness and sensory traits from non-enhanced and injection-enhanced beef. In *Proceedings 53rd international congress of meat science and technology* (pp. 527-528), 5-10 August 2007, Beijing, China.
- Hunt, M.C., Mancini, R.A., Hachmeister, K.A., Kropf, D.H., Merriman, M., & DelDuca, G. (2004). Carbon monoxide in modified atmosphere packaging affects color, shelf life, and microorganisms of beef steaks and ground beef. *Journal of Food Science*, 69, C45-C52.
- Jaffe, D. & Chavasse, L. (1999). Comparing the CO content of cigarette smoke and auto exhaust using gas chromatography. *Journal of College Science Teaching*, 172-176.
- Jakobsen, M. & Bertelsen, G. (2000). Color stability and lipid oxidation of fresh beef- Development of a response surface model for predicting the effects of temperature, storage time, and modified atmosphere composition. *Meat Science*, 54, 49-57.
- Jayasinh, P., Cornforth, D.P., Carpenter, C.E., & Whittier, D. (2001). Evaluation of carbon monoxide (CO) treatment in modified atmosphere packaging or vacuum packaging to increase color stability of fresh beef. *Meat Science*, 59, 317-324.
- Jayasinh, P., Cornforth, D.P., Brennand, C.P., Carpenter, C.E., & Whittier, D.R. (2002). Sensory evaluation of ground beef stored in high-oxygen modified atmosphere packaging. *Journal of Food Science*, 67, 3493-3496.
- John, L., Cornforth, D.P., Carpenter, C.E., Sørheim, O., Pettee, B., & Whittier, D.R. (2004). Comparison of color and thiobarbituric acid (TBA) values of cooked hamburger patties after storage of fresh beef chubs in modified atmospheres. *Journal of Food Science*, 69, 608-614.
- John, L., Cornforth, D.P., Carpenter, C.E., Sørheim, O., Pettee, B., & Whittier, D.R. (2005). Color and thiobarbituric acid values of cooked top sirloin steaks in modified atmospheres of 80% oxygen, or 0.4% carbon monoxide, or vacuum. *Meat Science*, 69, 441-449.
- Kader, A.A. (1983). Physiological and biochemical effects of carbon monoxide added to controlled atmospheres on fruits. *Acta Horticulturae*, 138, 221-225.
- Kader, A.A. (2000). Advances in CA/MA applications. *Perishables Handling Quarterly, Issue 104 (November)*, 8-9.
- Kowalski, W.R. (1999). Process for manufacturing tasteless super-purified smoke for treating seafood to be frozen and thawed. U.S. patent no. 5,972,401.
- Krause, T.R., Sebranek, J.G. Rust, R.E., & Honeyman, M.S. (2003). Use of carbon monoxide packaging for improving the shelf life of pork. *Journal of Food Science*, 68, 2596-2603.
- Kusmider, E.A., Sebranek, J.G., Lonergan, S.M., & Honeyman, M.S. (2002). Effects of carbon monoxide packaging for improving the shelf life of irradiated ground beef. *Journal of Food Science*, 67, 3463-3468.
- Landrock, A.H. & Wallace G.A. (1955). Discoloration of fresh red meat and its relationship to film oxygen permeability. *Food Technology*, 9, 194.
- Lanier, T. C., Carpenter, J. A., Toledo, R. T., & Reagan, J.O. (1978). Metmyoglobin reduction in beef systems as affected by aerobic, anaerobic and carbon-monoxide-containing environments. *Journal of Food Science*, 43, 1788-1792, 1796.
- Leone, A., Mori, L., Bertanelli, F., Fabiano, P., & Filippelli, M. (1991). Indoor passive smoking: its effect on cardiac performance. *International Journal of Cardiology*, 33, 247-251.
- Lund, M.N., Lametsch, R., Hviid, M.S., Jensen, O.N., & Skibsted, L.H. (2007). High-oxygen packaging atmosphere influences protein oxidation and tenderness of porcine *longissimus dorsi* during chill storage. *Meat Science*, 77, 295-303.
- Luño, M., Roncalés, P., Djenane, D., & Beltrán, J.A. (2000). Beef shelf life in low O₂ and high CO₂ atmospheres containing different low CO concentrations. *Meat Science*, 55, 413-419.
- MacDougall, D.B. & Taylor, A.A. (1975). Colour retention in fresh meat stored in oxygen – a commercial scale trial. *Journal of Food Technology*, 10, 339-347.
- Mancini, R.A. & Hunt, M.C. (2005). Current research in meat color. *Meat Science*, 71, 100-121.
- Mancini, R.A., Hunt, M.C., Hachmeister, K.A., Kropf, D.H., & Johnson, D.E.. (2005). Exclusion of oxygen from modified atmosphere packages limits beef rib and lumbar vertebrae marrow discoloration during display and storage. *Meat Science*, 69, 493-500.
- Manu-Tawiah, W., Ammann, L.L., Sebranek, J.G., & Molins, R.A. (1991). Extending the color stability and shelf life of fresh meat. *Food Technology*, 45(3), 94-102.

- Meischen, H.W., Huffman, D.L., & Davis, G.W. (1987). Branded beef – product of tomorrow today. In *Proceedings 40th reciprocal meat conference* (pp. 37-46), 28 June – 1 July, 1987, Saint Paul, Minnesota.
- Mermelstein, N.H. (1977). Success or failure? How the food technology industrial achievement award winners have fared over the years. *Food Technology*, 31(7), 36-58.
- Mullan, M. & McDowell, D. (2003). Modified atmosphere packaging. In R. Coles, D. McDowell, & M. Kirwan, *Food packaging technology* (summarized at <http://www.dairyscience.info/map-science.htm>), London: Blackwell Publishing.
- National Fire Protection Association (2007). http://www.enmetgasdetection.com/articles/CO_fact_sheet.htm#Table1. Accessed Dec. 6, 2007.
- Nissen, H., Alvseike, O., Bredholt, S., Hoick, A., & Nesbakken, T. (2000). Comparison between the growth of *Yersinia enterocolitica*, *Listeria monocytogenes*, *Escherichia coli* O157:H7 and *Salmonella* spp. in ground beef packaged by three commercially used packaging techniques. *International Journal of Food Microbiology*, 59(3), 211-220.
- Nissen, H., Sørheim, O., & Dainty, R. (1996). Effects of vacuum, modified atmospheres and storage temperature on the microbial flora of packaged beef. *Food Microbiology*, 13, 183-191.
- Olson, B. E., & Brinsmade, D. B. (2004). Seafood preservation process. *U.S. Patent No. 6,777,012*. Washington, DC: U.S. Patent and Trademark Office.
- Ott, W., Switzer, P., & Willits, N. (1994). Carbon monoxide exposures inside an automobile traveling on a urban arterial highway. *Journal of the Air and Waste Management Association*, 44, 1010-1018.
- Ott, W. R., Klepeis, N. E., & Switzer, P. (2003). Analytical solutions to compartmental indoor air quality models with application to environmental tobacco smoke concentrations in a house. *Journal of the Air and Waste Management Association*, 53, 918-936.
- Pierson, M. D., Collins-Thompson, D. L., & Ordal, Z. J. (1970). Microbiological, sensory, and pigment changes of aerobically and anaerobically packaged beef. *Food Technology*, 24, 1171-1175.
- Roth, L. A., & Clark, D. S. (1972). Studies on the bacterial flora of vacuum-packaged fresh beef. *Canadian Journal of Microbiology*, 18,1761-1766.
- Rulis, A. M. (2002, February 21). Agency response letter, re: GRAS notice no. GRN 000083. Retrieved December 7, 2007, from U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition Web Site: <http://vm.cfsan.fda.gov/~rdb/opa-g083.html>
- Sandusky, C. L., Reynhout, G. S., & Jones, T. S. (2006). Method of extending color life of modified atmosphere packaged fresh red meat using Labiatae plant extracts. *U.S. Patent No. 7,037,543*. Washington, DC: U.S. Patent and Trademark Office.
- Seideman, S. C., Carpenter, Z. L., Smith, G. C., Dill, C. W., & Vanderzant, C. (1979). Physical and sensory characteristics of beef packaged in modified gas atmospheres. *Journal of Food Protection*, 42, 233-239.
- Seideman, S. C., & Durland, P. R. (1983). Vacuum packaging of fresh beef – A review. *Journal of Food Quality*, 6, 29-47.
- Seyfert, M., Mancini, R. A., & Hunt, M. C. (2004a). Internal premature browning in cooked ground beef patties packaged from high oxygen modified atmosphere packaging. *Journal of Food Science*, 69, 142-146.
- Seyfert, M., Mancini, R. A., & Hunt, M. C. 2004b. Internal premature browning in cooked steaks from enhanced beef round muscles packaged in high oxygen and ultra-low oxygen modified atmospheres. *Journal of Food Science*, 69, 721-725.
- Siegel, M. (1993). Involuntary smoking in the restaurant workplace. A review of employee exposure and health effects. *Journal of the American Medical Association*, 270, 490-493.
- Silliker, J. H., Woodruff, R. E., Lugg, J. R., Wolfe, S. K., & Brown, W. D. (1977). Preservation of refrigerated meats with controlled atmospheres: Treatment and post-treatment effects of carbon dioxide on pork and beef. *Meat Science*, 1, 195-204.
- Silliker, J. H., & Wolfe, S. K. (1980). Microbiological safety considerations in controlled-atmosphere storage of meats. *Food Technology*, 34(3), 59-63.
- Sørheim, O., Aune, T., & Nesbakken, T. (1997). Technological, hygienic and toxicological aspects of carbon monoxide used in modified-atmosphere packaging of meat. *Trends in Food Science and Technology*, 8, 307-312.
- Sørheim, O., Grini, J. A., Nissen, H., Andersen, H. J., & Lea, P. (1995). Pork loins stored in carbon dioxide – colour and microbiological shelf life. *Fleischwirtschaft*, 75, 679-681.
- Sørheim, O., Nissen, H., Aune, T., & Nesbakken, T. (2001). Use of carbon monoxide in retail meat packaging. *Proceedings 54th reciprocal meat conference* (pp. 47-51), 24-28 July, 2001, Indianapolis, Indiana.
- Sørheim, O., Nissen, H., & Nesbakken, T. (1999). The storage life of beef and pork packaged in an atmosphere with low carbon monoxide and high carbon dioxide. *Meat Science*, 52, 157-164.
- Tarantino, L. M. (2004, July 29). Agency response letter, re: GRAS notice no. GRN 000143. Retrieved December 7, 2007, from U.S. Food and Drug Administration, Center for Food Safety and Applied Nutrition Web Site: <http://vm.cfsan.fda.gov/~rdb/opa-g143.html>
- Tørngren, M. A. (2003). Effect of packaging method on color and eating quality of beef loin steaks. In *Proceedings 49th international congress of meat science and technology* (pp. 5495-496), Campinas, Brazil.
- Watts, D. A., Wolfe, S. K., & Brown, W. D. (1978). Fate of [¹⁴C] carbon monoxide in cooked or store ground beef samples. *Journal of Agricultural and Food Chemistry*, 26, 210-214.
- Wilkinson, B. H. P., Janz, J. A. M., Morel, P. C. H., Purchas, R. W., & Hendriks, W. H. (2006). The effect of modified atmosphere packaging with carbon monoxide on the storage quality of master-packaged fresh pork. *Meat Science*, 73, 605-610.
- Willes, S. R., Fitzgerald, T. K., Permutt, T., Proud, D., Haley, N.J., & Bascom, R. (1998). Acute respiratory response to prolonged, moderate levels of sidestream tobacco smoke. *Journal of Toxicology and Environmental Health A*, 53, 193-209.
- Wolfe, S. K. (1980). Use of CO- and CO₂-enriched atmospheres for meats, fish, and produce. *Food Technology*, 34(3), 55-58, 63