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EFFECT OF PACKAGE BARRIER ON THE OXIDATIVE STABILITY OF VACUUM PACKAGED FRESH PORK PATTIES SUBJECTED TO "IN-PACKAGE" HEAT TREATMENT

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BY

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ABSTRACT

EFFECT OF PACKAGE BARRIER ON THE OXIDATIVE STABILITY OF VACUUM PACKAGED FRESH PORK PATTIES SUBJECTED TO "IN-PACKAGE" HEAT TREATMENT

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The effects of oxygen barrier (nylon, saran, SiO_2), and salt (1.5%) on lipid oxidation in vacuum packaged (VP), in-package pasteurized (PA) pork patties were evaluated. The influence of VP and PA on oxygen permeation rate (OTR), seal strength (SS), mechanical strength, and sealing integrity of these oxygen barriers was also investigated.

Both TBARS value (TBARS) and hexanal content (HEX) increased linearly (R^2=0.74 to 0.99) with storage time (1, 15, 30,60, 90, 120 days). Conversion factors of 1.76 (nylon) and 1.46 (saran) [(mg/kg)/(ppm)] were obtained when converting HEX into TBARS. Oxygen barrier significantly ($P<0.0001$) reduced the lipid oxidation of the patties during storage. OTR of the barriers were SiO_2 < saran << nylon. During 120 days of storage at 3°C, headspace CO_2 (CO_2/HS) declined but headspace O_2 (O_2/HS) stayed the same in all 3 barriers. TBARS and HEX of SiO_2 packaged samples had almost no change. Salt (1.5%) reduced the HEX both in saran and SiO_2 samples. High within-treatment correlation coefficients between lipid oxidation (TBARS & HEX) and O_2/HS were found in both nylon and saran packaged samples.

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INTRODUCTION

Among the currently important areas of consumers' interest and preference are "fresh" foods. Consumers perceive precooked refrigerated meat products to be fresher and more convenient than frozen products. To convey a fresh or homemade appeal many of the new refrigerated meat products are microwaveable, formulated without any or limited preservatives, vacuum packaged and heat pasteurized.

In the absence of nitrite, and because these products are pasteurized and vacuum packaged, they provide a suitable medium for the growth of clostridia or other psychotropic microorganisms, particularly if there is any temperature abuse (Sofos, 1986a). In-package pasteurization has been reported to extend product shelf life by eliminating microbial re-contamination, oxidation, and by increasing water retention (Siegel, 1982; Simunovic et al., 1985b; Jones et al., 1987; Smith & Alvarez, 1988; Prabhu et al., 1988; Stites et al., 1989).

Besides microbial growth, the shelf-life of these products is also greatly effected by physicochemical changes which include lipid oxidation, discoloration, and textural changes. These changes can be effected by the presence of oxygen in the product. Vacuum level and packaging film oxygen barrier combinations are critical to maintain an acceptable flavor and color in meat products (Terlizzi et al., 1984). Smith and Alvarez (1988) reported that

2-Thiobarbituric acid (TBA) numbers will double within 2-4 hours after opening uncured, vacuumed turkey products. This indicates that these products are particularly sensitive to oxygen. Unfortunately, no data have been reported relative to the effect of a packaging oxygen barrier on lipid oxidation in uncured, vacuum packaged, in-package pasteurized meat products. The presence of salt also affects lipid oxidation in precooked meat products (Rhee et al., 1983a; Matlock et al., 1984a; Greene and Price, 1975; Ang and Young, 1989). Different results have been reported concerning the effect of salt in meat products, but its action is still poorly understood (Gray and Weiss, 1988).

In-package pasteurization using steam or hot water will increase the moisture content of many plastic barriers and will deteriorate their barrier properties. Loss of oxygen barrier will change not only the rate of lipid oxidation but also may influence the nature of the bacterial population. High temperature processing may cause loss of seal integrity and loss of the mechanical strength of the packaging system, both of which may result in a serious food safety problem.

The specific objectives of this study are: 1. To study the effects of different oxygen barriers and headspace O_2 on the oxidative stability of vacuum packaged pork patties during 120 days of refrigerated storage. 2. To study the effects of salt on lipid oxidation of vacuum packaged pork patties during refrigerated storage for 120 days. 3. To

study the effects of 24 hours extended storage (no barrier) on lipid oxidation in vacuum packaged pork patties during 120 days of refrigerated storage. 4. To study the change in oxygen permeation rate, mechanical properties, and sealing integrity of three barrier materials, nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP after vacuum packaging and in-package pasteurization.

REVIEW OF LITERATURE

Lipids and Their Oxidation

Lipids in muscle structure

Lipids in meat can be classified as adipose or intermuscular, and as intramuscular or tissue lipids. Intermuscular lipids are generally stored in specialized connective tissues, while the intramuscular lipids are normally associated with protein and composed of a large amount of phospholipids which are widely distributed throughout the muscle tissues (Love and Pearson, 1971). Triglycerides are the principal lipid components of adipose tissue, whereas phospholipids are found in higher amounts in tissue lipids.

Triglycerides are the most abundant of the lipids. According to Hornstein et al. (1961), beef muscle contains 2.0-4.0% and pork muscle contains 5.0-7.0% triglycerides. Triglycerides are composed of a molecule of glycerol and three fatty acids each attached to a hydroxyl of glycerol via an ester linkage. They are mainly straight-chained, even-numbered carbon fatty acids, typically containing sixteen and eighteen carbon atoms (Dugan, 1971). The most abundant and widespread unsaturated fatty acid is oleic acid, whereas the most prominent saturated fatty acid is palmitic acid (Pearson et al., 1977). The phospholipid content is 0.8-1.0% in beef and 0.7-0.9% in pork (Hornstein,

1961). Though the phospholipid content of meat is relatively small, the susceptibility of phospholipids to oxidation makes them important to meat flavor. The lability of the phospholipid is a result of their high unsaturated fatty acid content. Over 50% of the fatty acids in phospholipids contain two or more double bonds, while only 10% of the fatty acids of triacylglycerols have two or more double bond (Hornstein et al., 1961). O'Keefe et al. (1968) found that the percentage of unsaturated fatty acids in phospholipids was 70.47%, 68.23%, and 70.36, whereas the unsaturated fatty acid percentage of neutral lipids was 52.47%, 51.93%, and 54.08% for beef semitendinosus, longissimus dorsi, and triceps brachii muscles, respectively. They also found a high percentage of $C_{20:3}$ (2.9-3.99%) and $C_{20:4}$ (12.54-15.23%) fatty acids in phospholipid in contrast to their near absence in the neutral lipid fraction. Thus, phospholipids are characterized by their high level of polyunsaturated fatty acids. In addition, the greatest concentration of polyunsaturated fatty acids (PUFAS) is found in the phosphatidyl ethanolamine (PE) fraction (17-43%). Phosphatidyl choline (PC) and sphingomyelin have 17-25% and 1-4% of PUFAS, respectively (Body and Shorland, 1974).

Dietary influences are more controllable and are more obvious in nonruminants than in ruminants. Hilditch and Williams (1964) showed that the polyunsaturated fatty acids of meat triglycerides from ruminants were fairly constant at

3-4% of the total fatty acid content and were influenced very little by the diet (Shorland, 1952). On the other hand, the PUFAS triglycerides from nonruminants vary directly with the diet. Therefore, it is important to recognize that in all nonruminant species, dietary fatty acids determine tissue fatty acid composition (Gibson and Worthington, 1977). Swine (nonruminant animals) are unable to hydrogenate unsaturated fatty acids. Consequently, lipids are deposited unaltered in swine adipose tissue (Allen et al., 1976). Ruminant animals, on the other hand, (through rumen microbial modification) hydrogenate the unsaturated fatty acids and are not as sensitive to dietary influence.

Fatty acid composition and degree of unsaturation of pork carcass fat may vary also with anatomical location. Kuchmak and Dugan (1965) analyzed the fatty acid composition of phospholipids from the belly, ham, loin and rib areas in pork. They found that the belly muscles had the highest linoleic ($C_{18:2}$) and arachidonic acid ($C_{20:4}$) concentrations and that the ham had slightly more linoleic acid ($C_{18:2}$) than the loin and rib. The loin and rib had similar amounts. The linolenic acid ($C_{18:3}$) content of both rib and loin was similar and the belly had slightly less, and the ham the least. Jeremiah (1982) also reported that back fat samples had a higher percentage of total unsaturated fatty acids and a lower percentage of total saturated fatty acids than did belly fat samples.

Mechanisms of lipid oxidation

Autoxidation of unsaturated fatty acids is believed to proceed by a free radical mechanism which can be described in terms of an initiation, propagation, and termination process (Frankel, 1980) as follows:

$$\text{Initiation: RH.} \xrightarrow{\text{initiator}} \text{R.} + \text{H.}$$

$$\text{Propagation: R.} + O_2 \longrightarrow \text{ROO.}$$

$$\text{ROO.} + \text{RH} \longrightarrow \text{ROOH} + \text{R.}$$

$$\text{Termination: ROO.} + \text{ROO.} \longrightarrow \text{ROOR} + O_2$$

$$\text{R.} + \text{R.} \longrightarrow R_2$$

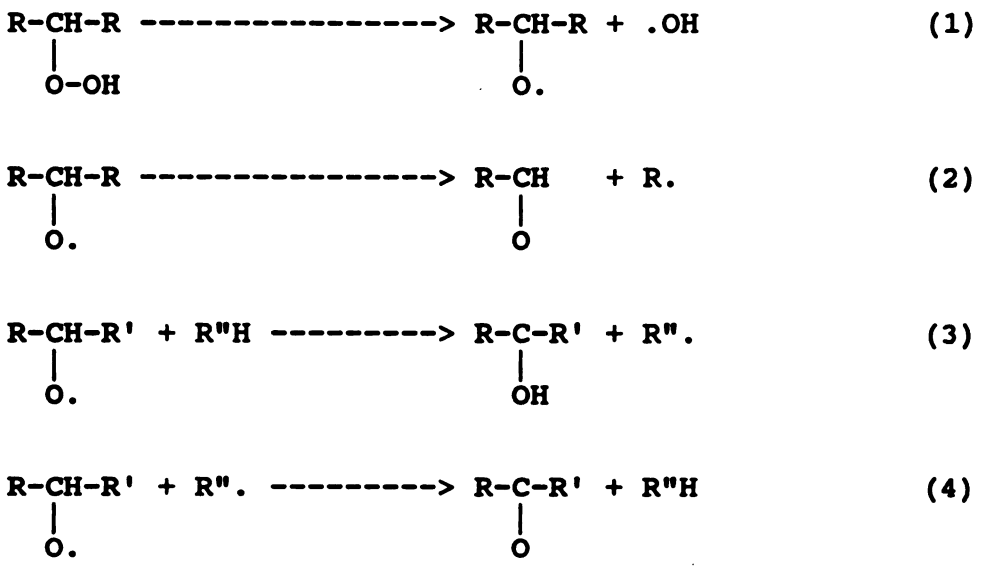
$$\text{R.} + \text{ROO.} \longrightarrow \text{ROOR}$$

Free radical formation in the initiation step may be catalyzed by direct thermal dissociation, metal catalysis and exposure to light (Lundberg, 1962; Gray, 1978; Frankel, 1980). Once the relatively stable "allyl radical" (R. is formed, it can react with oxygen to form the peroxy radical (ROO.). The peroxy radical may attack another allylic hydrogen (H.) on an unsaturated fatty acids (RH), so a peroxide (ROOH) will be formed and the allyl (R.) radical will again be regenerated. Once the propagation step has started, it propagates continuously, and about one hundred hydroperoxides (ROOH) will be produced before the radical (R., ROO.) terminates with another free radical (Bolland and Gee, 1946). Furthermore, many different isomers of

hydroperoxides will be formed after the rearrangement of e^- (delocalization of the e^-) within these free radicals (R.) and the recombination between radicals has been completed.

Besides free radical autoxidation, there is a photosensitized oxidation, which is not initiated by a free radical process. It is initiated by a different mechanism than that of autoxidation (Frankel, 1984). A sensitizer such as chlorophyll becomes excited when exposed to light energy. In coupling with triplet (ground) state oxygen, the sensitizer transfers its energy to oxygen and an excited singlet oxygen is formed. Singlet oxygen reacts directly with carbon-carbon double bonds by a concerted 'ene' addition and hydroperoxides are formed. An even distribution of hydroperoxides would be expected; however, the result (Frankel, 1984) is an uneven distribution in linoleate and linolenate due to the formation of cyclic peroxides (Neff et al., 1982).

Hydroperoxides, the primary products of autoxidation or photosensitized oxidation, are believed to be decomposed by many routes leading to the formation of a variety of volatile and non-volatile secondary products (Kenny, 1962). Although fat hydroperoxides are generally tasteless and odorless, their decomposition products have a great impact on flavor and lead to the formation of a variety of volatile and nonvolatile secondary products, i.e., aldehydes, ketones, acids, alcohols, hydrocarbons, lactones and esters (Kenny, 1962; Frankel, 1980, 1982) as follows:



The first step involves a homolytic cleavage of the O-O bond to yield the alkoxy and hydroxy free radical (1). The alkoxy radical then reacts to form an aldehyde and a new free radical (2). This reaction involves C-C bond scission and can occur on either side of the radical so that the aldehyde formed can be a short, volatile compound, or a long, glyceride attached non-volatile compound. The volatile aldehydes are known to play a major role in the oxidized flavor of fats. Abstraction of a hydrogen atom from another molecule can yield an alcohol and a new free radical (R") (3). The free radicals (R. and R") formed from (2) and (3) may participate in propagation of the chain reaction. Interaction of two free radicals can yield non-radical products and terminate the chain reaction, leading to the formation of ketones (4).

Lipid oxidation in meat

Phospholipids are found to be more unsaturated than triglycerides (Lea, 1957). Since they are normally susceptible to oxidation, their presence leads to severe deterioration in flavor of cooked meat products (Younathan and Watts, 1960). The ratio of triglycerides to phospholipids is about 8:1 in pork, and 4:1 in beef (Hornstein et al., 1961). In one study of precooked, vacuum and non-vacuum packaged beef patties (Gokalp et al., 1983), oxidation of the phospholipid fraction was more advanced in the first 3 months than that of the neutral lipid fraction, while after 3 months of frozen storage, oxidation of the neutral lipid fraction (triglyceride) was more rapid than the phospholipid fraction. This observation agrees with the work of El-Gharbawi and Dugan (1965) who found that tissue lipid oxidation occurred in two stages: the phospholipids were oxidized first and the neutral lipids second. The neutral fat fraction developed off-flavors less readily than the phospholipid fraction, leading to the conclusion that phospholipids contributed to poor flavor. Besides, neutral fat may trap volatile decomposition products of polar lipids and thus reduce their effect on off-flavor development in meat (Love and Pearson, 1971).

Wilson et al. (1976) examined meat from several species and evaluated their susceptibility to warmed-over flavor (WOF). After cooking and refrigerated storage, turkey was found to be most susceptible to WOF followed closely by

chicken, pork, beef, and mutton. Red muscles had higher TBA values than white muscles, indicating that red muscles were more susceptible to oxidative deterioration. Yamauchi et al. (1980) reported that in both total lipids and phospholipids, red pork muscle contained more polyunsaturated fatty acids than white pork muscle. . (1979) reported that chicken red muscle had a considerably higher phospholipid content than white muscle.

Warmed-over flavor (WOF) in meat products

The flavor of cooked meat changes rapidly during subsequent storage and is often referred to as "warmed-over flavor" (WOF). The term WOF was first introduced by Tims and Watts (1958), because it becomes most noticeable after rewarming or reheating (Pearson et al., 1983b), it is also called "reheated flavor" or "meat flavor deterioration". WOF may be caused and/or affected by several factors such as the composition of meat lipids, state of the iron ion in meat, role of metal ion chelators, antioxidants, packaging, storage temperature, and rigor state of the meat.

Composition of meat lipids Tims and Watts (1960) showed that phospholipids were responsible for the intense off-flavor induced by heating ground pork. Wilson et al. (1976) found that WOF becomes more evident as the amount of phospholipids increase. This observation suggests that triglycerides play only a minor role in the development of WOF. Igene and Pearson (1979) concluded that phosphatidyl

ethanolamine (PE) is the most important phospholipid which leads to WOF. A greater loss of polyunsaturated fatty acids (particularly arachidonic acid, $C_{20:4}$) due to WOF were found in PE than in phosphatidylcholine (PC). Love and Pearson (1971) indicated that loss of $C_{20:4}$ fatty acid was consistent with its greater propensity to undergo autoxidation, especially when associated with PE. . (1981) found that the decline of PE and PC were much greater in cooking than during frozen storage, and after cooking PE and PC were more stable at -18°C than at 4°C.

Role of heme and nonheme iron Love (1983) reviewed the role of heme and nonheme iron in lipid oxidation of red meats and pointed out that further knowledge of various forms of iron present in muscle is needed to elucidate the role of heme and nonheme iron. Pearson et al. (1983a) reported that there is a far stronger case for the direct involvement of nonheme iron than that of metmyoglobin in WOF. Sato and Hegarty (1971) studied the role of heme and nonheme iron in meats by using a pigment-extracted model system. Heme compounds were found to have little effect on the development of WOF; the reaction was apparently catalyzed by nonheme iron. Furthermore, they indicated that the catalytic effect of ferrous ion (Fe^{+2}) was higher than that of ferric ion (Fe^{+}_{3}). Love and Pearson (1974) further confirmed the observation of Sato and Hegarty (1971), which showed that there was no acceleration in lipid oxidation when 1.0 to 10.0 mg/g of metmyglobin was added to a

water-extracted, pigment-free beef muscle system. However, adding Fe^{+3} at a level of only 1.0 ppm catalyzed lipid oxidation. Igene et al. (1979) observed that the percentage of bound heme iron in fresh meat pigment extract was about 90% of the total iron, while the level of free nonheme iron was less than 10%. Cooking fresh meat pigment extract increased the nonheme iron to a level of 27% and accelerated lipid oxidation. Chen et al. (1984) noted that the heating rate and final temperature influenced the release of nonheme iron from meat pigment extracts. Slow heating increased the amount of nonheme iron more than fast heating and the optimum temperature for release of nonheme iron was about 63-70°C.

At low levels (<100ppm), ascorbic acid has been shown to catalyze development of WOF (Tims and Watts, 1958; Sato and Hegarty, 1971; Love and Pearson, 1974). Kelley and Watts (1957) postulated that a copper-ascorbic acid complex was the causative agent for the prooxidant effect of ascorbic acid. Kanner and Mendel (1977) proved that either Fe^{+3} or Cu^{+2} coupled with ascorbic acid at a low concentration ($<10^{-3}$M) caused a prooxidant effect. At higher levels (>1,000 ppm), ascorbic acid retarded oxidation (Sato and Hegarty, 1971), probably by upsetting the balance between the ferrous and ferric ion, or by acting as an oxygen scavenger.

Role of metal ion chelators and antioxidants Sato and Hegarty (1971) demonstrated that WOF was completely

eliminated by adding nitrite at a level of 220 ppm and was greatly inhibited at a level of 50 ppm. Nitrite is effective because nitric oxide, which is produced by reduction of nitrite, is an efficient free radical acceptor; in addition, nitrite appears to prevent the release of nonheme iron by stabilizing the porphyrin ring and blocking its breakdown (Chen et al., 1984). Fooladi et al. (1979) showed, that nitrite at 156 ppm inhibited WOF development in cooked meat, with a two-fold reduction for beef and chicken and a five-fold reduction for pork. Igene et al. (1979), used a cooked meat system with or without pigments (heme compound), to show that nitrite at 156 ppm decreased TBA numbers from 1.93 to 0.21 (mg malonaldehyde/kg sample). Taste panel results further proved that nitrite is an effective inhibitor of WOF.

Tims and Watts (1958) showed that addition of phosphates (pyro-, tripoly- and hexameta-) at a 0.5% level to cooked meat protected against WOF due to metal sequestering. Sato and Hegarty (1971) verified the effects of phosphates and rated their effectiveness as: tripoly- > hexameta- > pyro-. Matlock et al. (1984a, b) added sodium tripolyphosphate (STPP) at a 0.375% level (with or without NaCl), to precooked pork patties which were put into frozen storage. They observed that STPP would lower the TBA value no matter if the NaCl was added or not. Sofos (1986b) reviewed the use of phosphates in meat products and showed that their effect on oxidative rancidity was to chelate

proxidant metal ions and to increase the pH value which increased their ability to chelate metal ions.

Packaging and storage temperature Oxygen is one of the most reactive participants in lipid oxidation; its presence would be expected to enhance lipid oxidation. Vacuum packaging reduces the presence of oxygen, and is expected to reduce lipid oxidation. Uebersax et al. (1978) evaluated mechanically deboned turkey meat (MDTM) which was mixed under different treatments: a>. control (vacuum packaged), b>. exposed to air, c>. nitrogen gas, and d>. carbon dioxide gas. Treated MDTM was then packaged in pouches under air, or vacuum packaged, at 4°C up to six days, and frozen at -18°C up to six months. TBA numbers of MDTM samples stored in 4°C were not appreciably different between the air exposed samples and the vacuum packaged samples; however, vacuum packaged samples stored at -18°C had the lowest TBA numbers, with gas (N_2 or CO_2) packed the second lowest, and air exposed samples the highest. Jantawat and Dawson (1980) use frozen MDTM (-18°C) and then refrigerated storage for 72 hours at 4°C, and re-frozen to -18°C, they found vacuum and N_2 packaged samples had a significantly higher unsaturated fatty acid ratio and lower TBA numbers than CO_2 packaged samples with corresponding treatments. A significantly higher loss of polyunsaturated fatty acids and an increase in TBA numbers were found in all treatments which were held at 4°C, for 72 hours before freezing. The rate of oxidation was relatively faster in

refrigerated storage than in frozen storage. Miles et al. (1986) also verified that vacuum packaging reduced discoloration scores and lowered TBA numbers of restructured pork for up to 16 days at 4°C storage when compared with a non-vacuum packaged treatment. Matlock et al. (1984a, b) also showed that vacuum packaging was an effective method which controlled rancidity in precooked sausage patties for up to eight weeks of frozen storage. Igene et al. (1981) found that PE and PC were more stable when stored at -18°C than 4°C even after cooking. This implies that the development of WOF may become more serious when products are stored at 4°C than at -18°C.

Effects of rigor state and pH value The pH of muscle is recognized as an important factor which influences the rate and extent of lipid oxidation in meat. Keskinel et al. (1964) reported an inverse relationship between pH and lipid oxidation in raw beef. Owen and Lawrie (1975) observed that a high pH (6.14) in raw pork was associated with inhibition of lipid oxidation. Judge and Aberle (1980) reported that postrigor dark porcine muscles were more susceptible to lipid oxidation than light muscles; however, prerigor light muscles showed higher oxidative susceptibility than did dark muscles, possibly due to the higher pH values in prerigor dark muscles. Tay et al. (1983) reported that postrigor ground pork had a slightly higher free iron content than prerigor ground pork. Their pH values were in agreement with results reported by Judge

and Aberle (1980), who showed that the ultimate pH of prerigor processed ground pork is higher than that of postrigor processed ground pork. Tichivangana and Morrissey (1985) reported an inverse relationship between pH and TBA values in metal-catalyzed prooxidant activities of Fe^{+2}, Cu^{+2}, and Co^{+2} in heated muscle systems; however, heme-catalization was influenced less by pH, and TBA values increased with increasing pH.

Role of sodium chloride (salt) on lipid oxidation

The effect of sodium chloride as a causative agent in the lipid oxidation of meat has been studied by many researchers, but its action is still poorly understood (Gray and Weiss, 1988;). Knowledge of the independent oxidative influence of NaCl, trace metal ions, and heme pigments is necessary to clarify the mechanisms involved (Cross et al., 1987).

Lea (1939) suggested that NaCl is not by itself a pro-oxidant, but promotes the activity of lipoxidase in meat. Chang and Watts (1950) reported that the presence of hemoglobin or muscle extract does not enhance the rancidity that is caused by salt. They also reported that the catalytic effect of salt depends on its concentration and the moisture in the system. Aqueous salt solutions over 15% were antioxidative, but when the contact between lard and NaCl was extensive, a pro-oxidant effect was found. In methyl linoleate and linoleic acid emulsions in aqueous

solution, Mabrouk and Dugan (1960) observed that NaCl had an inhibitory effect, and concluded that it may have resulted from the decreased solubility of oxygen in the emulsion with increased concentration of NaCl. Consequently, the availability of oxygen would be a limiting factor in the reaction. Bolland (1946) stated that at oxygen pressures above 200 mm.Hg, the oxidation rate of ethyl linoleate is independent of oxygen pressure.

Ellis et al. (1968) studied the oxidation of freezer-stored, NaCl-cured pork by measuring carbonyl and peroxide content of the products. They found that increasing the concentration of NaCl accelerated autoxidation, but did not affect hydroperoxide decomposition to monocarbonyl compounds. This was consistent with the results of Chang and Watts (1950) who found that NaCl is a pro-oxidant when in direct contact with lard. Rhee et al. (1983a) evaluated raw and cooked ground pork, stored at 4°C and -20°C, prepared with NaCl, KCl, and $MgCl_2$ at ionic strengths (IOS) of 0.70 or 0.35 (for NaCl they are 2.5 or 1.25%) to study development of rancidity. In most cases, increased IOS increased rancidity. At both 4°C and -20°C, NaCl and $MgCl_2$ increased the rancidity of raw and cooked samples, whereas, KCl increased the rancidity of raw samples only. In cooked samples, $MgCl_2$ increased rancidity more than NaCl when stored at 4°C, but the opposite was true for samples at -20°C. Rhee et al. (1983b) reported that in raw ground beef, held at 4°C and -20°C, TBA values increased

with increasing NaCl up to 2%; TBA values decreased as NaCl increased form 2 to 3%. Matlock et al. (1984a,b) used raw and precooked frozen pork patties treated with 0.0, 0.5, 1.0, and 1.5% NaCl and studied their oxidative stability. Precooked samples with the higher NaCl levels had higher TBARS values, while raw pork samples were not affected ($P>0.05$) by NaCl level. Chen et al. (1984) also showed that 2% NaCl increased TBARS values of precooked meat stored at 4°C. Dawson et al. (1978) reported that N_2 and vacuum packaged, mechanically deboned turkey containing 1.5% NaCl and stored at 4°C, showed a linear pro-oxidant effect, all samples had relatively low TBARS values which were believed to be the result of mixing under N_2 and vacuum packaging.

Greene and Price (1975) used precooked fresh ground pork containing up to 2.0% NaCl with/without 0.02 $NaNO_2$ to study the meat flavor and lipid oxidation. They found that NaCl, in both cured and non-cured systems, appeared to be the major factor responsible for cured meat flavor rather than sodium nitrite or an absence of lipid oxidation. Moreover, NaCl levels up to 1.5% decreased lipid oxidation, but NaCl levels to 2.0% increased lipid oxidation in both systems. Ang and Hamm (1986) also reported that NaCl and sodium tripolyphosphate (STPP) had an antioxidative effect in refrigerated, precooked broiler breast meat. They reported that a combination of NaCl and STPP most effectively reduced TBARS values during storage followed by STPP, NaCl and lastly H_2O (the control) alone. Ang and

Young (1989) comprehensively studied the effects of pH, STPP, and ionic strength (IOS, adjusted with NaCl) on the oxidative stability of broiler breast patties. They found that adding 2.1% NaCl (increasing the IOS by 0.6) raised TBARS values in raw meat and during cooking, but it had a reverse effect on cooked-stored patties, which was probably because the IOS caused a lower increase in nonheme iron content in the cooked patties. They also found that when IOC (NaCl) was increased up to 0.6, nonheme iron content in the raw sample did not drop significantly, but did in the cooked sample.

Packaging materials for in-package pasteurized meat products

Harte (1985) outlined the requirements of barrier used for cook-in meat products. The barries should be able to maintain a product's flavor, color, moisture content, and prevent microbial spoilage. Besides, the barrier should also be physically tough, formable, transparent, heat sealable, compatible with product and temperature resistant. Due to the fact that properties required for such purposes are seldom found in a single component, multilayered materials composed of various coextruded or laminated polymers have been developed. Some typical oxygen barriers used for in-package pasteurized products are: nylon/EVA (outside structure layer/inside sealing layer), nylon/Surlyn®, nylon/PVDC/Surlyn, PET/LLDPE, PET/PVDC/LDPE, LDPE/EVOH/EVA, and nylon/EVOH/Surlyn (Siegel, 1982;

Terlizzi, 1984; Harte, 1985.; ; Simunovic et al., 1985; Jones et al., 1987; Stites et al., 1989). Among various polymer used, PE, EVA, and Surlyn are used as sealants and moisture barriers, while nylon, PET, PVDC, and EVOH are used as barriers and/or to provide mechanical strength. Generally, a good oxygen barriers are also a good CO_2 barriers with its CO_2 transmission rate 2 to 4 times greater than its O_2 transmission rate.

Moisture and temperature may influence the performance of barrier materials. EVOH, an excellent O_2 barrier under dry conditions (<1 $ml/m^2/24$ hr/atm) is heat stable but inherently moisture sensitive, and becomes increasingly permeable as it absorbs moisture. Therefore, for in-package pasteurization, deposition of desiccant in a layer of the multilayer structure results in much smaller oxygen permeation (Rice, 1987). PVDC, although not as good an O_2 barrier (about 10-30 $ml/m^2/24$ hr/atm) as EVOH, is a satisfactory barrier for oxygen, water, fat, and aromas. Its disadvantages include limited heat stability, brittleness and difficulty in processing (Sacharow, 1988). Nylons are also sensitive to moisture and temperature changes and are only intermediate O_2 barriers. To choose between these barriers is a matter of compromise and depends on the shelf-life required, performance required, and economic considerations.

Recently, Toppan Printing Co. (Japan) and Toyo Ink Co (Japan) test marketed a silica-coated PET film laminated to

cast polypropylene film in a heat sealable, retortable grade. This barrier is claimed to have 0.5 oxygen permeation rate (OTR, ml/m^2/24 hr/atm) at 25°C, 100% R.H. and can retain more than 75% of its OTR after boiling or retort treatment (Anon, 1988a, b; Watanabe, 1988; Brody, 1988). This silica barrier is a microwavable and transparent retortable barrier with only limited gas barrier changes both at retort temperature and humidity. A potential weakness of this super barrier may be the brittle nature of the silica, but there are no available data which tell how much this will affect the OTR of the material.

Role of packaging barriers on the lipid oxidation of vacuum packaging/modified atmosphere packaging of precooked refrigerated foods (VPPRF) Low residual oxygen in the headspace of VPPRF is the most important factor influencing lipid oxidation in VPPRF products. A high residual O_2 in the headspace of products may also create discoloration, loss of flavor, and encourage microbial spoilage (aerobic). Residual oxygen needs to be below 1% and CO_2 above 20% in order to best maintain product quality in precooked chilled foods (Coulon and Louis, 1989). A high oxygen barrier is critical to maintain product quality (Ahvenainen et al., 1989,1990a; Yen et al. 1988; Terlizzi et al., 1984; Amundson et al. 1982; Lin and Sebranek, 1979). Data relative to the minimum oxygen transmission rate (OTR) required for cook-in meat products are available for cured meats but not for non-cured meats. Terlizzi et al. (1984) concluded that for

2-3 lb consumer style cook-in hams, OTR should be lower than 15 $ml/m^2/24$ hr/atm which is the same level as Rizvi (1981) recommended for vacuum packaged processed (cured) meat. Lin and Sebranek (1979) studied the color stability and rancidity of sliced bologna during 35 days of storage and summarized that the minimum OTR required to prevent lipid oxidation is 60 $ml/m^2/24$ hr/atm (23°C, 0% R.H.) with a vacuum level > 25 in. Hg. For color maintainence, an OTR of < 7 $ml/m^2/24$ hr/atm with a vacuum level of >29.6 in.Hg was needed.

Vacuum packaging/modified atmosphere packaging of precooked refrigerated foods (VPPRF)

Vacuum packaging/modified atmosphere packaging

Brody (1989) defined vacuum packaging (VP) as the removal of all air within the package without replacement with another gas. Modified atmosphere packaging (MAP) is the initial alteration of the gaseous environment in the immediate vicinity of the product, permitting the packaged product interactions to naturally vary their immediate gaseous environment. Vacuum packaging is often regarded as MAP in the sense that elevated levels (10-20%) of CO_2 are produced within vacuum packages by microorganisms as they consume residual oxygen or by respiring products (Silliker and Wolfe, 1980). MAP technology and microbiology have been reviewed by Genigeorgis (1985), Hintlian and Hotchkiss (1986), and Hotchkiss (1988).

Safety concerns and regulations Ironically, the same atmospheric modification that delivers the benefits of MAP is also the main cause of controversy surrounding the potential health hazards that accompany such atmospheric modification (Palumbo, 1986). Atmospheric modification, particular O_2 reduction in the package headspace will, favor the growth of anaerobic bacteria (i.e. *C. botulinum*). Growth of aerobic bacteria that normally spoil the product will be inhibited. Consequently, it is possible to have the right conditions for optimum growth of anaerobic toxin producers which, in the absence of competing aerobic organisms, will cause no organoleptic degradation. Post et al. (1985) conducted studies on fish fillets inoculated with *C. botulinum* and packaged using MAP. They concluded that organoleptic spoilage would not always cause the product to be rejected before the fillets became toxic. Recently, "sous vide" products which were first introduced by Floury Micron (Paris, France) and others have created significant controversy with regards to food safety. By pasteurizing after vacuum packaging, the shelf life is extended by destroying the heat sensitive spoilage bacteria, though the heat treatment does not provide a guarantee against food pathogens (Randall, 1988; Chomon, 1988; Lioutas, 1988).

These so called "new generation refrigerated foods," including vacuum packaged, pasteurized, refrigerated foods (VPPRF) and sous vide, are among the current focus of the recently established National Advisory Committee on

Microbiological Criteria for Foods (NACMCF), which is sponsored jointly by Federal agencies including FDA and USDA. This committee is currently using the Hazard Analysis and Critical Control Point System (HACCP) to address the product safety needs for refrigerated foods (Corlett, 1989). The HACCP system has been recommended by the National Food Processors Association (NFPA) as part of a total program to assure the microbiological safety of refrigerated foods (NFPA, 1988a,b) and is under active consideration and development by FDA and USDA. To integrate HACCP into the design of food products requires: 1. identification of risk factors; 2. formulation of the product to reduce risks by employing back-up microbiological barriers such as low pH, moisture control, competitive microbial flora, preservative, thermal processing, and partial reliance on modified atmosphere; 3. selection of appropriate packaging to produce and distribute safe products; and 4. education of consumers (Corlett, 1989). The technology currently used for extension of the shelf-life of fresh prepared food in the U.S. are: 1. temperature control, 2. oxygen lowering, and 3. CO_2 elevation. This gives 7-60 days longer shelf-life depending on the product and technology used (Labuza and Breene, 1989).

In-package pasteurization and "Sous Vide" technology

In-package pasteurization has been reported to extend product shelf-life by eliminating recontamination, oxidation, and increasing water retention (Siegel, 1982;

Simunovic et al., 1985; Jones et al., 1987; Stites et al., 1989). Other significant advantages of this technology are potential elimination of shrinkage and purge (Siegel, 1982). In-package pasteurization has been used with pork (Delaquis et al., 1986; Jones et al., 1987; Prabhu et al., 1988), beef (Simunovic et al., 1985; Stites et al., 1989; Mann et al., 1989), poultry (Sofos, 1986a; Smith and Alvarez, 1988), and fish (Eklund et al., 1988) to increase shelf-life. "Sous Vide," a system approach to vacuum cooking, is not just boil-in-the bag, but uses careful heat processing for an appropriate time to achieve balance between organoleptic quality and pasteurization level, and is then chilled quickly and stored chilled (Randall, 1988). Generally, these products have a claimed shelf life of 2-3 weeks when stored at 2-4°C (Lechowich, 1988). Magnetic (Labuza and Breene, 1989), microwave, and gamma ray irradiation (Barbut et al., 1987; Kreiger et al., 1983) pasteurization were also investigated.

Role of VP or MAP on microbial shelf life of precooked, refrigerated meat To achieve success with MAP in precooked foods, four elements are needed. 1. suitable gas mixture, 2. suitable packaging equipment, 3. clean and appropriate handling, and 4. protective films must interact in a balanced system (Coulon and Louis, 1989). Generally, only CO_2 and N_2 are used for precooked foods, O_2 is usually avoided to reduce oxidative rancidity. Lioutas (1988) reported that 20% CO_2 and 80% N_2 and storage at 0-2°C is

recommended for a non-respiring, chilled, cooked meal. For this category of product where gas exchange with the outside atmosphere is not desirable, high-barrier flexible packaging is usually used.

CO_2 is commonly used because of its bacteriostatic (especially gram negative) and fungistatic properties. However, CO_2 does not retard the growth of all types of microbes. Lactic acid bacteria's growth is actually improved in the presence of CO_2 under low O_2 tension. CO_2 also has little effect on yeast (Clark and Lentz, 1969; Silliker and Wolfe, 1980; Enfors and Molin, 1984; Brody, 1989a; Coulon and Louis, 1989). The inhibitory effect of CO_2 has been attributed to alteration of the cell's permeability, pH, and enzymatic inhibition (Young et al., 1988). In general, an increase of the CO_2 level to 25% results in a 2X increase in microbial generation time at 10°C, a 2.5X increase at 4.4°C, and a 3.5X increase at 0°C (Labuza and Breene, 1989). Normally, a minimum level of 20% CO_2 is required, although 5% has been reported to suppress microbial growth (King and Nagel, 1967; Coulon and Louis, 1989). However, CO_2 above 25% will result in a gray brown color in raw, red meat. For precooked products, the ratio of N_2 and CO_2 depends mainly on one key factor: the equilibrium relative humidity of the product. This factor determines the rate of microbial growth, oxidation and enzyme activity (Coulon and Louis, 1989). For products with higher water activity (A_w), the residual oxygen should be

less than 1%. Vacuum packaging and MAP with 20% CO_2/80% N_2 have been reported to have the same effectiveness in extending the shelf life of some refrigerated, precooked meat products (Silla and Simonsen, 1985; Ahvenainen et al., 1989, 1990a; Hwang et al., 1990) and some raw pork and beef (Seiderman et al. 1979a, b).

In-package pasteurization has proven to be very useful as a means of reducing microorganisms in VPPRF. Simunovic et al. (1985), evaluated restructured beef which was cooked to 82.2°C (internal T) for 15 min. (3D of lethality for *C. bot.*) and showed no aerobic or anaerobic bacterial growth during 16 weeks of storage at 4.4°C. Smith and Alvarez (1988) reported that in commercially prepared (cooked to internal T=71°C) turkey roll containing 1.5% salt and 0.5% phosphate, no psychotropic aerobic colony forming units (CFU) were detected during 87 days storage at 4°C. However, 188 and 100 Most Probable Number (MPN)/g of mesophilic anaerobic CFU (which may include *Clostridia*) were found on the core and in the surface, respectively. Stites et al. (1989) reported that in-package pasteurization of roast beef to an internal temperature of 70°C resulted in low total aerobic counts and subsequently slow microbial growth (<1000/g) after 28 days at 4°C. Prabhu et al. (1988) used a second in-the-bag cooking to 66°C after precooked pork chops were vacuum packaged. They found that without the secondary in-package cooking, pork chops spoiled after 15 days at

2-4°C, but those samples receiving a second cooking had more than 60 days of refrigerated shelf life.

Lipid oxidation of precooked, refrigerated meat under VP/MAP and subsequent storage Only a few studies have been done using refrigerated, precooked meat products which were susceptible to warmed-over flavor. Nolan et al. (1989), studied the effect of air, vacuum, N_2, and CO_2 packaging on lipid oxidation, and color in precooked pork and turkey packaged in a high oxygen barrier bag at 4°C for 18 hours or -20°C for 3 months. They concluded that for refrigerated and frozen storage, N_2 and CO_2 samples were less "oxidized" (sensory and TBARS values) than those stored in air but were more "oxidized" than vacuum packaged meat. Vacuum packaging was particularly effective for turkey, whereas CO_2 and N_2 offered only minimal improvement over packaging in air. Hwang et al. (1990) evaluated cooked beef loin slices packaged in vacuum, 80% N_2 and 20% CO_2, and air, using an EVOH oxygen barrier material and stored at -20_oC for 11 weeks to study their effect on lipid oxidation and color change as measured by TBARS values, hexanal content, sensory analysis, and Hunter Spectra color values. The results showed that vacuum packaging and CO_2/N_2 packages were more meaty, had less warmed-over flavor, less oxidation and had lower TBARS values and lower hexanal contents than those packed in air. Vacuum packaged beef samples were redder than those packaged in CO_2/N_2 and air and frozen storage which is consistent with Nolan et al. (1989), who

stated that vacuum packaging led to increased redness compared with meat packaged with 100% N_2, CO_2 and air. For precooked meat in refrigerated storage, Carr and Marchello (1987) showed that vacuum packaging also maintained a superior cooked beef color for 18 days and was better than samples containing 15% CO_2 with their headspace O_2 increase from 10 to 40% O_2. However, Smith and Alvarez (1988) reported that visual observation indicated the color of precooked turkey rolls at 4°C varied from white to pink initially, and started to fade to a grayish pink after 47 days of storage at 4°C Polyphosphate, salt-treated, vacuum packaged, and in-package pasteurization provided the best protection against microbial growth as well as lipid oxidation. Jones et al. (1987) reported that TBARS, oxidative rancidity and WOF of pork roasts did not change ($P<0.05$) during refrigerated storage which resulted in a shelf life in excess of 28 days. Similar results were reported by Simunovic et al. (1985) for 16 weeks with restructured beef; by Smith and Alvarez (1988) for 87 days with turkey rolls; and by Stites et al. (1989) for 28 days with beef roasts. The microbiological and organoleptic shelf life of these products depends on their: 1. pasteurization temperature and duration, 2. initial microbial load, 3. antioxidants used, 4. oxygen barrier used, 5. nature of the product itself, and 6. storage condition, i.e., temperature, light, and moisture.

Measurement of lipid oxidation

2-Thiobarbituric acid (TBA) test The reaction of malonaldehyde (MA) with 2-thiobarbituric acid (TBA) has been widely used to test for oxidative rancidity in foods. In fact, the TBA test has become the most widely used method for assessing the extent of oxidative deterioration in muscle foods (Tarladgis et al., 1960; Gray, 1978; Melton, 1983; Igene et al., 1985). Gray (1978) in his review paper on the measurement of lipid oxidation stated:

> "There is no ideal chemical method which correlates well with change in organoleptic properties of oxidized lipid throughout the entire course of autoxidation. The methods discussed each gives information about particular stages of the autoxidative process."

Pearson et al. (1983a) concluded that any chemical method used to measure WOF must show correlation between the measurement and sensory scores. They also felt that TBA values obtained through the distillation procedure of Tarladgis et al. (1960) were indicative of WOF, even though the method sometimes is not as precise as desired.

Melton (1983) reviewed the methods used to measure oxidation in muscle foods and stated that if the distillation method is to be used to assess lipid oxidation in muscle foods, it would be wise to modify the method by adding some antioxidants. Moerck and Ball (1974) modified the original method by adding an antioxidant mixture containing 20% BHA, 6% PG, and 4% citric acid in propylene glycol (Tenox II) at the distillation stage to prevent oxidation of chicken meat. Ke et al. (1977) added 100 mg

(per 10g meat) of each of PG and EDTA at the distillation stage to prevent further oxidation of mackerel samples. Rhee (1978) pointed out that the use of BHA during the distillation of chicken and fish may increase their TBA numbers by increasing the decomposition of lipid peroxides in muscle fat, yet addition of PG and EDTA may lower the TBA numbers. Pikul et al. (1983) added BHT during the heating step of the TBA assay and found that this reduced TBA numbers in a chicken meat system. Crackel et al. (1988) reported that replacing acetic acid with water in the TBA solution and adding antioxidant significantly reduced TBARS values of fish and chicken, but had no influence on beef or cooked chicken meat. Generally, TBA values as determined by the distillation method and absorbance of the malonaldehyde-TBA complex at 532 nm have been reported to be highly related to sensory scores of oxidized and warmed-over flavor in foods (Watts, 1962; Zipser et al., 1962; Igene et al., 1979; Igene and Pearson, 1979).

Although the TBA test provides a good indication of lipid oxidation, some problems exist with the reproducibility and specificity of the method. It is a misconception to assume that malonaldehyde is the only lipid peroxidation product capable of producing the TBA reaction. Some lipid oxidation products such as 2,4-alkdienals and 2-alkenals may also produce a positive TBA reaction (Marcuse and Johansson, 1973). Formaldehyde, which was found to increase during frozen storage in vacuum packaged minced

fish, is able to interfere with the TBA reaction by raising the turbidity of the TBA solution (Careche and Tejada, 1988). Malonaldehyde is able to react with protein and amino acids in muscle foods (Melton, 1983; Gokalp et al. 1983).

Hexanal analysis Hexanal is a major secondary product of linoleate oxidation (Frankel et al., 1981) and is formed only when $C_{18:2}$ is present in an oxidizing meat system. Mottram et al. (1982) reported that pork has substantially higher levels of linoleate than beef, and, therefore, a larger amount of hexanal will be in the pork-based sample. Early studies have shown hexanal to be a useful marker of lipid oxidation in precooked meat in extended storage (St. Angelo et al., 1987; Dupuy et al., 1987; Shahidi et al.,1987; Lai, 1989; Hwang et al., 1990; Ang and Lyon, 1990). Shahidi et al. (1987) reported that hexanal measurement is a very sensitive technique and more consistently measures lipid oxidation than the TBA test, especially in the early stages of lipid oxidation. Hwang et al. (1990) reported correlation coefficients of 0.89 and 0.89, between warmed-over flavor and hexanal content, and warmed-over flavor and TBARS values, respectively, for vacuum packaged cooked beef slices after 11 weeks of frozen storage. Similar results were reported in cooked ground pork refrigerated in a plastic bag (r=0.98, by Shahidi et al., 1987), and in refrigerated stored beef roasts (r=0.8, by St. Angelo et al., 1987).

Several methods have been developed to detect lipid oxidation products in meat systems. Direct headspace analysis has been used to detect aldehydes and ketones in heated pork fat (Yasuhara and Shibamoto, 1989). The major problem with this method is its limited sensitivity, hence only major peaks are identified. Use of a porous trap such as Tenax GC to concentrate the volatiles prior to separation by gas chromatography (GC) enables a more sensitive measurement. Normally, the sample is heated in a closed system, then purged with a carrier gas or extracted in vacuum (Vercellotti et al., 1987) so that the volatiles are trapped in the Tenex glass liner trap. This trap is later discharged with solvent (Shahidi et al., 1987; Stoick, 1989) or is secured in an external closed inlet device (ECID, Scientific Instruments, River Ridge, LA)(Dupuy et al., 1987) where the volatiles are purged by heating. Direct GC analysis is another method, where the meat sample is packed in a glass liner which is then placed into the injection port of a gas chromatography for purging and separation. This method gave satisfactory results (Dupuy et al., 1987).

MATERIALS AND METHODS

Materials and Manufacturing Procedures

Boneless Boston shoulders and boneless picnic shoulders were obtained from the MSU Meat Laboratory within 48 hours of slaughter. The raw materials were first ground through a 9.5 mm (3/8 inch) plate, formulated to contain about 15% fat, and then ground again through a 3.2 mm (1/8 inch) plate. Ground pork was divided into two portions and each portion was blended (Hobart blender, Troy, OH) for 5 minutes with/without 1.5% NaCl (AKZO Salt Inc., Clarks Summit, PA). The ground pork was then stuffed into a 2½ fibrous casing (Teepak, maximum diameter = 72 mm) and immediately frozen at -18°C overnight before slicing into 1.7 cm thick patties (65 grams each). Each pouch (15.2 cm x 21.6 cm) was designed to contain two patties and the filled pouch was then vacuum packaged (while the contained patties are still solid hard) at 614 mmHg vacuum level (calibrated by putting a vacuum gauge in a pouch prior to vacuum packaging) by using a Multivac vacuum packaging machine (Sepp Haggenmuller, W. Germany). Three oxygen barrier materials were used as the packaging material: 1. nylon/PE (Koch, Kansas City, MO, thickness= 0.09 mm), 2. nylon/saran/Surlyn, (Culon Grade 850, Curwood Inc., New London, WI, thickness= 0.08 mm, manufacture reported OTR: <15.5 ml/day.M^2.atm at 23°C, 0% RH), and 3. PET/SiO_2-PET*/CPP (Toppan Printing Co., Tokyo,

Japan, thickness: PET= 0.012 mm; SiO_2-PET= ceramic coated PET film (Toppan GL film), 0.012 mm; CPP=Cast Polypropylene, 0.060 mm; actual total laminated thickness is about 0.10 mm; OTR= 0.5 ml/day.M^2.atm at 25°C, 75% RH). Vacuum packaged samples were then placed on racks and cooked in a smokehouse equipped with a microprocessor (PC 5809, Butcher & Packer Supply Co.). Pork patties were steam cooked (pasteurized) from an internal temperature of 0°C to 82±2°C (it takes about 20 minutes to reach) and stay at that temperature for 20 minutes. Two sample packages were individually monitored for their internal temperature by inserting a thermocouple to each patty and connected the thermocouple to a recording potentiometer (Model F2157, Honeywell Co. Minneapolis, MN). After cooking was completed, the pork patties were chilled in a cold water shower until an internal temperature of 23°C was reached. Residual water on the sample packages was wiped off with dry towels and the samples were stored in a 3±1°C walk-in cooler for up to 120 days.

Treatment Descriptions

There are total four different sets of treatments were used in this study. In the first set of treatments, the purpose is to determine the effects of in-package pasteurization and vacuum packaging on the oxygen transmission rates of the three selected oxygen barriers. Each barrier received the following treatments:

1. Control (No vacuum, No pasteurization)
2. Vacuum packaging (to 635 mmHg of vacuum level)
3. Pasteurization (cooked at 82°C water for 30 min)
4. Vacuum packaging then pasteurization

Each treatment was replicated four times. In the vacuum packaging treatment, two frozen (solid hard) patties were put side by side into the barrier pouch and vacuum packaged.

The second set of treatments was designed to study the effect of oxygen barrier and salt on product lipid oxidation and on the headspace gas concentration (CO_2%, O_2%) during storage. The sample patties were vacuum packaged and cooked under the following variables:

1. Nylon/PE barrier
2. Nylon/PE barrier + salt (1.5%)
3. Nylon/saran/Surlyn barrier
4. Nylon/saran/Surlyn barrier + salt (1.5%)
5. PET/SiO_2-PET/CPP barrier
6. PET/SiO_2-PET/CPP barrier + salt (1.5%)

These samples were stored for 1, 15, 30, 60, 90, and 120 days and then analyzed. Each variable at each sampling day had 7 replications. For TBA measurement, at each sampling day, each sample package was opened, take one patty for analysis, the other patty in the same package was stored (open package) for 24 hours, then analyzed. 2 observations for each sample patty were made. For hexanal analysis, 1 observation for each sample was made. For headspace CO_2/O_2 analysis, 3 observations for each sample were made.

The third set of treatments was designed to monitor the microbial growth of the samples which packaged in 3 different oxygen barriers. The sample patties were packaged in 1. nylon/PE pouch, 2. nylon/saran/Surlyn, 3. PET/SiO_2-PET/CPP barriers then cooked to 82°C for 20 minutes. These samples were stored for 1, 15, 30, 60, 90, and 120 days at 3°C then analyzed. This set of treatments were repeated four times and each treatment has 2 samples at each sampling day.

The fourth set of treatments was designed to study the effect of in-package pasteurization and storage on tensile strength, seal strength, and seal integrity of the three oxygen barrier pouch systems. Samples were vacuum packaged, in-package pasteurized and refrigerated for 7 days. Each barrier received the following treatments:

1. Control (No pasteurization, No storage)
2. Pasteurization (cooked at 82°C for 30 min)
3. Pasteurization and subsequent refrigeration

Both machine direction and cross direction tensile strength and seal strength of the barriers were examined. Each treatment was replicated 10 times.

Analytical Methods

Oxygen transmission

Oxygen transmission of the barriers was determined as described in ASTM Standard D1434. To test the effect of cooking (pasteurization) on oxygen transmission of the barriers, each of 4 nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP barrier pouches were impulse (0.8 second) sealed on the Multivac vacuum packaging machine before and after cooking in an 82°C water bath for 30 min. To test the effect of vacuum draw on the oxygen transmission of the materials, each of 4 nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP pouches containing two frozen (solid hard) pork patties was vacuumed and impulse (0.8 second) sealed using the Multivac vacuum packaging machine under 635 mmHg of vacuum. Material from these barrier pouches was then tested before and after cooking in an 82°C water bath for 30 min. The oxygen transmission rate (ml/day.M^2.atm) of the three different barriers was determined using an OX-TRAN 100 (Modern Controls, Inc., Minneapolis, MN) oxygen transmission tester equipped with a 100 cm^2 film sample cell. Wet bubbling was used to generate a near 75% RH (15.8g H_2O/m^3, at 23°C) environment in both the oxygen and carrier (nitrogen/hydrogen) gas. The carrier gas flow rate was set at 20 ml/min, while a standard reference (Material #1470, polyester plastic film for oxygen gas transmission, U.S.

Department of Commerce, National Bureau of Standard, Washington, D.C.) with a transmission rate of 63.08 ml/day.M^2.atm was used to calibrate the OX-TRAN tester. A flat-bed recorder (Model L6512, Linseis Inc., Princeton, NJ) with a chart speed of 2.54 cm/hr was used to record the oxygen transmission data.

Measurement of lipid oxidation

Thiobarbituric acid test (TBA test) The TBA distillation method of Tarladgis et al. (1964) as modified by Rhee (1978) was used to measure the development of oxidative rancidity in the sample patties. Twenty five grams of meat, 80 ml distilled water and 15 ml of antioxidant solution (1 g propyl gallate + 1 g of EDTA in 40 ml alcohol and H_2O up to 200 ml) were added in a beaker prior to sample homogenization (Tekmar homogenizer, Tekmar Company, Cincinnati, OH) into a slurry. Forty eight grams of the slurry (containing 10 g meat sample) was weighed and transferred into a Kjeldahl flask with an additional 49.5 ml distilled water. Two ml of HCl (4N) solution was added to adjust the pH to 1.5. 0.5 ml of antifoam solution plus 2-3 boiling chips were added to the slurry. This mixture was then distilled until 50 ml distillate were collected. Triplicate analyses were made using 5 ml aliquots of the distillate. Each sample distillate was put into a test tube along with 5 ml of TBA solution (0.02M TBA in distilled water). These were heated for 35 minutes in a boiling water

bath (100°C) and then cooled for 10 minutes in tap water. The optical density of the solution was read against a blank TBA solution at 532 nm using a Bausch and Lomb Spectronic 2000. A conversion factor of 6.2 (Crackel et al., 1988) was used to calculate TBA values as mg malonaldehyde/kg meat.

Hexanal analysis by purge-and-trap/solvent extraction procedure The pretrapping procedure of Dupuy et al. (1987) was modified due to the instability and lack of reproducibility in the preliminary test, especially the sample patties did not contain any polyphosphate (as antioxidant). Therefore, this procedure was modified -- by adding antioxidants (propyl gallate/EDTA) into the sample solution and flushing with nitrogen during sampling. Glass traps were packed with 200 mg of Tenax TA (60/80 mesh), and secured with glass wool at both ends of a glass liner (84*9 mm). Glass traps were conditioned in a stream of nitrogen (20 ml/min) at 200°C for 24 hrs. Pork patties were cut into cubes (about 0.3 cm in each dimension) and 10 g samples were weighed into a 500 ml flat bottom flask to which 4 ml (16.6 μg) of aqueous 2-heptanone internal standard solution (5μl 2-heptanone in 10 ml methanol and adding it up to 1000 ml water solution), 1.5 ml of propyl gallate solution (2.5 g/50 ml MtOH), 1.5 ml of EDTA solution (2.5 g/50 ml H_2O) and 26 ml of distilled water were added. The flask was fitted on to a 24/40 condenser topped with a rubber stopper (Fig. 1). On the stopper, two short glass tubes were fitted to the

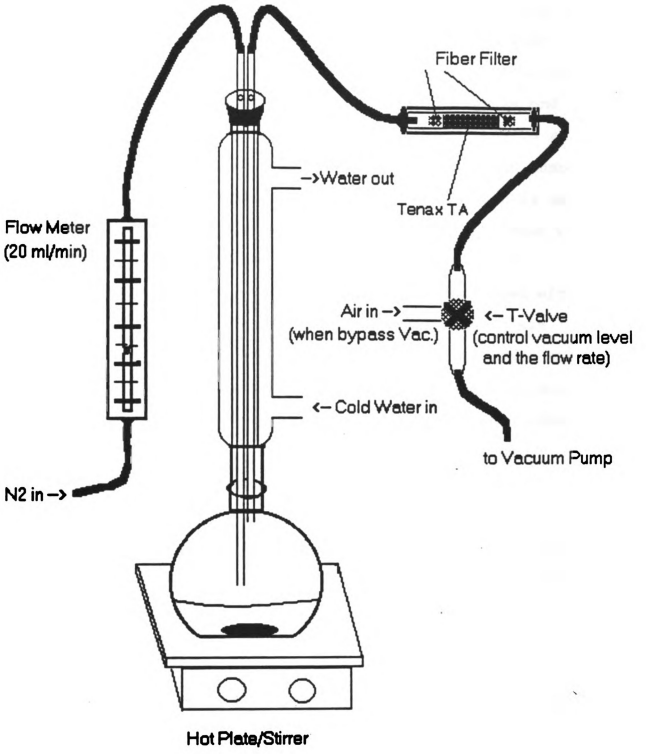


Figure 1. Detail of the hexanal analysis apparatus.

inside of the stopper. One end of the first glass tube was connected to a TYGON tube inside the condenser and down to the mouth of the flask, while the other end was connected to a nitrogen source equipped with a flowmeter. The second glass tube was connected to the Tenax trap and then to a vacuum pump. A T-shaped valve was installed between pump and trap in order to adjust the vacuum level. Samples were held at 60±3°C for 1.5 hr at a vacuum level of 254 mmHg with constant stirring on a hot plate/stirrer. After the pre-trapping procedure, the Tenax trap was discharged with 1.5 ml of 2-methylbutane and allowed to stand for 2 min. before centrifuging at 400 rpm for 15 seconds to elude the volatiles. This step was repeated three times and produced about 4.5 ml of extract. The extract was evaporated under nitrogen to 1.5 ml and a 2 µl sample was immediately injected onto the GC column using a pre-chilled 10 µl syringe. A HP 3392A integrator and an HP 5890 GC (Hewlett-Packard Co., Avondale, PA) equipped with a Flame Ionization Detector (FID) and a 60m * 0.25mm ID, Carbowax 10 fused silica capillary column (Supelco Inc., Bellefonte, PA) were used. The oven temperature was initially set at 40°C for 10 minutes, then increased at a rate of 2 °C/min to 80 °C, and then increased at a rate of 30 °C/min to 200 °C for 20 minutes. Injector and detector temperature were 200 and 275 °C, respectively. The flow rate of the helium carrier gas was 0.5 ml/min.

Retention times of samples and the standard hexanal were compared to identify hexanal. A standard solution composed of 4 ml (16.6 μg) hexanal solution (5 μl hexanal in 10 ml MeOH was added to distilled water to make 1000 ml) and 4 ml (16.6 μg) of 2-heptanone internal standard solution (5 μl 2-heptanone in 10 ml MeOH and distilled water to make 1000ml) were freshly prepared and poured into a flat bottom flask. The retention time, peak area of hexanal, and 2-heptanone of the standard solution were determined by GC analysis using the same purge-and trap/solvent extraction procedure. The hexanal/2-heptanone area ratio of this standard solution was then compared against those ratios (hexanal/2-heptanous) from sample solution (meat sample plus internal standard) so that the absolute hexanal contents in meat samples can be calculated.

Headspace Analysis

After each storage interval, sample packages (vacuum packaged) were removed from storage and injected with 5 ml helium gas to mix with the CO_2, O_2 inside the package and to provide an enough volume of gas for further sampling. A rubber patch, which with one side has adhesive, was used to attach to the pouch and functions as an air tight injection port. This air tight injection port prevents/eliminates any O_2 or N_2 from atmosphere getting into or out of the sample package during sampling. To further ensure that no air can get into barrier pouches while sampling, sample barrier

pouches were put into a Ziploc plastic bag (Dow Consumer Products Inc.) and flushed with helium gas and closed. The Ziploc bags were also attached with the rubber patch and served as an injection port. A 5 ml of helium gas was injected into the sample pouch through the injection ports by using a air tight syringe. After 2 minutes, a 0.5 ml gas sample from the headspace of the sample pouch was drawn hermetically by using a glass gas-tight syringe and immediately analyzed using a Carle AGC Gas Chromatograph (model-111, Anaheim, CA) equipped with a thermistor detector (TCD) under the following conditions: carrier gas flow rate= 30 ml/min, column temp.= 50°C; column 1= Molecular Sieve 5A, 42/60 mesh; 6'x" O.D.; column 2= 80 % Porapak N, 20 % Porapak Q, 50/80 mesh, 8'x" O.D. Each sample package was tested three times (3 observations) before the package was opened and the meat used for hexanal analysis. A recorder (model-056-03001; Hitachi Ltd. Tokyo, Japan) was used to record the signal output and the gas composition was determined by comparing the signal output from a stand reference gas composed of 5% CO_2, 21% O_2, 74% N_2 (Linde Division, Union Carbide Co., Danbury, CT).

Total bacterial count

The total aerobic and anaerobic counts for each sample were determined using the APHA Standard Plate Count method (Busta et al., 1984). Each package was opened and 2 grams of the meat sample were weighed into a test tube containing

18 ml of sterile peptone water (0.1%). The sample test tube was mixed by shaking and votexing (in a votexer) for 3 min. Serial dilutions from 10^{-2} to 10^{-3} were prepared and the number of microbes evaluated using Plate Count Agar (Difco Co., Detroit, MI). The conditions used for incubation were: Aerobic: 37°C for 48-72 hours; Anaerobic: 37°C for 48-72 hours in a GasPak anaerobic jar with gas pack system (BBL Microbiology System, Cookeysville, MD). Colony numbers (CFU/g) were reported as the average of duplicated samples and expressed as Log_{10} CFU/g.

Tensile strength and seal strength

Tensile strength Tensile strength was determined as described in ASTM Standard D882-83. Each of 10 nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP pouches was sealed using an Impulse Heat Sealer (Sentinel Packaging Industries, Hyannis, MA). Impulse time was 0.8 second and sealing pressure was 50 psi. These samples were tested before and after cooking in a 82°C water bath for 30 minutes. Cooked samples were tested before and after refrigerated storage at 3°C for 7 days. Sample films were cut into 1" wide strips in both machine direction (MD) and cross direction (CD) using a sample cutter (Thwing-Albert Instrumental Co., PA). Sample strips were tested using an Instron tensile testing instrument (model 4201, Instron Corp., Canton, MA) at a cross-head speed of 50.8 cm/min. Tensile strength was reported in N(Newton)/m^2 (PA), and strain in cm/cm (ratio).

Seal strength The procedure used for evaluating seal strength is essentially the same as used in tensile strength, except sample strips were cut to include the seal portion. Each of 10 nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP pouches was sealed using an Impulse Heat Sealer (Sentinel Packaging Industries, Hyannis, MA). Impulse time was 0.8 second and sealing pressure was 50 psi. These samples were tested before and after cooking in a 82°C water bath for 30 minutes. Only the MD seal strength was determined.

ARO vacuum chamber seal integrity test

Seal integrity of the sample packages was tested as described in ASTM Standard D3078-84. An ARO package tester (Model F-099-1080, ARO Corp., Buffalo, NY) equipped with a vacuum pump was used to determine the seal integrity of the pouches. Each of 6 nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP pouches was sealed (impulse time= 0.8 second, vacuum= 635 mmHg) using the vacuum packaging machine. These samples were tested before and after cooking in an 82°C water bath for 30 minutes. Cooked samples were tested before and after refrigerated storage at 3°C for 7 days. Each package was submerged into the chamber which was filled with water. The vacuum level in the chamber was increased slowly until it reached 711 mmHg (1 atm= 760 mmHg) and then held at this level for 2 minutes. If no bubbles

were observed coming from the package or seal area, and if no water was found inside the sample package, it was recorded as having no leakage.

Statistical analysis

Statistical analyses of sample treatments was performed using the Statistical Analysis System (SAS, 1985) program. Analysis of variance was performed using the general linear model (GLM) procedure. Tukey's multiple comparisons and correlation coefficients for comparing mean TBARS values, mean hexanal contents, mean headspace CO_2%, mean headspace O_2%, mean tensile and seal strength were calculated according to Gill (1978).

RESULTS AND DISCUSSION

Effects of pasteurization and vacuum packaging on the oxygen permeation of the oxygen barriers

The oxygen transmission rates (OTR) of nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP before pasteurization and vacuum packaging were 155, 33, and 0.4 ml/day/m^2/atm (23°C, 75% RH), respectively (Table 1).

Table 1. Effect of in-package cooking (pasteurization) and/or vacuum packaging on the mean oxygen transmission rate (OTR, ml/day.M^2.atm) of nylon, saran, silica barrier pouches.

Treatments	Oxygen Barrier*					
	NYLON		SARAN		SiO_2	
	OTR	Std.	OTR	Std.	OTR	Std.
Control	155.4 b**	13.8	33.3 a	3.5	0.4 d	0.06
Cook	215.1 a	29.3	36.8 a	3.7	0.5 c	0.14
Vacuum	161.3 b	31.5	36.4 a	7	1.8 b	0.52
Cook & Vac	198.3 a	12.7	35.3 a	1.3	2.4 a	0.31

* Each data point is the average of 4 replications, each sample has only 1 observation.

** Means within the column followed by a common letter are not significantly different (P<0.05).

According to manufacturers' data, the OTR (ml/day.M^2.atm) for nylon/PE and, nylon/saran/Surlyn were 140 (at 4°C), 15.5 (at 23°C, 0% RH), respectively. The OTR for PET/SiO_2-PET/CPP is 0.5 (at 25°C, 75% RH) (Brody, 1988). These data are in general agreement with the results reported here. The testing conditions may have caused some minor variation. Pasteurization (cooking) increased the OTR

of all three barriers (Table 1 and Fig. 2), but only changes to nylon and SiO_2 were significant ($P<0.05$). After pasteurization, nylon's OTR increased almost 39% and that of the SiO_2 film by almost 29%. Since the OTR for SiO_2 was very low, the difference caused by pasteurization was actually quite small. Nylon's increase in OTR was expected and is due to the hydrophilic nature of the nylon barrier. Vacuum packaging causes some crevices in the SiO_2 barrier, which can be visually seen on the barrier surface. This increased the OTR of the SiO_2 barrier almost 3.5 times (Fig. 3). Vacuum drawing can create physical cracking of the structure. Wrinkles in the pouch material following vacuum packaging were evident. Vacuum packaging and cooking plus storage for 7 days further increased the OTR of the SiO_2 barrier.

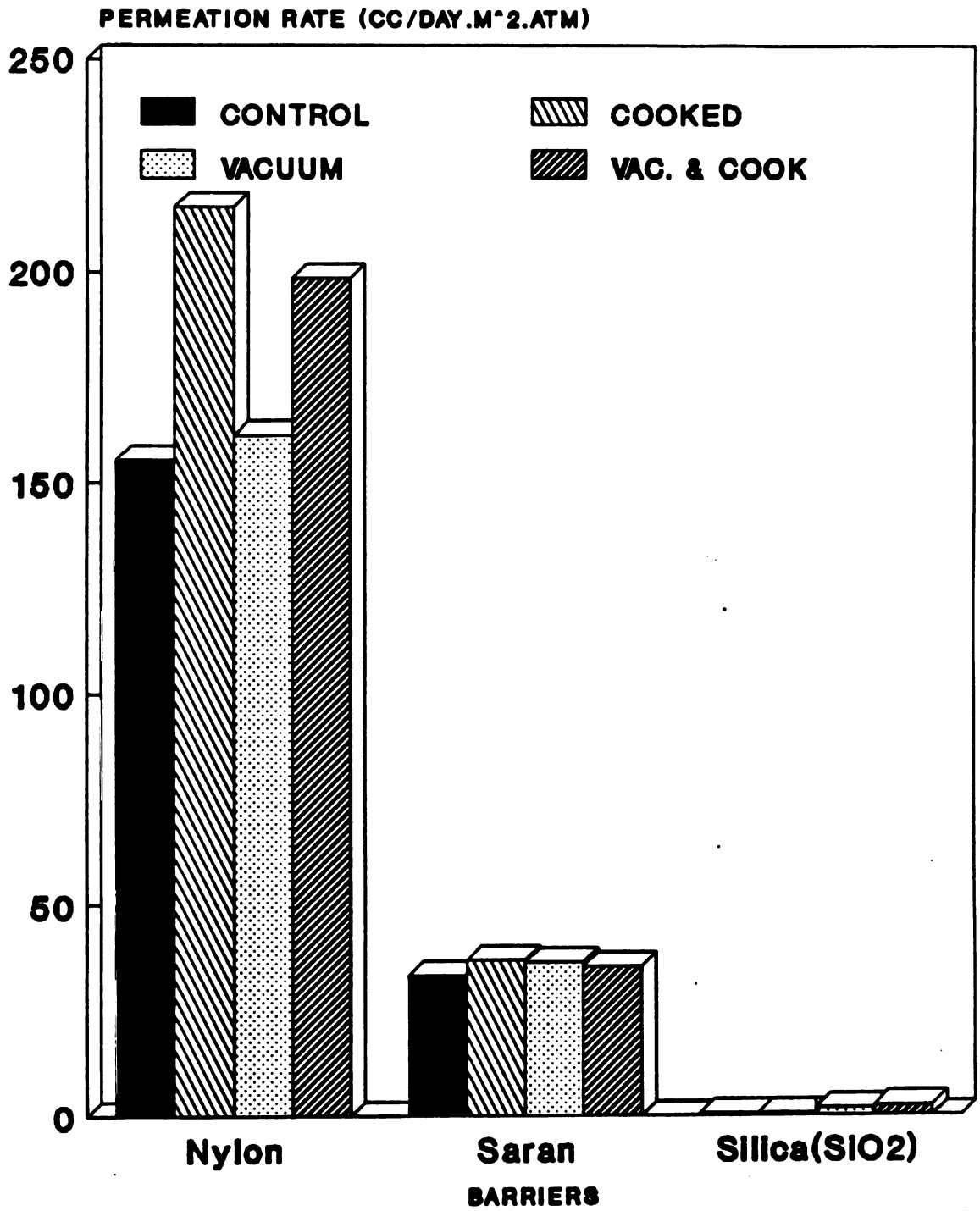


Figure 2. Effect of in-package cooking (pasteurization) and/or vacuum packaging on the mean oxygen transmission rate through nylon, saran, and silica barriers.

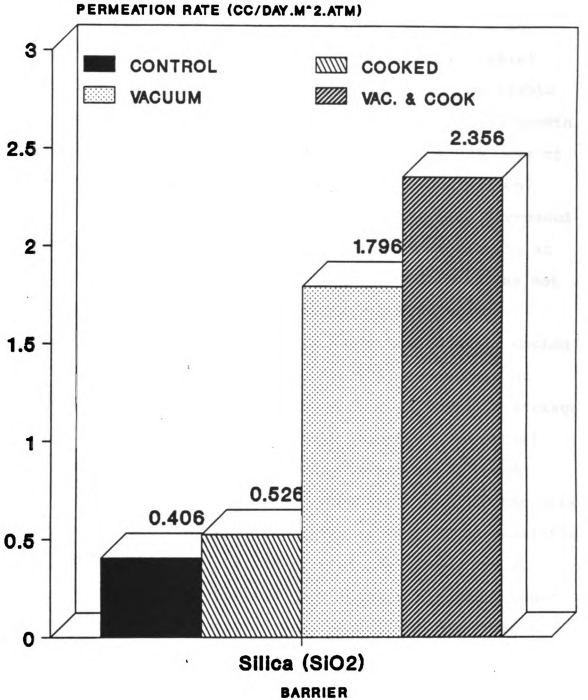


Figure 3. Effect of in-package cooking (pasteurization) and/or vacuum packaging on the mean oxygen transmission rate through the silica barriers.

Microbial growth in the pork patties - aerobic and anaerobic total count

Only about 6% and 8% of the samples experienced any detectable (>2 log CFU) aerobic and anaerobic microbial growth during the 120 days of refrigerated storage (Table 2). For those samples that had detectable microbial growth, growth ranged from 2.0 to 4.0 (log CFU) during 120 days of refrigerated storage. A parallel study (Manomaiwiboon, 1990) showed that growth of aerobes and anaerobes increased approximately 1.5 log cycles during 40 days of storage at 4-6°C. Furthermore, the microbial growth detected was not significantly different in the different barriers. Simunovic et al. (1985) found for restructured beef, cooked in-package to 82.2°C for 15 minutes, that no aerobic or anaerobic bacterial growth occurred over 16 weeks of storage at 4.4°C. Smith and Alvarez (1988) inspected commercial cook-in-bag turkey rolls which were cooked to 71°C and stored at 4°C for 87 days. No psychotropic aerobic bacteria were found, but an average 188 MPN/g of mesophilic anaerobic bacteria were detected in the core of the turkey rolls. Stites et al. (1989) reported that the total aerobic count of vacuum cook-in-bag (70°C) beef roasts was less than 3.3 log CFU/g. Thus, the data in this study, are in apparent agreement with the previous work in the area in that microbial growth is generally low in these types of products.

Table 2. Total counts of aerobic and anaerobic microorganisms in pork patties vacuum packaged in nylon, saran, and SiO_2 barriers, pasteurized, and stored at 3°C for up to 120 days.

	Log CFU per gram of meat*							
Total aerobic count					Total anaerobic count			
Nylon Pouch	Rep1	Rep2	Rep3	Rep4	Rep1	Rep2	Rep3	Rep4
Day 1	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
5	<2.0	<2.0	<2.0	--	<2.0	<2.0	<2.0	--
30	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
60	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
90	**3.0**	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
120	**4.2**	<2.0	**2.0**	<2.0	**4.0**	<2.0	<2.0	<2.0
Saran Pouch	Rep1	Rep2	Rep3	Rep4	Rep1	Rep2	Rep3	Rep4
Day 1	<2.0	<2.0	<2.0	--	<2.0	<2.0	<2.0	--
15	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
30	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	**3.9**
60	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	**3.7**
90	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
120	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
SiO_2 Pouch	Rep1	Rep2	Rep3	Rep4	Rep1	Rep2	Rep3	Rep4
Day 1	<2.0	<2.0	<2.0	--	<2.0	<2.0	<2.0	--
15	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
30	**2.0**	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0
60	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	**2.0**	<2.0
90	<2.0	<2.0	**3.3**	<2.0	<2.0	<2.0	**3.3**	<2.0
120	<2.0	<2.0	<2.0	<2.0	<2.0	<2.0	**2.3**	<2.0

* Data represents an average log number of 2 replications.

Extensive microbial growth in the product could significantly influence lipid oxidation as well as the headspace gas composition (Ahvenainen et al., 1989). The results indicate that in-package pasteurization eliminated

most of the microorganisms and created an acceptable system in which to study lipid oxidation and its relationship to oxygen barrier level and headspace gas composition.

Lipid oxidation as measured by TBA test

Analysis of variance of the TBARS values was performed using the General Linear Model (GLM) procedure (SAS, 1985) based on a three factorial Randomized Complete Block (RCB) design with different batches as the block (replication) effect. The analysis of variance (ANOVA) for TBARS values of vacuum packaged pork patties evaluated during refrigerated storage is shown in Table 3.

Table 3. Analysis of variance of the TBARS values of vacuum packaged, in-package pasteurized pork patties stored at 3°C.

Source of Variation	DF	Sum of Square	Mean Square	F Value	Pr > F
Replication (R)	6	1.72	0.29	5.96	0.0001
Storage Day (D)	5	74.13	14.83	306.9	0.0001
Barrier (B)	2	167.96	83.98	1738.15	0.0001
Salt (S)	1	0.04	0.04	0.74	0.3918
D * B	10	85.47	8.55	176.9	0.0001
D * S	5	0.21	0.04	0.85	0.5126
B * S	2	0.73	0.37	7.6	0.0007
D * B * S	10	0.63	0.06	1.3	0.232
Error	198	9.57	0.05		
Total	239	340.46			

Replication (block), day (storage time), and barrier were all highly significant ($P<0.0001$). In addition, significant interactions were found between storage time and barrier,

(P<0.0001) and between barrier and salt (P<0.0007). Salt alone did not have any significant effect. Barrier level had the greatest effect on TBARS values and storage time had the second most important impact. There are three major factors (barrier level, storage time, salt level) that were considered in this test design. To evaluate the effect of one particular factor (either barrier, salt, or storage time) on lipid oxidation in the product, mean TBARS values of a second factor was compared and their significance determined. For example, when the effect of oxygen barrier on lipid oxidation was evaluated, the mean TBARS values for each storage day (the second factor) were compared with each other. This TBARS value is the average from samples of different levels of salt (the third factor) packaged in a specific barrier and stored for a particular time.

The initial quality of the raw pork meat was also measured using the TBA test. The mean TBARS value of 0.18 (standard deviation= 0.06, n=5) indicates that the level of lipid oxidation following slaughter and handling was low. TBARS values for all treatments increased almost linearly with storage time (Table 4, Fig. 4). Although statistic data did not show that salted samples had significant lower TBARS values than their non-salted counterparts, mean TBARS values of salted samples (except for SiO_2 samples) were always lower than non-salted samples in every storage day. SiO_2 sample had the lowest TBARS values and did not increase with storage time. Generally, nylon samples had

significantly ($P<0.05$) higher TBARS values than saran samples which TBARS values were higher but not significantly than SiO_2 samples.

Table 4. Effect of oxygen barrier and salt on the mean TBARS values* (mg/kg) of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	TBARS values (mg/kg) at storage day						
Treatment	1 **	15	30	60	90	120	Avg.
Nylon	0.41 ab (0.08)	1.07 a (0.28)	1.72 a (0.29)	2.76 a (0.26)	3.75 a (0.26)	4.58 a (0.68)	2.33 a (1.48)
Nylon/salt	0.47 a (0.09)	1.17 a (0.12)	1.62 a (0.25)	2.38 b (0.28)	3.42 a (0.44)	4.29 a (0.39)	2.23 a (1.34)
Saran	0.28 b (0.05)	0.49 bc (0.12)	0.57 b (0.14)	0.74 c (0.16)	0.90 b (0.15)	1.13 b (0.12)	0.69 b (0.30)
Saran/salt	0.39 ab (0.07)	0.62 b (0.17)	0.64 b (0.10)	0.8 c (0.13)	1.05 b (0.11)	1.31 b (0.21)	0.81 b (0.33)
SiO_2	0.33 ab (0.12)	0.49 bc (0.04)	0.38 b (0.04)	0.32 d (0.07)	0.32 c (0.08)	0.42 c (0.07)	0.37 b (0.09)
SiO_2/salt	0.3 b (0.06)	0.37 c (0.07)	0.41 b (0.08)	0.27 d (0.05)	0.36 c (0.10)	0.4 c (0.07)	0.35 b (0.09)

*All values represent the average of seven replications. Number in parentheses is the standard deviation of the replications.

**Means within the column followed by a common letter are not different ($P<0.05$).

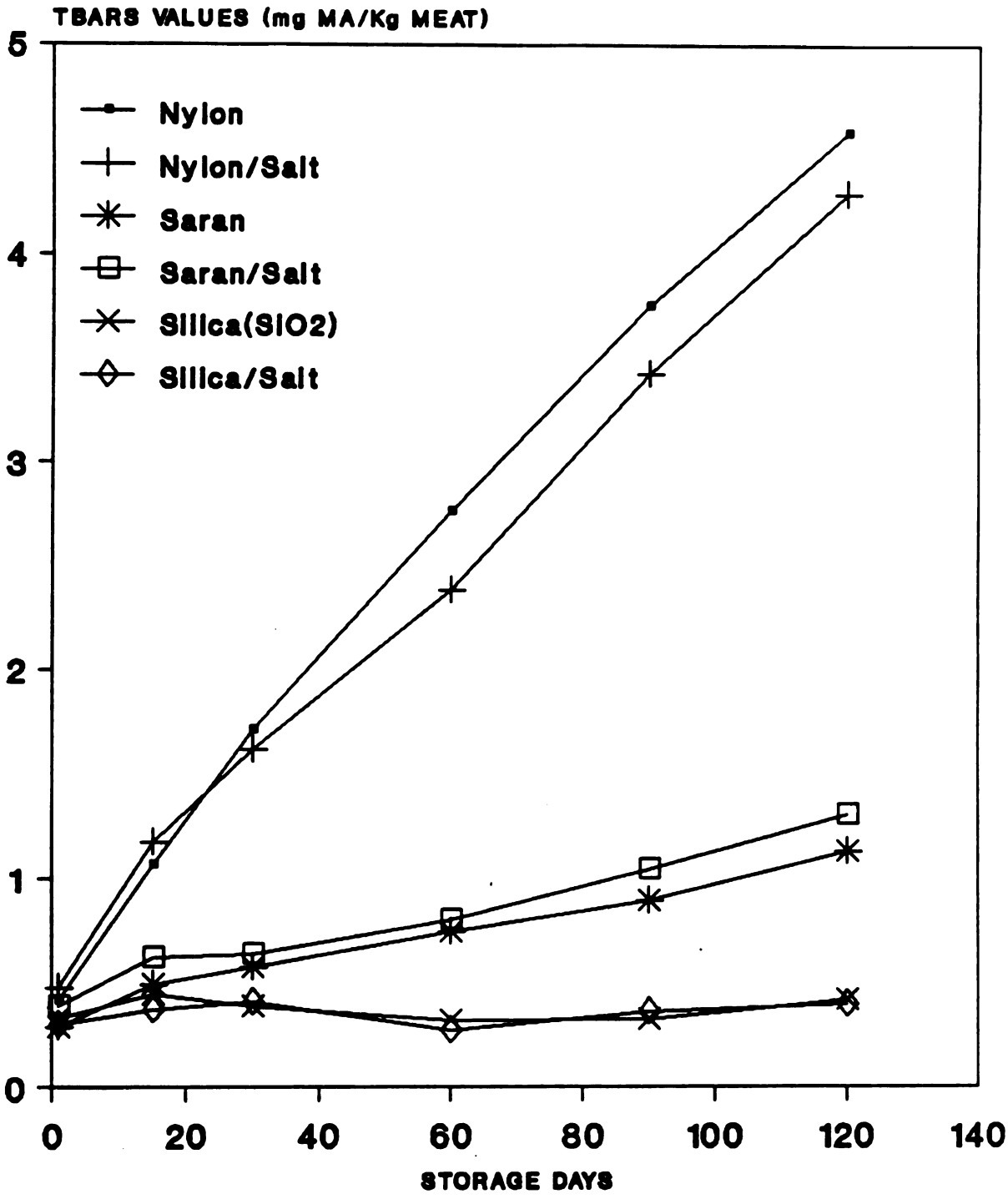


Figure 4. Effect of oxygen barrier and salt on mean TBARS values of vacuum packaged, in-package cooked pork patties stored at 3°C.

In Table 5 are shown the linear regression values for lipid oxidation (TBARS values) and storage time and between hexanal content and storage time. Samples packaged in nylon and saran barriers had a very high (R^2= 0.995 and 0.981, respectively) linear relationship between TBARS value and storage time. For the silica barrier, however, there was poor correlation (R^2=0.02) probably because: First, the silica barrier has a very low OTR, therefore, the corresponding change in TBARS values was very low and unable to distinguish the difference among storage days. Second, the TBA test seems unable to consistently measure the initial stages of lipid oxidation (Shahidi et al., 1987), especially in the low oxidation (SiO_2 packaged) samples.

Table 5. Linear regression data (R square) between TBARS values and storage time of pork patties vacuum packaged in nylon, saran, and silica barriers, in-package pasteurized, and stored at 3°C.

	TBA Test	
Treatments	R Square	Slope
Nylon	0.995	0.0329
Saran	0.981	0.0068
SiO2	0.024	0.00017

TBA values sometimes decline during the final stage of extended storage, possibly because malonaldehyde (MA) reacted with proteins or amino acids in the meat (Melton, 1983; Gokalp et al., 1983), however, were not found here. The linear relationship between TBARS values and storage

time demonstrates that the TBA test is an effective method that can be used to monitor Lipid oxidation during storage. If a relationship between TBARS values and sensory evaluation data can be established, then TBA test can also be an useful tool to predict the shelf life of the product. Many researchers have shown that TBARS values are related to the sensory scores of oxidized and warmed-over flavors (Igene and Pearson, 1979; Igene et al, 1979). Threshold oxidized flavor expressed as TBARS values was 0.6-2.0 in cooked beef (Greene and Cumuze, 1981) and 0.5-1.0 in ground pork (Tarladgis et al., 1960).

Effect of oxygen barrier on lipid oxidation as measured by the TBA test The type of oxygen barrier had a significant impact on lipid oxidation in pork patties during storage. In , the effect of oxygen barrier on the mean TBARS values of vacuum packaged, precooked pork patties is shown. Mean TBARS values of saran and SiO_2 packaged samples were about the same and both were lower than that of nylon packaged samples at day one of storage. After 15 days of storage, TBARS values of the saran samples were higher ($P<0.05$) than that of SiO_2 samples. Both saran and SiO_2 samples had TBARS values lower than nylon samples. The better the oxygen barrier, the lower the TBARS values. These results concur with that of Simunovic et al. (1985) who used an aluminum-laminated retort pouch to package restructured beef (very low OTR) and reported TBARS values

of 0.1-0.2 during 16 weeks of refrigerated storage. Stites et al. (1989) used a CN 350 barrier (Cryovac, OTR= 30 $ml/m^2/24$ hr) to package beef roast and reported TBARS values of 0.30 - 0.45 during 4 weeks of refrigerated storage. Smith and Alvarez (1988) used a nylon/PE (OTR about 155 $ml/m^2/24$ hr) material to package beef and reported TBARS values of 0.30 - 1.0 during 10 weeks of refrigerated storage.

Table 6. Effect of storage time and oxygen barrier on the mean TBARS values* (mg/kg) of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

STORAGE DAYS	Oxygen Barriers											
	NYLON			SARAN			SiO_2			AVERAGE		
	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**
1	0.44	0.09	f^{A}	0.34	0.08	e^{B}	0.31	0.1	bc^{B}	0.37	0.11	f
15	1.12	0.22	e^{A}	0.56	0.16	d^{B}	0.43	0.07	a^{C}	0.72	0.35	e
30	1.67	0.28	d^{A}	0.61	0.13	d^{B}	0.4	0.07	ab^{C}	0.93	0.59	d
60	2.57	0.33	c^{A}	0.77	0.15	c^{B}	0.3	0.07	c^{C}	1.24	1.01	c
90	3.58	0.4	b^{A}	0.97	0.15	b^{B}	0.34	0.09	ab c^{C}	1.66	1.43	b
120	4.42	0.56	a^{A}	1.23	0.2	a^{B}	0.41	0.07	ab^{C}	2.02	1.77	a
AVERAGE	2.28	1.41	^{A}	0.75	0.32	^{B}	0.36	0.09	^{C}	1.14	1.19	

*All values represent the average of salt and no-salt samples. Each sample point is the result of fourteen replications. Std= Standard deviation.

**Means within the same column (subscript) or same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Effect of extended storage on lipid oxidation as measured by TBA test The effect of refrigerated storage on the mean TBARS values of vacuum packaged, precooked patties is shown in Table 6. Inhibition of lipid oxidation due to the silica coated barrier was so effective that TBARS values were always below 0.43, no difference ($P<0.05$) could be found among samples during 120 days of refrigerated storage. Saran, the second best barrier, also performed well and TBARS values were below 1.0 at 90 days of storage; however, the increase in TBARS values was significant ($P<0.05$) as storage time increased. According to Tarladgis et al. (1960), a rancid flavor can first be perceived at a TBARS values of 0.5-1.0. Thus, in the saran barrier, pork patties may have 15-90 days of shelf life without development of rancid flavor. Nylon was the poorest barrier used in the test. Mean TBARS values after 15 days of storage exceeded 1.0 and increased rapidly up to 4.4 at the 120th day. Smith and Alvarez (1988) used turkey breast samples which were first vacuum packaged in a nylon bag and then cooked-in the bag with each sample containing 1.5% salt and 0.5% phosphate and found TBARS values ranging from 0.3 (0 day) to 1.0 after 68 days. Although nylon materials have been used as cook-in barriers (Siegel, 1982; Terlizzi et al., 1984) for many years, antioxidants including phosphate, have often been added to meat packaged in nylon in order to prolong the product's shelf life.

Sheu (1988) used a similar meat model system (prerigor meat) to study lipid oxidation of pork patties during refrigerated storage. The samples were first cooked in a cooking bag and then removed and resealed (vacuum packaged) in a saran barrier bag. Higher initial TBARS value and a greater increase in TBARS values during storage were reported than those in the current study. The analytical procedure (TBA test) used for the two studies was the same. In-package cooking reduced the chance of samples being exposed to oxygen after sealing and cooking. This helps to explain why there were low initial TBARS and slow increase in lipid oxidation during storage. Vacuum packaging with subsequent in-package cooking has been reported to extend meat product shelf-life by inhibiting lipid oxidation and microbial growth (Siegel, 1982; Terlizzi et al., 1984; Simunovic et al., 1985; Sofos, 1986a; Jones et al., 1987; Prabhu et al, 1988; Randall, 1988; Stites et al., 1989).

Effect of salt on lipid oxidation as measured by TBA test The effect of salt on the mean TBARS values of vacuum packaged, precooked patties is shown in Table 7. Nylon/Salt samples had lower (although not significantly) mean TBARS values than that of the Nylon/No-salt samples. Saran/Salt samples, on the contrary, had small but significantly higher mean TBARS values than that of Saran/No-salt samples. In the silica barrier, no significant difference in TBARS values could be detected between salt and non-salt samples during storage. No difference can be detected between the

average TBARS value of salted (1.16) and non-salted (1.15) samples, indicated that TBA test has limitation in detecting the effect of salt (1.5%) on lipid oxidation in the system. Since the effect of salt in a meat system is still poorly understood (Gary and Weiss, 1988), further study on the effect of salt is required, especially in precooked, vacuum packaged refrigerated products.

Table 7. Effect of salt (1.5%) on the mean TBARS values* (mg/kg) of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**
No Salt	2.33	1.48	a	0.69	0.3	a	0.37	0.09	a	1.15	1.24	a
Salted	2.23	1.34	a	0.81	0.33	b	0.35	0.09	a	1.16	1.14	a
AVERAGE	2.28	1.41	A	0.75	0.32	B	0.36	0.09	C	1.14	1.19	

*All values represent the average of six testing days with each test day replicated seven times. Std= Standard deviation.

**Means within the same column (subscript) or the same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Effect of 24 hours open-package storage after extended storage on lipid oxidation as measured by TBA test In Fig. 5 and Table 8 are shown the effects of extended open-package storage on lipid oxidation of the pork patties during 120 days of refrigerated storage. All sample packages were stored at 3°C for 1, 15, 30, 60, 90, 120 days, opened, and stored for another 24 hours before analyzing the sample. After one day of storage (1 day closed, followed by 24 hours of open storage), all the samples exposed to air were close to a TBARS value of 2.0. Using barriers like silica and saran did not have much advantage in reducing lipid oxidation over nylon. Instead, TBARS values of these samples were close to a level that was found in the nylon barrier samples. Nevertheless, when storage time (closed only) increased, the high barrier silica samples did not show significant ($P<0.05$) increase in TBARS values. The TBARS values in the lower barrier saran and nylon increased during the 120 days of (closed) storage (Table 9). Saran and silica packaged samples, after 60 days and 24 hours of open storage had lower ($P<0.05$) TBARS values than the nylon samples (Table 9).

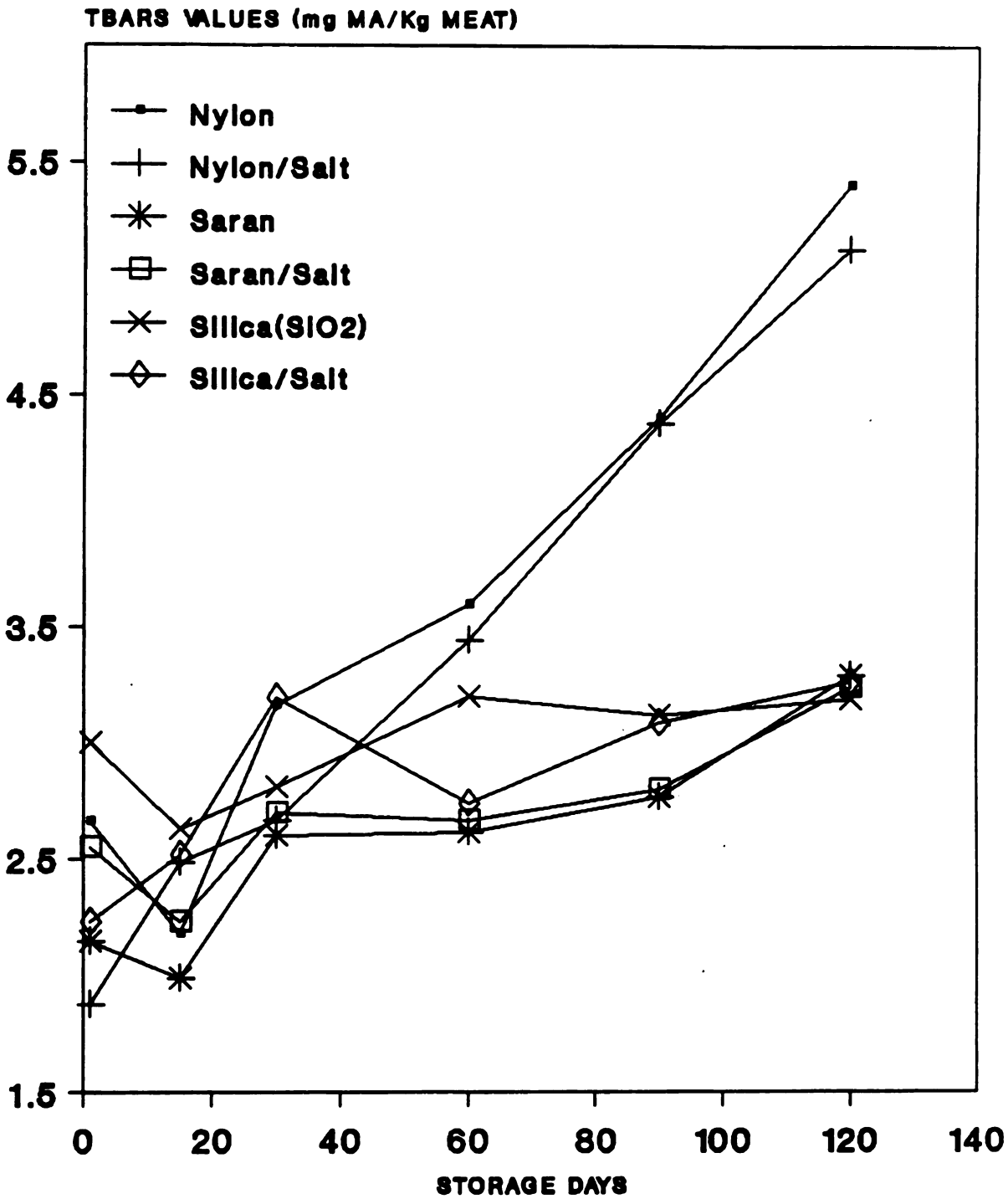


Figure 5. Effect of oxygen barrier and salt on mean TBARS values of vacuum packaged, cooked pork patties stored at 3°C for 1, 15, 30, 90, 120 days, opened, and restored for 24 hrs.

Table 8. Effect of oxygen barrier and salt on the mean TBARS values (mg/kg) of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, stored at 3°C for 1, 15, 30, 60, 90, 120 day, at each day, package was opened and stored for additional 24hrs.

	TBARS values* (mg/kg) at storage day						
Treatment	1 **	15	30	60	90	120	Avg.
Nylon	2.66 a (0.89)	2.18 a (0.58)	3.16 a (0.66)	3.6 a (0.51)	4.4 a (0.57)	5.4 a (0.60)	3.62 a (1.26)
Nylon/salt	1.88 a (0.54)	2.49 a (0.60)	2.67 a (0.39)	3.44 a (0.29)	4.38 a (0.83)	5.13 a (0.79)	3.47 ab (1.31)
Saran	2.15 a (1.26)	1.99 a (0.74)	2.61 a (0.75)	2.62 a (0.23)	2.77 b (0.31)	3.3 b (0.51)	2.59 c (0.82)
Saran/salt	2.55 a (0.38)	2.23 a (0.60)	2.7 a (0.32)	2.67 a (0.20)	2.8 b (0.59)	3.25 b (0.50)	2.72 c (0.56)
SiO_2	2.66 a (0.80)	2.63 a (0.75)	2.81 a (0.26)	3.2 a (0.73)	3.12 b (0.69)	3.19 b (1.20)	2.99 abc (0.81)
SiO_2/salt	2.23 a (0.64)	2.52 a (0.70)	3.2 a (0.54)	2.74 a (0.59)	3.09 b (0.95)	3.27 b (1.21)	2.88 bc (0.72)

*All values represent the average of seven replications. Number in parentheses is the standard deviation of the replications.

**Means within the column followed by a common letter are not different ($P<0.05$).

Table 9. Effect of storage time and oxygen barrier on the mean TBARS values* (mg/kg) of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, stored at 3°C for 1, 15, 30, 60, 90, 120 day, at each day, package was opened and stored for additional 24hrs.

STORAGE DAYS	Oxygen Barriers											
	NYLON			SARAN			SiO_2			AVERAGE		
	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**
1	2.27	0.83	d^{A}	2.35	0.95	b^{A}	2.62	0.82	a^{A}	2.41	0.88	de
15	2.32	0.61	d^{A}	2.11	0.68	b^{A}	2.58	0.73	a^{A}	2.34	0.7	e
30	2.92	0.59	cd^{A}	2.65	0.58	ab^{A}	3.01	0.33	a^{A}	2.86	0.54	dc
60	3.52	0.42	c^{A}	2.64	0.22	ab^{B}	2.97	0.66	a^{AB}	3.05	0.59	bc
90	4.39	0.71	b^{A}	2.79	0.47	ab^{B}	3.11	0.72	a^{B}	3.43	0.95	b
120	5.26	0.72	a^{A}	3.27	0.51	a^{B}	3.23	0.96	a^{B}	3.92	1.21	a
AVERAGE	3.45	1.29	^{A}	2.66	0.7	^{C}	2.94	0.77	^{B}	3	1.03	

*All values represent the average of salt and no-salt samples. Each sample point is the result of fourteen replications. Std= Standard deviation.

**Means within the same column (subscript) or same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

When compare Table 6 with Table 9, the effect of 24 hours open-storage actually increase the TBARS values by 2 to 6.5 times depends on how long the extended (close package) storage was. The longer the extent storage time was, the lower the effect of 24 hours open-storage on TBARS values increasing will be. Smith and Alvarez (1988) reported that TBA numbers will double within 2-4 hours after opening uncured, vacuumed turkey rolls. Simunovic et al. (1985) reported only slight increases in TBARS values in

retort pouch packaged restructured beef after 2 weeks of open storage. Because malonaldehyde is able to react with protein and amino acids in muscle foods (Melton, 1983; Gokalp et al., 1983) therefore, TBARS values will be smaller than expected. Those studies confirm the current study that high level of malonaldehyde (from extent and open-storage) may reacted with amino acids and protein, then resulted in a lower TBARS values than expected.

Although the samples with salt have lower average TBARS values than non-salted samples, differences were not significant ($P>0.05$). The effect of salt on TBARS values of refrigerated samples after 24 hours of open storage is shown in Table 10).

Table 10. Effect of salt (1.5%) on the mean TBARS values* (mg/kg) of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for 1, 15, 30, 60, 90, 120 day, at each day, package was opened and stored for additional 24hrs.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**	TBARS	Std	**
No Salt	3.62	1.26	a	2.72	0.82	a	2.99	0.81	a	3.06	1.07	a
Salted	3.47	1.31	a	2.59	0.56	a	2.88	0.72	a	3.02	0.97	a
AVERAGE	3.54	1.29	A	2.66	0.7	C	2.94	0.77	B	1.14	1.03	

*All values represent the average of six testing days with each test day replicated seven times. Std= Standard deviation.

**Means within the same column (subscript) or the same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Lipid oxidation as measured by hexanal content

The analysis of variance (ANOVA) for hexanal contents of vacuum packaged pork patties during refrigerated storage is shown in Table 11. Replication (block), day (storage time), barrier, and salt all show highly significant ($P<0.0001$) effects. In addition, significant interaction effects were found between day and barrier ($P<0.0001$). Unlike that found for the TBA test, hexanal showed that salt alone significantly increased the hexanal content in the system. Barrier had the greatest, storage time the second, and salt the third largest effect on hexanal content in the patties.

Table 11. Analysis of variance of the hexanal contents of vacuum packaged pork patties, pasteurized, stored at 3°C for 120 days.

Source of Variation	DF	Sum of Square	Mean Square	F Value	Pr > F
Replication (R)	6	604.67	100.77	24.25	0.0001
Storage Day (D)	5	2081.79	416.35	100.19	0.0001
Barrier (B)	2	6146.52	3073.26	739.5	0.0001
Salt (S)	1	84.1	84.1	20.24	0.0001
D * B	10	2141.67	214.16	51.53	0.0001
D * S	5	10.1	2.02	0.49	0.78
B * S	2	2.07	1.03	0.25	0.779
D * B * S	10	12.24	1.22	0.29	0.98
Error	198	706.49	4.15		
Total	239	11789.7			

Generally, product hexanal content vs. storage time (Fig. 6 & Table 12) of all six treatments showed the same

trend as that of TBARS vs. storage time (Fig. 4). Except for nylon barrier, statistic data (Table 17) did show that salted samples had significant lower hexanal contents than their non-salted counterparts. Mean hexanal contents of salted samples (all 3 barrier samples) were always lower than non-salted samples in every storage day.

Table 12. Effect of oxygen barrier and salt on the mean hexanal content* (ppm) of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Hexanal content (ppm) at storage day						
Treatment	1 **	15	30	60	90	120	Avg.
Nylon	0.47 $_{a}$ (0.08)	0.8 $_{a}$ (0.28)	1.28 $_{a}$ (0.29)	1.64 $_{a}$ (0.26)	2.31 $_{a}$ (0.26)	2.77 $_{a}$ (0.68)	1.6 $_{a}$ (1.48)
Nylon/salt	0.42 $_{ab}$ (0.09)	0.71 $_{a}$ (0.12)	1.09 $_{a}$ (0.25)	1.64 $_{a}$ (0.28)	2.26 $_{a}$ (0.44)	2.55 $_{a}$ (0.39)	1.55 $_{a}$ (1.34)
Saran	0.33 $_{abc}$ (0.05)	0.41 $_{b}$ (0.12)	0.58 $_{b}$ (0.14)	0.77 $_{b}$ (0.16)	0.83 $_{b}$ (0.15)	0.89 $_{b}$ (0.12)	0.66 $_{b}$ (0.30)
Saran/salt	0.29 $_{bc}$ (0.07)	0.3 $_{bc}$ (0.17)	0.4 $_{bc}$ (0.10)	0.5 $_{c}$ (0.13)	0.7 $_{b}$ (0.11)	0.78 $_{b}$ (0.21)	0.51 $_{bc}$ (0.33)
SiO_2	0.28 $_{bc}$ (0.12)	0.32 $_{bc}$ (0.04)	0.42 $_{bc}$ (0.04)	0.4 $_{cd}$ (0.07)	0.45 $_{bc}$ (0.08)	0.41 $_{bc}$ (0.07)	0.38 $_{bc}$ (0.09)
SiO_2/salt	0.22 $_{c}$ (0.06)	0.2 $_{c}$ (0.07)	0.21 $_{c}$ (0.08)	0.26 $_{d}$ (0.05)	0.27 $_{c}$ (0.10)	0.27 $_{c}$ (0.07)	0.24 $_{c}$ (0.09)

*All values represent the average of seven replications. Number in parentheses is the standard deviation of the replications.

**Means within the column followed by a common letter are not different ($P<0.05$).

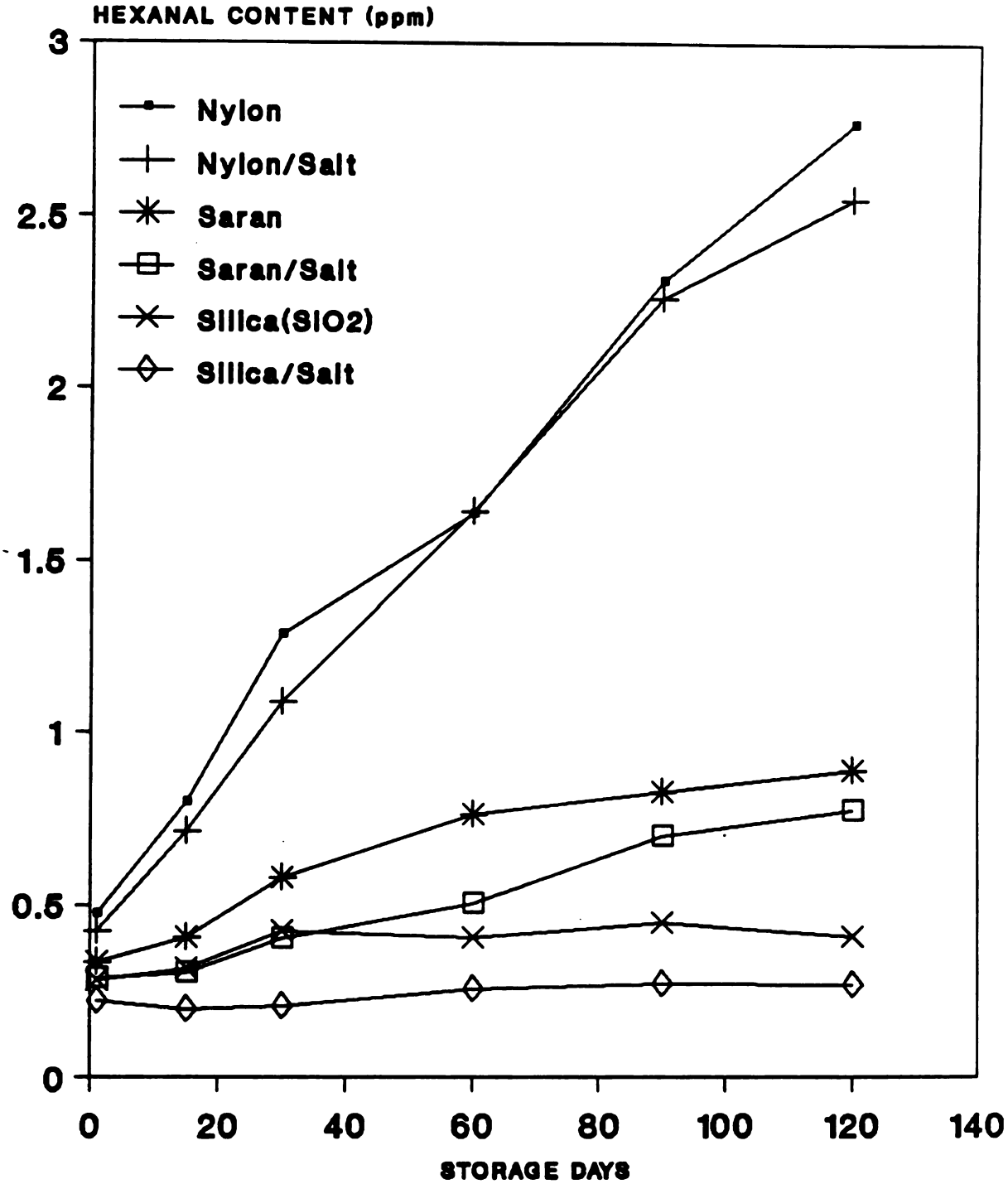


Figure 6. Effect of oxygen barrier and salt on mean hexanal content of vacuum packaged, in-package cooked pork patties, stored at 3°C.

SiO_2 samples had the lowest hexanal contents and did not increase with storage time. Generally, nylon samples had significantly ($P<0.05$) higher TBARS values than saran samples, and saran samples had significantly higher hexanal contents than SiO_2 samples.

Mean hexanal contents of pork patties for all treatments increased linearly with storage time. The linear regression analysis of hexanal and storage time is shown in Table 13. Both nylon and saran barrier had a very high (R^2= 0.99 and 0.97, respectively) correlation between hexanal content and storage time. Silica barrier had a moderate correlation (R^2=0.74) because only limited change in hexanal was observed during the storage time. The hexanal analysis was more sensitive and more consistent than the TBA test in measuring lipid oxidation, especially in the early stages of the oxidation. This finding was in agreement with Shahidi et al. (1987). Hexanal can be a useful marker of lipid oxidation in precooked meat in

Table 13 Linear regression data (R square) between hexanal contents (ppm) and storage time of pork patties vacuum packaged in nylon, saran, and silica barriers, in-package pasteurized, and stored at 3°.

	Hexanal Analysis	
Treatments	R Square	Slope
Nylon	0.99	0.01872
Saran	0.973	0.0046
SiO2	0.739	0.00084

extended storage (St. Angelo et al., 1987; Dupuy et al., 1987; Shahidi et al., 1987; Lai, 1989; Ang and Lyon, 1990; Hwang et al., 1990). Unfortunately, no other hexanal data have been reported for vacuum packaged, precooked, meat in extended refrigerated storage.

The high correlation between hexanal content and storage time not only demonstrated that hexanal analysis is a useful method to measure lipid oxidation in this product, but also that it could be used as a tool to predict the shelf life of the product if a relationship between hexanal and sensory evaluation data can be established. Hwang et al. (1990) studied vacuum packaged, cooked, sliced beef after 11 weeks of frozen storage. They reported a correlation coefficient of 0.89 between warmed-over flavor and hexanal content, and 0.89 between warmed-over flavor and TBARS values. Similar results were reported in cooked ground pork refrigerated in a plastic bag (r=0.98, Shahidi et al., 1987), and in refrigerated beef roasts (r=0.8, St. Angelo et al., 1987). Unfortunately, none of above two report have the threshold hexanal content (ppm) in terms of lipid oxidation.

Three things in the current study that make conversion between TBARS values and hexanal contents possible (Table 5 and Table 13): 1. the high linear relationship between TBARS values and storage time. 2. the high linear relationship between hexanal content and storage time and 3. high correlation coefficients between TBARS values and

hexanal contents. In Table 5, the slope of the TBA-storage curve is 0.0329 (nylon), and the hexanal-storage curve is 0.0187 (nylon), hence, a conversion factor of 1.76 (0.0329/0.0187) is calculated between TBARS values and hexanal content in the nylon barrier. Likewise, the conversion factor for patties in the saran barrier is 1.46. The silica barrier was poorly correlated, so calculation of a conversion factor is meaningless. These factors concur with the work of Dupuy et al. (1987), who used a direct GC technique to measure the hexanal content of a refrigerated roast beef. Using the same approach, an equivalent conversion factor of 1.65 was obtained based on Dupuy et al., (1987) data. Since TBARS values at 0.5-1.0 have been reported (Tarladgis et al., 1960) to be the threshold level of rancidity, we can speculate that hexanal content at around 0.63 ppm may be the threshold level of rancidity for this system. Further studies to correlate sensory evaluation are necessary to establish this level.

Correlation coefficients between TBARS values and hexanal contents In Table 14 & Table 15 are shown the within-time and within-treatment correlation coefficients between TBARS values and hexanal in pork patties for the different treatments and storage times. The within-time relationships between TBARS values and hexanal contents are significantly correlated ($r = 0.69-0.89$, $P<0.0001$) except in samples which were packaged in the silica barrier (Table

14). Low correlation coefficients were found for the silica barrier, due to limited lipid oxidation.

Table 14. Within-time correlation coefficients between TBARS values and hexanal content of pork patties for each treatment.

Treatment	Correlation Coeff.	Significant Level P
nylon	0.87	<0.0001
nylon/salt	0.89	<0.0001
saran	0.8	<0.0001
saran/salt	0.69	<0.0001
SiO_2	0.07	NS*
SiO_2/ salt	-0.01	NS*
Pooled	0.93	<0.0001

* No Significant difference (P>0.05)

Table 15. Within-treatment correlation coefficients between TBARS values and hexanal content of pork patties at each storage time.

Storage Days	Correlation Coeff.	Significant Level P
1	0.41	<0.0229
15	0.85	<0.0001
30	0.81	<0.0001
60	0.95	<0.0001
90	0.94	<0.0001
120	0.91	<0.0001
Pooled	0.93	<0.0001

Correlation coefficients between TBARS values and hexanal contents were: 0.81 (0.01) in vacuum packaged, frozen beef loin slices (Hwang et al., 1990), 0.92 in beef roast (St.

Angelo et al., 1987), and 0.995 ($P<0.05$) in cooked ground pork refrigerated in a plastic bag (Shahidi et al., 1987).

Lai (1989) reported a correlation coefficient of 0.68 (pooled, within treatment) between hexanal contents and sensory scores of frozen chicken nuggets. A correlation coefficient of 0.55 between TBARS values and sensory scores was also reported. It was concluded that because of the poor correlation coefficient between hexanal content and TBARS values, the use of hexanal analysis may be more sensitive than the TBA test for evaluation of the warmed-over flavor of chicken nuggets during 6 months of frozen storage. A correlation coefficient of 0.93 (pooled) was obtained between TBARS values and hexanal content in the present study.

A high correlation coefficient between hexanal content and TBARS values does not guarantee that TBA truthfully represents lipid oxidation throughout storage. Shahidi et al. (1987) reported that even when there is a high correlation coefficient ($r= 0.995$) between TBA values and hexanal, the TBA test still does not accurately measure lipid oxidation of the cooked pork samples during the early stages of storage. Shahidi et al. (1987) reported that after 2 days of storage, all of the samples had almost identical TBA values. Accordingly, hexanal analysis maybe a better measure of the oxidative state of the cooked meat in the early stages of storage. The within-treatment relationships between TBARS values and hexanal contents

(Table 15) were well correlated (r= 0.81-0.95, P<0.0001) throughout storage, except for day 1 where a correlation coefficient of 0.41 was found. The excellent correlation between TBARS values and hexanal content plus the fact that both TBARS values and hexanal contents were linear with storage time implies that both the TBA test and hexanal analysis effectively monitored the formation of lipid oxidation throughout the 120 days of refrigerated storage.

Effects of oxygen barrier on lipid oxidation as measured by hexanal analysis In general, hexanal results were similar to those found with the TBA test. The Oxygen barrier significantly retarded lipid oxidation in the pork patties during storage. The effect of oxygen barrier on the mean hexanal contents of vacuum packaged, precooked pork patties in refrigerated storage is shown in Table 16. The mean hexanal contents of saran and SiO_2 packaged samples were lower than that of nylon packaged samples at day one of storage, which is the same as found for TBARS values (Table 6). At the 60th day of storage, the hexanal content of saran samples was significantly higher than in the SiO_2 samples (Table 16). In contrast, the TBA method first detected a significant difference at the 15th day of storage (Table 6). The reason why the hexanal analysis is not able to detect a significant difference between saran and SiO_2 packaged samples in the early stage of storage is probably because of some data were lost during the early stage of hexanal analysis. The missing data resulted due to high

level of autooxidation occurring during distillation in the hexanal analysis. This problem was later corrected by adding antioxidant in the distillation procedure.

Table 16. Effect of storage time and oxygen barrier on the mean hexanal content*(ppm) of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	HEX	Std	**	HEX	Std	**	HEX	Std	**	Hex	Std	**
1	0.45	0.13	e^{A}	0.31	0.07	d^{B}	0.25	0.07	c^{B}	0.34	0.12	d
15	0.76	0.14	e^{A}	0.35	0.06	d^{B}	0.26	0.10	bc^{B}	0.46	0.24	d
30	1.19	0.34	d^{A}	0.49	0.12	c^{B}	0.32	0.13	abc^{B}	0.66	0.44	c
60	1.64	0.20	c^{A}	0.63	0.18	b^{B}	0.33	0.12	abc^{C}	0.86	0.59	b
90	2.28	0.31	b^{A}	0.77	0.21	a^{B}	0.35	0.10	a^{C}	1.17	0.85	a
120	2.65	0.56	a^{A}	0.84	0.21	a^{B}	0.34	0.09	ab^{B}	1.28	1.05	a
AVERAGE	1.57	0.84	^{A}	0.59	0.25	^{B}	0.31	0.11	^{C}	0.79	0.75	

*All values represent the average of salt and no-salt samples. Each sample point is the result of fourteen replications. HEX=hexanal content, Std.= Standard deviation.

**Means within the same column (subscript) or same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

The overall performance of oxygen barriers was as follows: silica > saran >> nylon. The lower the OTR of the oxygen barrier, the lower the hexanal content in the pork

patties. Hwang et al. (1990) reported that hexanal content in vacuum and air packaged (EVOH pouch) cooked beef loin slices after 11 weeks at -20°C was 0.3 and 0.82 ppm, respectively. Since 0.3 ppm is a relatively low hexanal level, it is believed that high oxygen barriers such as EVOH do effectively inhibit lipid oxidation during frozen storage.

Effect of extended storage on lipid oxidation as measured by hexanal analysis The effect of storage time on the mean hexanal contents of vacuum packaged, precooked patties is shown in Table 16. The silica coated barrier almost completely inhibited lipid oxidation and production of hexanal as values were always below 0.35 ppm during the 120 days of refrigerated storage. However, unlike the TBA test, which was unable to detect any change, hexanal analysis first detected an increase in lipid oxidation in silica barrier samples at the 90th day. Saran, the second best barrier in the test, also inhibited production of hexanal and values were below 0.63 ppm at 60 days of storage. The hexanal contents of saran packaged samples increased ($P<0.05$) slowly with storage time. Nylon (the poorest barrier) samples had mean hexanal contents in excess of 0.6 ppm after 15 days of storage, which continued to increase to 2.65 ppm at the 120th day.

Effect of salt on lipid oxidation as measured by hexanal analysis The effect of salt on the mean hexanal contents of vacuum packaged, precooked patties is shown in

Table 17. Throughout the storage period, nylon/salt samples had lower, although not significantly lower, mean hexanal contents than nylon/No-salt samples. The antioxidant effect of salt in saran and silica packaged pork patties, was more profound and statistically significant ($P<0.05$). The antioxidant effectiveness of salt seems more prominent in high barrier materials than in low barrier. In the silica barrier samples, salt reduced the mean hexanal content by 38%, whereas it was 23% in saran and 3% in nylon.

Table 17 Effect of salt (1.5%) on the mean hexanal content (ppm) of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	HEX	Std	**	HEX	Std	**	HEX	Std	**	HEX	Std	**
No Salt	1.6	1.48	a	0.66	0.3	a	0.38	0.09	a	0.88	1.24	a
Salted	1.55	1.34	a	0.51	0.33	b	0.24	0.09	b	0.78	1.14	b
AVERAGE	1.57	0.84	A	0.59	0.25	B	0.31	0.11	C	0.79	0.75	

*All values represent the average of six testing days with each test day replicated seven times. HEX= Hexanal Content. Std= Standard deviation.

**Means within the same column (subscript) or the same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

One of the reasons that salt has an antioxidant effect may be due to its ability to reduce nonheme iron in

precooked meat systems. Ang and Young (1989) studied the effect of ionic strength (IOS, adjusted with NaCl) on oxidative stability of broiler breast patties. They found that increasing IOS to 0.6 (2.1% NaCl) increased TBARS values in raw meat and during cooking, but had a reverse effect on cooked-stored patties, which was due to the lower nonheme iron content of cooked patties. They also found that when IOS (NaCl) was increased to 0.6, nonheme iron content in cooked samples dropped significantly but not in raw samples. Another reason that support salt be an antioxidant is its ability to reduce oxygen solubility in vacuum packaged meat systems. Mabrouk and Dugan (1960) observed the inhibitory effect of NaCl in methyl linoleate and linoleic acid emulsions in aqueous solution. They concluded that it may result from the decreased solubility of oxygen in emulsion with increased concentrations of NaCl. Consequently, oxygen availability would be a limiting factor in the reaction rate.

Bolland (1946) stated that the oxidation rate of ethyl linoleate is dependent on oxygen pressure when oxygen pressure is below a critical level of 200 mmHg. According to Raoult's law (Maron and Lando, 1976), the amount of oxygen in a solution (a closed system) can be represented by the oxygen pressure in the headspace. Data (Table 26) from the present study support this finding and show that salt did reduce package headspace oxygen. However, Dupuy et al. (1987) found that addition of 0.5% NaCl solution to ground

beef prior to cooking in a boil-in-bag pouch doubled hexanal formation during storage at 4°C for one day. Because Dupuy et al. (1987) did not vacuum package nor seal the cooking bag, oxygen availability was never a limiting factor in the system. It is speculated that the reason that salt behaves as an antioxidant in the current study because: 1. vacuum packaging of the pork patties makes oxygen availability a limiting factor, 2. addition of salt decreases the solubility of oxygen in the system (Mabrouk and Dugan, 1960), and 3. nonheme iron content in the precooked patties decreases as salt (or ionic strength) level increases (Ang and Young, 1989). The results (Table 26) show that oxygen availability did become a limiting factor in the system and that the better the oxygen barrier, the greater the antioxidant activity of the salt. Since the effect of salt in a meat system is still poorly understood (Gray and Weiss, 1988), further study on the effect of salt is required.

Lipid oxidation and headspace CO_2 concentration

There are three major catalytic systems involved in lipid oxidation--heme iron, nonheme iron and microsomal enzymes (Love, 1983). The oxidative deterioration of muscle foods has been described as being primarily due to nonenzymatic oxidation reactions (Gardner, 1980). Love (1983) stated that the nonenzymatic systems have greater significance at higher temperatures, especially in cooked meat. Enzymes like lipoxygenase or other lipid-oxidation

enzymes are normally inactivated by heat treatment. Researchers (Laleye et al., 1984a, b; Ahvenainen et al., 1990a, b) have reported that package headspace CO_2 increased in vacuum packaged, cured, cooked meat due to increased microbial growth during storage. Laleye et al. (1984a) reported that headspace CO_2% in vacuum packaged pastrami, stored at 3°C, was 40% on day one and 90% at the 49th day of storage. They concluded that the increased CO_2 reflected predomination of the product by a *Lactobacillus* spp. In this study, product headspace CO_2% decreased during storage and was always below 12% (Table 18 & Fig. 7). Therefore, microbial growth should not have been a problem in this system, which is in agreement with the results reported previously (Table 2).

Most enzymes will be partially or totally inactivated when heated to 79°C (Desrosier and Desrosier, 1977). In the current study, pork patties were cooked at 82°C for 20 minutes, so most of the microorganisms and thus enzyme activity should have been destroyed by the in-package pasteurization. Therefore, lipid oxidation should be mainly due to nonenzymatic reactions, and the presence of headspace CO_2 in the precooked, vacuum packaged system should mainly due to the end product of autoxidation of the meat. Thus, it may be possible to monitor lipid oxidation from the headspace CO_2% change in the system. CO_2 may also migrate through the packaging barrier, and thus, headspace CO_2

concentration depends as well on the CO_2 transmission rate through the barrier material.

Table 18. Effect of oxygen barrier and salt on the mean headspace CO_2 percentage* of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	headspace CO_2 % at storage day						
Treatment	1 **	15	30	60	90	120	Avg.
Nylon	8.2 b (1.22)	6.3 c (1.29)	4.1 c (1.38)	2.3 c (0.92)	1.0 c (0.54)	0.8 c (0.47)	3.82 e (2.91)
Nylon/salt	6.0 c (0.90)	5.6 c (1.28)	3.5 c (1.00)	2.5 c (1.00)	1.0 c (0.49)	0.9 c (0.53)	3.24 e (2.20)
Saran	8.7 b (1.27)	9.5 ab (1.34)	8.6 b (1.34)	8.2 b (1.54)	6.9 b (1.08)	6.8 b (1.27)	8.15 c (1.64)
Saran/salt	7.3 bc (1.35)	8.1 cb (0.93)	7.3 b (0.66)	7.1 b (0.84)	6.2 b (1.26)	5.2 b (0.76)	6.85 d (1.35)
SiO_2	11.2 a (1.78)	12.0 a (2.48)	11.8 a (1.93)	11.7 a (2.39)	11.8 a (1.68)	11.0 a (1.96)	11.65 a (2.08)
SiO_2/salt	9.0 b (1.47)	10.1 ab (2.32)	9.1 b (1.56)	9.6 ab (1.43)	10.0 a (1.38)	9.6 a (1.51)	9.57 b (1.69)

*All values represent the average of seven replications. Number in parentheses is the standard deviation of the replications.

**Means within the column followed by a common letter are not different ($P<0.05$).

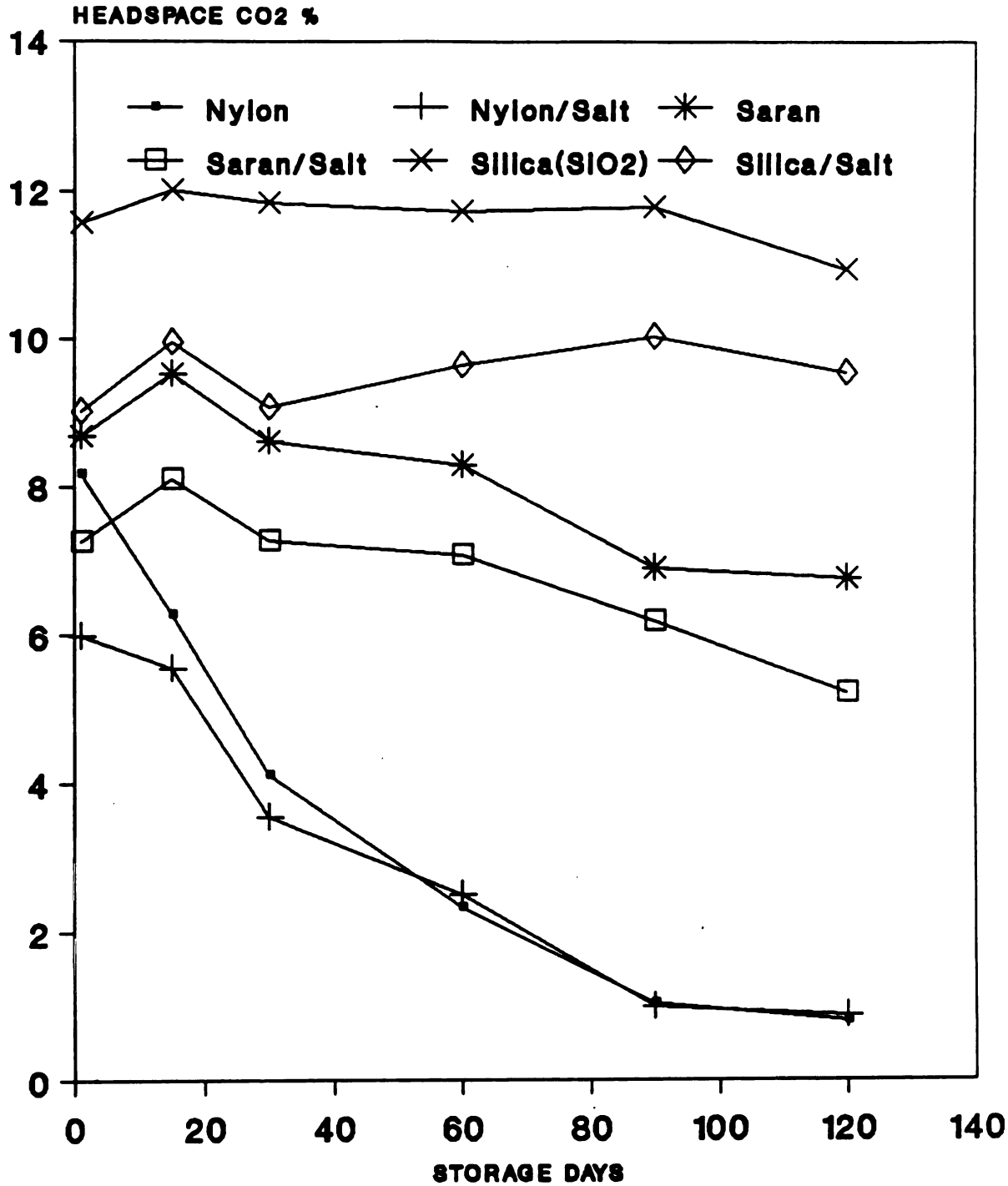


Figure 7. Effect of oxygen barrier and salt on mean package headspace CO_2 % of vacuum packaged, in-package cooked pork patties, stored at 3°C.

Effect of oxygen barrier on headspace CO_2 concentration during refrigerated storage In Table 18 & Fig. 7 are shown the headspace CO_2% of precooked, vacuum packaged pork patties in the different barriers, with/without salt, and refrigerated for up to 120 days. CO_2 declined as storage time increased, especially for the nylon barrier. The effect of oxygen barrier and storage time on mean headspace CO_2% of pork patties is given in Table 19. The amount of CO_2 in the silica barrier package did not significantly ($P>0.05$) change since it remained about 11% during the 120 days of storage. Percent CO_2 in saran samples declined slightly (8.8-6.0%) during storage and after 90 days had significantly decreased. Percent CO_2 in nylon samples declined from the first day (7.1%) to the 120th day (0.8%). Because of its polar nature, nylon is normally more sensitive to moisture and may loose barrier in high moisture environments. In this study, samples were cooked in steam and stored under refrigeration (RH= 50-60%), this may help explain why nylon samples had low headspace CO_2 percentage after cooking and storage.

Table 19. Effect of storage time and oxygen barrier on the mean headspace CO_2 percentage* of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

STORAGE DAYS	Oxygen Barriers											
	NYLON			SARAN			SiO_2			AVERAGE		
	CO_2	Std	**	CO_2	Std	**	CO_2	Std	**	CO_2	Std	**
1	7.07	1.53	a^{B}	7.97	1.5	ab^{B}	10.29	2.07	a^{A}	8.47	2.22	a
15	5.92	1.33	b^{C}	8.81	1.36	a^{B}	11.07	2.61	a^{A}	8.6	2.79	a
30	3.83	1.24	c^{C}	7.94	1.25	ab^{B}	10.45	2.23	a^{A}	7.41	3.18	b
60	2.42	0.83	d^{C}	7.69	1.38	b^{B}	10.68	2.23	a^{A}	7.04	3.74	b
90	1.01	0.51	e^{C}	6.54	1.23	c^{B}	10.89	1.77	a^{A}	6.15	4.24	c
120	0.84	0.51	e^{C}	5.95	1.28	c^{B}	10.2	1.86	a^{A}	5.54	4.08	c
AVERAGE	3.53	2.59	^{C}	7.49	1.64	^{B}	10.6	2.17	^{A}	7.2	3.62	

*All values represent the average of salt and no-salt samples. Each sample point is the result of fourteen replications. Std= Standard deviation.

**Means within the same column (subscript) or same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Effect of salt on the headspace CO_2 concentration

The presence of salt decreased headspace CO_2 in all three barrier systems (Table 20). Lower CO_2 levels indicates reduced lipid oxidation, hence they were in agreement with the TBA, hexanal, and the headspace O_2 analysis (discussed below), which all support the conclusion that salt reduces O_2 solubility (Mabrouk and Dugan, 1960) and/or reduces nonheme iron due to increased ionic strength (Ang and Young, 1989).

Table 20. Effect of salt (1.5%) on the mean headspace CO_2* percentage of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	CO_2	Std	**	CO_2	Std	**	CO_2	Std	**	CO_2	Std	**
No Salt	3.82	2.91	a	8.15	1.64	a	11.66	2.08	a	7.87	3.94	a
Salted	3.24	2.2	b	6.85	1.35	b	9.57	1.69	b	6.55	3.15	b
AVERAGE	3.53	2.59	C	7.49	1.64	B	10.6	2.17	A	7.2	3.62	

*All values represent the average of six testing days with each test day replicated seven times. Std= Standard deviation.

**Means within the same column (subscript) or the same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Correlation coefficients between TBARS values and headspace CO_2 %, and between hexanal contents and CO_2 %

Negative within-time correlation coefficients for all barriers except silica/salt were obtained between TBARS values and headspace CO_2%, and between hexanal content and CO_2% (Table 21). Except for the silica barrier, the decrease in CO_2% was highly correlated with the increase in lipid oxidation during storage. The low correlation coefficients for silica barriers was due to the low level of lipid oxidation in this barrier material. Negative within-treatment correlation coefficients were found for all barriers except for hexanal vs CO_2% at day one (Table 22). Except for hexanal vs CO_2% at day one, the decrease in CO_2% at all storage days was highly correlated with the increase

in lipid oxidation in the different barriers. The low correlation coefficients for silica barriers at day one was probably due to some missing data as was indicated earlier.

Table 21. Within-time correlation coefficients between TBARS values and headspace CO_2, and between hexanal content and headspace CO_2 of pork patties for each treatment.

	TBARS vs Headspace CO_2		Hexanal* vs Headspace CO_2	
Treatment	Correction Coeff.	Significant Level P	Correction Coeff.	Significant Level P
Nylon	-0.91	< 0.0001	-0.81	< 0.0001
Nylon/salt	-0.9	< 0.0001	-0.83	< 0.0001
Saran	-0.55	< 0.0002	-0.48	< 0.0034
Saran/salt	-0.6	< 0.0001	-0.58	< 0.0002
SiO_2	-0.37	< 0.023	-0.25	NS**
SiO_2/salt	-0.23	NS**	0.37	NS**
Pooled	-0.82	< 0.0001	-0.79	< 0.0001

* Hexanal test and CO_2 headspace test used the same sample patty, while the samples used for TBARS test were not used for CO_2 headspace test.

** No significant difference (P >0.05)

Table 22. Within-treatment correlation coefficients between TBARS values and headspace CO_2, and between hexanal content and headspace CO_2 of pork patties for each storage time.

	TBARS vs Headspace CO_2		Hexanal* vs Headspace CO_2	
Storage Days	Correction Coeff.	Significant Level P	Correction Coeff.	Significant Level P
1	-0.58	< 0.0001	-0.34	NS**
15	-0.69	< 0.0001	-0.63	< 0.0002
30	-0.83	< 0.0002	-0.64	< 0.0001
60	-0.89	< 0.0001	-0.84	< 0.0001
90	-0.92	< 0.0001	-0.89	< 0.0001
120	-0.91	< 0.0001	-0.84	< 0.0001
Pooled	-0.82	< 0.0001	-0.79	< 0.0001

* Hexanal test and CO_2 headspace test used the same sample patty, while the samples used for TBARS test were not used for CO_2 headspace test.

** No significant difference (P >0.05)

Typically, measurement of packaging headspace CO_2 in vacuum packaged cooked meats has been done to determine the influence of microbial growth on product quality (Laleye et al., 1984 a, b; Ahvenainen et al., 1989, 1990 a, b). However, in this study it has been determined that headspace CO_2% may be used as a marker to monitor lipid oxidation in different oxygen barriers during storage, if microbial growth and enzymatic activity are inhibited. The highly correlated relationship ($P<0.00001$, Table 21 & Table 22) between lipid oxidation and the loss of headspace CO_2 in the package can be the results of the following: 1. the transmission rate of CO_2 in most barriers is proportional to their O_2 transmission rate; 2. the CO_2 that was generated is the result of consumption of O_2 during lipid oxidation in the meat system; 3. the amount of CO_2 lost is always proportional to the amount of CO_2 generated from the lipid oxidation in each barriers; 4. the amount of CO_2 lost is always proportional to the amount of CO_2 generated from the lipid oxidation at any storage time; and 5. a constant CO_2 and O_2 transmission rate is existed (under same T^{o} and RH) at any storage time for each barrier. The last argument is actually confirmed by the data in Table 22 and Table 23. These findings are consistent with the ideal gas law in that the transmission rate of an ideal gas is proportional to its partial pressure in the system.

Table 23. Within-treatment correlation coefficients between TBARS values and headspace O_2, and between hexanal content and headspace O_2 of pork patties for each storage time.

	TBARS vs Headspace O_2		Hexanal* vs Headspace O_2	
Storage Days	Correction Coeff.	Significant Level P	Correction Coeff.	Significant Level P
1	--	--	--	--
15	0.76	< 0.0001	0.84	< 0.0001
30	0.76	< 0.0001	0.9	< 0.0001
60	0.63	< 0.0001	0.65	< 0.0001
90	0.68	< 0.0001	0.78	< 0.0001
120	0.5	< 0.0034	0.67	< 0.0001
Pooled	0.54	< 0.0001	0.64	< 0.0001

* Hexanal test and O_2 headspace test used the same sample patty, while the samples used for TBARS test were not used for O_2 headspace test.

Lipid oxidation and headspace O_2 concentration

Effect of oxygen barrier on headspace O_2 concentration during refrigerated storage In Table 24 & Fig. 8 are shown the headspace $\%O_2$ for the precooked, pork patties, vacuum packaged in nylon, saran, and silica barriers, with/without salt, and refrigerated for up to 120 days. Except at day one, all O_2-Storage curves were at the same level regardless of storage time. This implies that a steady state was achieved between replenishment of O_2 from atmosphere through transmission and the consumption of O_2 due to lipid oxidation (or any other reactions) that consumed oxygen in the system. The initial $\%O_2$ at day one is because the whole system has not yet reached steady state and most of the O_2 in the headspace has not yet been consumed so that $O_2\%$ at day one is still high. Also, the

in-bag pasteurization probably increased the oxygen transmission of these barrier materials and hence increased the headspace O_2.

Table 24. Effect of oxygen barrier and salt on the mean headspace O_2 percentage* of pork patties, vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	headspace O_2 % at storage day						
Treatment	1 **	15	30	60	90	120	Avg.
Nylon	3.62 $_{ab}$ (0.92)	2.79 $_{a}$ (0.40)	2.46 $_{a}$ (0.18)	2.56 $_{a}$ (0.57)	2.59 $_{a}$ (0.32)	2.33 $_{a}$ (0.13)	2.75 $_{a}$ (0.66)
Nylon/salt	4.12 $_{a}$ (1.42)	2.47 $_{a}$ (0.38)	2.27 $_{a}$ (0.50)	2.2 $_{ab}$ (0.67)	2.44 $_{ab}$ (0.50)	2.2 $_{ab}$ (0.54)	2.62 $_{a}$ (1.01)
Saran	2.61 $_{ab}$ (0.56)	1.94 $_{b}$ (0.35)	1.85 $_{b}$ (0.10)	1.99 $_{cb}$ (0.31)	2.04 $_{cb}$ (0.42)	1.99 $_{abc}$ (0.65)	2.06 $_{b}$ (0.49)
Saran/salt	2.00 $_{b}$ (0.23)	1.77 $_{bc}$ (0.27)	1.75 $_{bc}$ (0.23)	1.75 $_{cb}$ (0.24)	1.78 $_{cd}$ (0.34)	1.67 $_{dbc}$ (0.32)	1.78 $_{b}$ (0.29)
SiO_2	2.58 $_{ab}$ (0.89)	1.41 $_{c}$ (0.15)	1.37 $_{d}$ (0.10)	1.44 $_{c}$ (0.16)	1.34 $_{e}$ (0.12)	1.36 $_{d}$ (0.09)	1.59 $_{b}$ (0.59)
SiO_2/salt	3.75 $_{a}$ (1.50)	1.48 $_{bc}$ (0.25)	1.45 $_{dc}$ (0.13)	1.52 $_{c}$ (0.14)	1.51 $_{ed}$ (0.17)	1.48 $_{d\ c}$ (0.17)	1.87 $_{b}$ (1.05)

*All values represent the average of seven replications. Number in parentheses is the standard deviation of the replications.

**Means within the column followed by a common letter are not different ($P<0.05$).

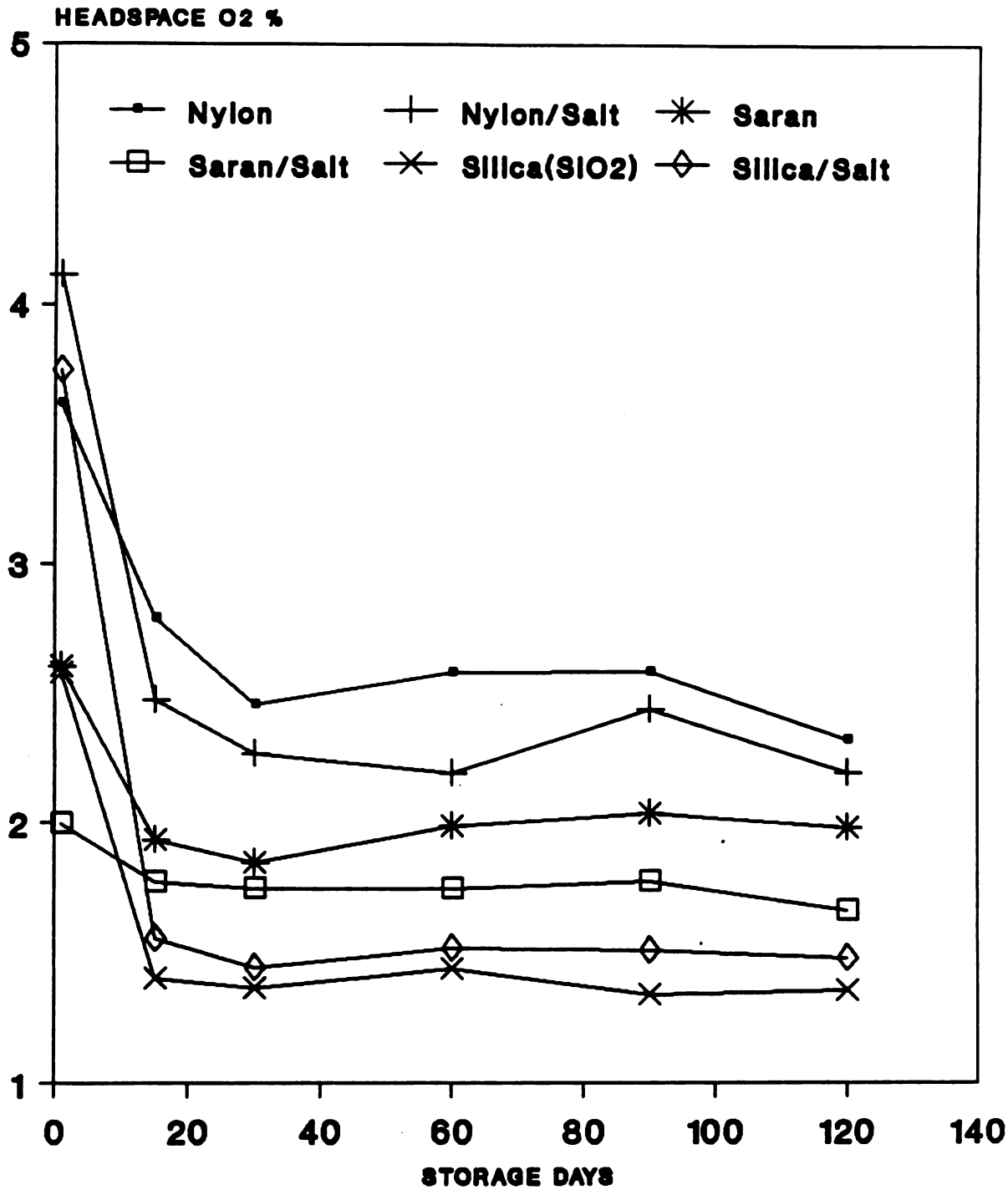


Figure 8. Effect of oxygen barrier and salt on mean package headspace O_2 % of vacuum packaged, in-package cooked pork patties, stored at 3°C.

The effect of oxygen barrier and the effect of storage time on mean headspace $\%O_2$ of the packaged pork patties are shown in Table 25. Except for day one, the headspace $O_2\%$ did not significantly change ($P<0.05$) during storage for any of the barriers and the average headspace $\%O_2$ was 2.7, 1.9, and 1.7% for nylon, saran, and silica barriers respectively. Headspace $O_2\%$ for the different barrier packaging can be distinguished from each other after the 15th day of storage. If not for the high headspace $O_2\%$ in silica samples at day one, it might be possible to tell the $\%O_2$ difference between all barriers beginning at day one. The effect of oxygen barrier on headspace $O_2\%$ (Table 25) is related to TBA analysis (Table 4) and hexanal analysis (Table 16). Generally, the first significant change in headspace $O_2\%$ occurred at day 15 for all three barriers. The headspace O_2 in the system had not reached steady state at day one (Fig. 8), which may be why headspace O_2 analysis can not detect any difference between samples packaged in different barriers. Instead, TBA and hexanal analysis were able to detect a significant difference in lipid oxidation between nylon and saran as well as nylon and silica barrier at day one.

Table 25. Effect of storage time and oxygen barrier on the mean headspace O_2 percentage* of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	O_2	Std	**	O_2	Std	**	O_2	Std	**	O_2	Std	**
1	3.87	1.22	a^{A}	2.3	0.53	ab^{B}	3.16	1.36	a^{AB}	3.02	1.25	a
15	2.63	0.42	b^{A}	1.86	0.32	a^{B}	1.44	0.22	a^{C}	1.87	0.54	a
30	2.36	0.39	c^{A}	1.79	0.19	ab^{B}	1.41	0.12	a^{C}	1.77	0.43	b
60	2.39	0.65	d^{A}	1.87	0.3	b^{B}	1.48	0.16	a^{C}	1.83	0.51	b
90	2.52	0.42	e^{A}	1.91	0.41	c^{B}	1.43	0.17	a^{C}	1.86	0.53	c
120	2.25	0.42	e^{A}	1.81	0.52	c^{B}	1.43	0.15	a^{C}	1.75	0.5	c
AVERAGE	2.68	0.86	^{A}	1.92	0.43	^{B}	1.73	0.87	^{C}	2.11	0.82	

*All values represent the average of salt and no-salt samples. Each sample point is the result of fourteen replications. Std= Standard deviation.
**Means within the same column (subscript) or same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Effect of salt on headspace O_2 concentration

Addition of salt reduced the headspace $\%O_2$ in both nylon and saran barriers, but increased the headspace $\%O_2$ in silica barriers (Table 26). There are two factors that together may explain why headspace $O_2\%$ in the SiO_2/salt samples was higher than that in the SiO_2/No-salt samples. First, the headspace $CO_2\%$ in SiO_2/No-salt samples was significantly (2%) higher than that in SiO_2/salt samples. Since headspace

CO_2 and O_2 are expressed simultaneously in relative percentages, a higher relative CO_2% results in lower O_2% in the same sample. Secondly, the silica material is such a good barrier, almost no transmission of O_2 occurs between system and environment. Any reaction (including lipid oxidation) which uses oxygen under this semiclosed system will cause an increase in CO_2% and reduce O_2%. Because SiO_2/salt samples have lower oxidation rates than SiO_2/No-salt samples in the system (Table 17 & Table 10), headspace $\%O_2$ in SiO_2/salt samples will be higher than SiO_2/No-salt samples.

Table 26. Effect of salt (1.5%) on the mean headspace O_2* percentage of pork patties vacuum packaged in nylon, saran, and silica pouches, pasteurized, refrigerated at 3°C for up to 120 days.

	Oxygen Barriers											
STORAGE DAYS	NYLON			SARAN			SiO_2			AVERAGE		
	O_2	Std	**	O_2	Std	**	O_2	Std	**	O_2	Std	**
No Salt	2.75	2.91	a	2.06	1.64	a	1.58	2.08	b	2.03	3.94	a
Salted	2.62	2.2	a	1.78	1.35	b	1.87	1.69	a	2.00	3.15	a
AVERAGE	2.68	0.86	A	1.92	0.43	B	1.73	0.87	C	2.11	0.82	

*All values represent the average of six testing days with each test day replicated seven times. Std= Standard deviation.

**Means within the same column (subscript) or the same row (capital, superscript) followed by a common letter are not significantly different ($P<0.05$).

Decreased headspace O_2 (Table 23) should lead to less lipid oxidation, which is in agreement with the results of the TBA test, hexanal analysis, and headspace CO_2. Thus, the results of this study support the conclusions that salt reduces the O_2 solubility (Mabrouk and Dugan, 1960) and/or reduced nonheme iron by increasing ionic strength (Ang and Young, 1989).

Correlation coefficients between TBARS values and headspace O_2 %, and between hexanal contents and O_2 % Low within-time correlation coefficients for all barriers were obtained between TBARS values and headspace O_2%, and between hexanal content and O_2% (Table 27). Increased %O_2 at any storage time was poorly correlated with increased lipid oxidation in the different barriers. This was because headspace O_2% stayed almost at the same level, after

Table 27. Within-time correction coefficients between TBARS values and headspace O_2, and between hexanal content and headspace O_2 of pork patties for each treatment.

	TBARS vs Headspace O_2		Hexanal* vs Headspace O_2	
Treatment	Correction Coeff.	Significant Level P**	Correction Coeff.	Significant Level P
Nylon	-0.2	NS	-0.14	NS
Nylon/salt	-0.22	NS	0.06	NS
Saran	0.19	NS	0.47	< 0.0090
Saran/salt	-0.11	NS	0.54	< 0.0018
SiO_2	0.20	NS	-0.11	NS
SiO_2/ salt	-0.24	NS	0.10	NS
Pooled	0.54	< 0.0001	0.64	< 0.0001

* Hexanal test and O_2 headspace test used the same sample patty, while the samples used for TBARS test were not used for O_2 headspace test.

** No significant difference (P >0.05)

reaching steady state (Fig. 8).

High within-treatment correlation coefficients for all storage times (except at day one) were obtained between TBARS values and headspace O_2%, and between hexanal content and O_2% (Table 23). Except at day one, the increased O_2% in the different barriers was highly correlated with the increase in lipid oxidation. At any particular storage time (except day one), the higher the headspace O_2% the higher the lipid oxidation in the pork patties.

Effect of pasteurization and subsequent storage on the tensile and seal strength of the oxygen barriers

Tensile strength of the oxygen barriers The initial tensile strengths of nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP barriers were 5764, 6838, and 9436 psi (1 psi= 6892 N/m^2), respectively. Pasteurization (82°C, 30 min.) caused a decrease, though not significantly, in the tensile strength of all 3 barriers. However, pasteurization plus refrigerated storage for 7 days significantly ($P<0.05$) reduced the tensile strength of nylon/PE and nylon/saran/Surlyn barriers in the machine direction (MD) and nylon/saran/Surlyn in the cross direction (Table 28). Pasteurization and pasteurization plus refrigerated storage did not have any significant ($P<0.05$) effect on tensile strength of PET/SiO_2-PET/CPP barriers. Nylon is hydrophilic and is more sensitive to moisture than PET or CPP (Cast Polypropylene). The results showed that nylon/PE and

nylon/saran/Surlyn lost some tensile strength due to pasteurization. Subsequent storage (3°C) after pasteurization further decreased the tensile strength of nylon/PE and nylon/saran/Surlyn barriers. This may be due to the high humidity in refrigeration, which allows moisture to penetrate into the polymers' matrix. This results in decreasing % crystallinity (Rodriguez, 1982), and hence less tensile strength.

Table 28. Effect of pasteurization & storage on mean tensile strength at break* (psi) of 3 oxygen barrier films after sealing in the machine direction (MD) and cross direction (CD).

	MD tensile Strength at Break (psi)								
Treatment	nylon			saran			SiO_2		
	Seal	Std	**	Seal	Std	**	Seal	Std	**
Control	5764	35	a	6838	505	a	9436	182	a
In-package past.***	5440	496	ab	6774	332	ab	9332	298	a
In-package Past. & Storage	4970	220	b	6162	704	b	9384	256	a
	CD tensile Strength at Break (psi)								
Control	3882	338	a	5742	255	a	8358	311	a
In-package pasteurization	3794	226	a	5612	602	a	8384	256	a
In-package Past. & Storage	3706	167	a	5096	309	b	8230	243	a

*All values represent the average of eight replications. Barrier: nylon= nylon/PE, 3.4 mil; saran= nylon/saran/Surlyn, 3.1 mil; SiO_2= PET/SiO_2-PET/CPP, 3.9 mil.

**Mans within the same column followed by a common letter are not different (P<0.05). Std= standard deviation.

***Pasteurization: 82°C, 30 min.; Pasteurized & Stored: Pasteurization at 82°C, 30 min. then stored at 3°C, RH%=85 for 7 days.

Percentage elongation of these barriers (Table 29) seemed to follow the same pattern as shown for tensile strength except that pasteurization in the saran barrier increases elongation rather than decreases it. Generally, pasteurization with refrigerated storage decreased the mechanical strength of all three barriers, especially for nylon/PE and nylon/saran/Surlyn. However, these effects were all relatively small, with most less than 15%.

Table 29. Effect of pasteurization & storage on mean tensile strength at break* (psi) of 3 oxygen barrier films after sealing in the machine direction (MD) and cross direction (CD).

	MD % Elongation								
Treatment	nylon			saran			SiO_2		
	Seal	Std	**	Seal	Std	**	Seal	Std	**
Control	502	24	a	436	20.5	b	101	9	a
In-package Past.***	480	35.5	a	491	18	a	101	15	a
In-package Past. & Storage	439	23.5	b	443	37.5	ab	104	9.5	a
	CD % Elongation								
Control	509	28	a	470	17	a	122	20	a
In-package pasteurization	515	23.5	a	488	27	a	126	23	a
In-package Past. & Storage	503	19.5	a	469	28.5	a	122	17	a

*All values represent the average of eight replications. Barrier: nylon= nylon/PE, 3.4 mil; saran= nylon/saran/Surlyn, 3.1 mil; SiO_2= PET/SiO_2-PET/CPP, 3.9 mil.

**Mans within the same column followed by a common letter are not different ($P<0.05$). Std= standard deviation.

***Pasteurization: 82°C, 30 min.; Pasteurized & Stored: Pasteurization at 82°C, 30 min. then stored at 3°C, RH%=85 for 7 days.

Seal strength of the oxygen barriers Seal strength is not a measure of seal continuity, nor can it be used to predict performance of a package. The term "seal strength" implies only the force necessary to separate the seal at the interface of the two layers of the sealant material (Martin, 1989).

The initial seal strengths of nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP barriers were 10.69, 11.33, and 23.04 lbs/in (1 lbs/in= 175 N/m), respectively (Table 30). The sealant layers in the nylon/PE, nylon/saran/Surlyn, and PET/SiO_2-PET/CPP barriers were LDPE, Surlyn, and CPP, respectively. Pasteurization significantly decreased ($P<0.05$) the seal strength of both PET/SiO_2-PET/CPP and nylon/saran/Surlyn but did not effect the seal strength of the nylon/PE barrier.

Table 30. Effect of pasteurization & storage on mean seal strength at break* (lbs/in.) of 3 oxygen barrier films after sealing in the machine direction (MD).

	MD Seal Strength at Break (lbs/in)								
Treatment	nylon			saran			SiO_2		
	Seal	Std	**	Seal	Std	**	Seal	Std	**
Control	10.69	0.24	a	11.33	0.56	a	23.04	1.9	a
In-package Past.***	10.44	0.19	a	5.48	0.35	b	19.03	1.85	b
In-package Past. & Storage	10.05	0.25	b	5.56	0.39	b	21.09	2.46	ab

*All values represent the average of eight replications. Barrier: nylon= nylon/PE, 3.4 mil; saran= nylon/saran/Surlyn, 3.1 mil; SiO_2= PET/SiO_2-PET/CPP, 3.9 mil.
**Mans within the same column followed by a common letter are not different ($P<0.05$). Std= standard deviation.
***Pasteurization: 82°C, 30 min.; Pasteurized & Stored: Pasteurization at 82°C, 30 min. then stored at 3°C, RH%=85 for 7 days.

Surlyn is a PE based polymer that is copolymerized with anionic carboxyl groups and linked by a cationic metal ion i.e. Na^{+}, Zn^{++}, K^{+}, Mg^{++} etc. between PE polymer chains (Briston, 1989). After pasteurization, the nylon/saran/Surlyn barrier became opaque (light purple) especially in the seal area. Since saran (PVDC) is very sensitive to high temperature, as temperature >75 ^{o}F, saran will cause shrinkage because the processing method that it was made (Hanlon, 1984). The shrinkage and an opaque color of saran are the evidences of loosing its crystallinity and of cause, its mechanical strength. The result is a seal strength loss in the nylon/saran/Surlyn barrier which broken starts from the saran-laminated layer. Only about 50% of the seal strength of nylon/saran/Surlyn was retained (Table 30). The percentage loss of elongation in the seal portion of nylon/saran/Surlyn barrier was even worse, with only about 7% of the elongation maintained (Table 31). Surlyn normally has excellent seal strength, puncture resistance, clarity, and toughness. The low % elongation of the saran barriers may sometimes cause a leakage problem if the package is pasteurized and then processed or handled under compression.

Table 31. Effect of pasteurization & storage on mean percentage elongation at break* of 3 oxygen barrier films after sealing in the machine direction (MD).

	MD % Elongation								
Treatment	nylon			saran			SiO_2		
	Seal	Std	**	Seal	Std	**	Seal	Std	**
Control	249	14	a	241	17	a	23.04	6.5	a
In-package pasteurization***	229	14.5	b	17	2	b	19.03	1.5	a
In-package Past. & Storage	188	11	c	14	1.5	b	21.09	5.5	a

*All values represent the average of eight replications. Barrier: nylon= nylon/PE, 3.4 mil; saran= nylon/saran/Surlyn, 3.1 mil; SiO_2= PET/SiO_2-PET/CPP, 3.9 mil.

**Mans within the same column followed by a common letter are not different ($P<0.05$). Std= standard deviation.

***Pasteurization: 82°C, 30 min.; Pasteurized & Stored: Pasteurization at 82°C, 30 min. then stored at 3°C, RH%=85 for 7 days.

Effects of pasteurization and storage on seal integrity of the oxygen barriers

Pasteurization and storage did not detrimentally effect the seal integrity of any of the three barriers. All samples survived the ARO vacuum chamber test and no sample leaks occurred under 28 in.Hg (711 mm Hg) vacuum for two minutes.

SUMMARY AND CONCLUSIONS

The effect of oxygen barrier (nylon, saran, and SiO_2), and salt (0 & 1.5%) on lipid oxidation of vacuumed, in-package pasteurized pork patties was evaluated using TBA, hexanal, and headspace (CO_2, O_2) analyses during 120 days of refrigerated storage. Correlations between TBARS values (TBARS) and hexanal (HEX), and between headspace composition (O_2 & CO_2) and lipid oxidation (TBARS & HEX) were established. Total aerobes and anaerobes were determined as a measure of the quality of samples during storage. The influence of vacuum packaging and in-package pasteurization on oxygen permeation rates, mechanical strength, and seal integrity were also investigated.

Both TBARS and HEX increased linearly (R^2=0.74 to 0.99) with storage time except for SiO_2 packaged samples which TBARS was poorly correlated. In a low lipid oxidation environment, i.e. in SiO_2 samples, hexanal analysis proved to be a better method than TBA test to monitor lipid oxidation. This is because hexanal analysis had a better linear relationship between storage time and HEX than the TBA test. The high linear relationship (in saran and nylon samples) between TBARS and storage time and between HEX and storage time, in addition to high correlation coefficients between TBARS and HEX, makes conversion between TBARS and HEX possible. The conversion factors for nylon and saran were 1.76 and 1.46 [(mg/kg)/(ppm)], respectively.

Consequently, a lipid oxidation measured as TBARS values= 1.0 was equal to HEX= 0.57 ppm in nylon sample, and equal to HEX= 0.68 ppm in saran samples.

Oxygen barrier had a highly significant ($P<0.0001$) effect on lipid oxidation of the pork patties during storage based on both TBARS and HEX. SiO_2 performed so well that TBARS and HEX were always below 0.43 (mg/kg) and 0.34 (ppm) respectively. The TBA test could not detect any difference ($P<0.05$) during 120 days of storage. Hexanal analysis began to distinguish a difference after 90 days of storage. Saran was the second best barrier in the test, and the TBARS were below 1.0 (mg/kg) after 90 days and the HEX below 0.63 ppm after 60 days of storage. Nylon was the poorest barrier, TBARS exceeded 1.0 after 15 days of storage and increased rapidly up to 4.4 at the 120th day. The better the oxygen barrier, the lower the lipid oxidation in the pork patties during refrigerated storage. The overall performance of these barriers was SiO_2 > saran >> nylon.

For all samples, stored for one day and then opened for 24 hours, TBARS increased to about 2.0. Essentially there was no advantage in using SiO_2 and saran over nylon in reducing lipid oxidation after open storage. Instead, the TBARS of SiO_2 and saran samples increased to a level close to that found in the nylon barrier sample.

Salt had a minor effect on the lipid oxidation of the pork patties during storage as measured by TBA test; however, HEX data showed that salt significantly ($P<0.05$)

reduced lipid oxidation both in saran and SiO_2 barriers during storage.

Headspace CO_2 was observed to decline as storage time increased. CO_2 levels in the poorer barrier (nylon) declined more while CO_2 in the best barrier (SiO_2) did not decline at all ($P<0.05$) during 120 days of storage. High negative within-time and within-treatment correlation coefficients for all barriers (except SiO_2) were obtained between TBARS and headspace CO_2%, and between HEX and headspace CO_2%. This indicates that the amount of CO_2 lost could be used as a marker of lipid oxidation mainly because the transmission rate of CO_2 in most barriers is proportional to their O_2 transmission rates. O_2-storage curves stayed the same for all barriers as storage time increased. High within-treatment correlation coefficients at all test times (except day one) were obtained between TBARS and headspace O_2%, and between HEX and headspace O_2%. These high correlation coefficients indicate that increased percent O_2 in the different barriers was highly correlated with increased lipid oxidation at any day except day one.

Vacuum packaging caused visible cracking on the SiO_2 barrier and increased the OTR by 3.5 times. Pasteurization (PA) increased the OTR of all three barriers, but only nylon and SiO_2 were significantly increased. PA seems to decrease, though not significantly, the tensile strength of all 3 barriers. However, pasteurization plus storage (PS) significantly reduced the tensile strength of both nylon and

saran barriers in the machine direction and saran in cross direction. PA significantly decreased the seal strength of SiO_2 (CPP sealant) and saran (Surlyn sealant) laminate but did not effect the strength of the nylon (LDPE sealant) laminate. The significant decrease of seal strength in saran barrier seems to start from the breakage of the saran laminate layer. PA and PS did not have any effect on seal integrity of the three barriers.

Based on the results from the current study, SiO_2 barrier (or barriers having the same OTR level) is recommended for premium products with 90 days or longer of required shelf-life. Vacuum packaging may cause some structural damage on the glass-like SiO_2 layer, and therefore a material with high tensile strength and O_2 barrier, i.e. PET, is a good choice to laminate with the brittle SiO_2 layer. Cost and the opaque appearance (the cracking of broken grass) caused by vacuum drawing are drawbacks of using the SiO_2 barrier.

The saran barrier (or barriers having the same OTR level) would be suitable for products with 60 days or less of required shelf-life, which is adequate for most refrigerated products. Saran normally is not recommended for cook-in-bag usage. The in-package pasteurization did not change barrier property of this nylon/saran/Surlyn lamination, but did reduce the seal strength and lower the % elongation. These may lead to a leakage problem if the package is processed or handled under compression. The

Nylon barrier is not recommended for uses in these in-package pasteurized products because of its high OTR.

BIBLIOGRAPHY

Ahvenainen, R., Skytta, E., and Kivikataja, R. L. 1989. Factors affecting the shelf-life of gas-and vacuum-packed cooked meat products: I. Sliced Ham. Lebensm. -Wiss. u. -Technol. 22:391.

Ahvenainen, R., Kivikataja, R. L., and Skytta, E. 1990a. Factors affecting the shelf-life of gas-and vacuum-packed cooked meat products: II. Vienna Sausage. Lebensm. -Wiss. u. -Technol. 23:130.

Ahvenainen, R., Kivikataja, R. L., and Skytta, E. 1990b. The influence of modified atmosphere packaging on the quality of selected ready-to-eat foods. Lebensm. -Wiss. u. -Technol. 23:139.

Allen, C. E., Beitz, D. E., Cramer, D. A. and Kauffman, R. G. 1976. Biology of fat in meat animals. North Central Regional Research Publ. No. 234.

Amundson, C. M., Sebranek, J. G., Kraft, A. A., Rust, R. E., Wagner, M. K., Gehrke, W. H. 1982. Effect of packaging film and vacuum level on regular and sorbate-cured bacon. J. Food Sci. 47:355.

Ang, C. Y., and Hamn, D. 1986. Effect of salt and sodium tripolyphosphate on shear, thiobarbituric acid, sodium, and phosphorus values of hot-stripped broiler breast meat. Poultry Sci. 65:1532.

Ang, C. Y. W., and Young, L. L. 1989. Factors relating to oxidative stability of cooked broiler breast patties treated with sodium tripolyphosphate. J. Food Sci. 54:1151.

Ang, C. Y. W., and Lyon, B. G. 1990. Evaluations of warmed-over flavor during chill storage of cooked broiler breast, thigh and skin by chemical, instrumental and sensory methods. J. Food Sci. 55:644.

Anon, 1988. Super-barriers from glass make new packages safer. Modern Plastics 1988(8):19.

Barbut, S., Maurer, A. J., and Thayer, D. W. 1987. Gamma--irradiation of clostridium botulinum inoculated turkey frankfurter formulated with different chloride salts and polyphosphates. J. Food Sci. 52:1137.

Body, D. R., and Shorland, F. B. 1974. The fatty acid composition of the main phospholipid fractions of the rumen and abomasum tissues of foetal and adult sheep. J. Sci. Food Agric. 25:197.

Bolland, J. L. 1946. Proceeding of the Royal Science, A186:218.

Bolland, J. L. and Gee, G. 1946. Kinetic studies in the chemistry of rubber and related materials: III. Thermochemistry and mechanisms of olefin oxidation. Trans. Faraday Soc. 42:244.

Briston, J. H. 1989. Ionomers. Ch. 6. In *Plastics Films*, 3rd. edition., Page 37. John Wiley and Sons Inc, New York, NY.

Brody, A. L. 1988. Silica super barrier coated films for the next generation of microwaveable food pouches. Presentation at Micro-Ready Foods '88, International Conference on Microwaveable Foods.

Brody, A. L. 1989. Modified atmosphere/vacuum packaging of Meat. Ch. 2, page 17, in Brody, A. L. ed. *Controlled/Modified Atmosphere/ Vacuum Packaging of Foods*. Food & Nutrition Press, Inc., Trumbull, CT.

Busta, F. F., Peterson, E. H., Adams, D. M., and Johnson, M. G. 1984. Colony count methods. page 63, in *Compendium of Methods for the Microbiological Examination of Foods*. American Public Health Assoc. (APHA), Washington D.C.

Careche, M., and Tejada, M. 1988. Interference by formaldehyde in the 2-thiobarbituric acid test for rancidity. J. Sci. Food Agric. 43:49.

Carr, T. P., and Marchello, J. A. 1987. Growth of aerobic psychrotrophs and color changes of precooked beef slices as affected by packaging procedure. J. Food Prot. 50:733.

Chang, I., and Watts, B. B. 1950. Some effects of salt and moisture on rancidity of fat. Food Res. 15:313.

Chen, C. C., Pearson, A. M., Gray, J. I., Fooladi, M. H. and Ku, P. K. 1984. Some factors influencing the nonheme iron content of meat and its implication in oxidation. J. Food Sci. 49:581.

Chomon, P. 1988. French cuisine cook in pack under vacuum (sous vide) at the ready-meal age. Page 117 in Future-Pak '88, Sixth International Ryder Conference on Packaging Innovation, Ryder Associates, Inc., Whippany, NJ.

Clark, D. S., and Lentz, C. P. 1969. The effect of carbon dioxide on the growth of slime-producing bacteria of fresh beef. J. Inst. Can. Sci. Technol. 2(2):72.

Corlett, Jr., D. A. 1989. Refrigerated foods and use of hazard analysis and critical control point principles. Food Technol. 43(2):91.

Coulon, M., and Louis, P. 1989. Modified atmosphere packaging of precooked foods. Ch. 8, page 135 in Brody, A. L. ed. Controlled/Modified Atmosphere/ Vacuum Packaging of Foods. Food & Nutrition Press, Inc., Trumbull, CT.

Crackel, R. L., Gray, J. I., Pearson, A. M., Booren, A. M., and Buckley, D. J. 1988. some further observations of the TBA test as an index of lipid oxidation in meats. Food Chem. 28:187.

Cross, H. R., Len, R., and Miller, M. F. 1987. Scope of warmed-over flavor and its importance to the meat industry. Page 1 in St. Angelo, A. J., and Bailey, M. E. ed. *Warmed-Over Flavor of Meat. Academic Press*, Inc., Orlando, FL.

Dawson, L. E., Uebersax, M. A., and Uebersax, K. L. 1978. Influence of antioxidants on stability of mechanically deboned turkey meat. Proc. World's Poultry Congress, Rio de Janeiro, Brazil, Sept. Page 2009.

Delaquis, P. J., Baker, R., and McCurdy, A. R. 1986. Microbiological stability of pasteurized ham subjected to a secondary treatment in retort pouches. J. Food Prot. 49(1):42.

Desrosier, N. W. and Desrosier J.N. 1977. Principles of Food Preservation by Canning. Ch. 6, Page 172. In *The Technology of Food Preservation*. 4th ed. AVI Publishing Co., Inc. Westport, CT.

Dugan, L. R. 1971. Fats. Page 133 in Price, J. F. and Schweigert, B. S. eds. *The Science of Meat and Meat Products*. 2nd ed. W. H. Freeman and Company, San Francisco, Calif.

Dupuy, H. P., Bailey, M. E., St. Angelo, A. J., Vercellotti, J. R., and Legendre, M. G. 1987. Instrumental analyses of volatiles related to warmed-over flavor of cooked meats. Page 165 in St. Angelo, A. J., and Bailey, M. E. eds. *Warmed-Over Flavor of Meat*. M. E. Academic Press, Inc., Orlando. FL.

Eklund, M. W. Peterson, M. E., Paranjpye, R., and Pelroy, G. A. 1988. Feasibility of a heat-pasteurization process for the inactivation of nonproteolytic clostridium botulinum type B and E in vacuum-packaged, hot-process (smoked) fish. J. Food Prot. 51(9):720.

El-Gharbawi, M. I. and Dugan, Jr. L. R. 1965. Stability of nitrogenous compounds and lipids during storage of freeze-dried raw beef. J. Food Sci.30:817.

Ellis, R., Currie, G. T., Thornton, F. E., Bollinger, N. C., and Gaddis, A. M. 1968. Carbonyls in oxidizing fat: II. The effect of the prooxidant activity of sodium chloride in pork tissue. J. Food Sci. 33:555.

Enfors, S. O., and Molin, G. 1984. Carbon dioxide evolution of refrigerated meat. Meat Sci. 10(3):197.

Fooladi, M. H., Pearson, A. M., Coleman, T. H. and Merkel, R. A. 1979. The role of nitrite in preventing development of warmed-over flavor. Food Chem. 4:238.

Frankel, E. N. 1980. Lipid oxidation. Prog. Lipid Res. 19:1.

Frankel, E. N., Neff, W. E., and Selke, E. 1981. Analysis of autoxidized fats by gas chromatography-mass spectrometry: VII. Volatile thermal decomposition products of pure hydroperoxides from autoxidized and photosensitized oxidized methyl oleate, linoleate, and linolenate. Lipids 16:279.

Frankel, E. N. 1982. Volatile lipid oxidation products. Prog. Lipid Res. 22:1.

Frankel, E. N. 1984. Recent advances in the chemistry of rancidity of fats. Ch. 6, page 89. In Bailey, A. J. ed. *Recent Advances in the Chemistry of Meat*. The Royal Society of Chemistry, London.

Gardner, H. W. 1980. Page 447 in Simic, M. G., and Karel, M. eds. *Autoxidation in Food and Biological System*. Plenum Press, New York, NY.

Genigeorgis, C. A. 1985. Microbial and safety implications of the use of modified atmosphere to extend the storage life of fresh meat and fish. Int. J. Food Microbiol. 1(5):237.

Gibson, T. A. and Worthington, R. E. 1977. Lipid change in frozen stored channel catfish grown by tank culture: Effects of dietary fat, freezing method, and storage temperature. J. Food Sci. 42:355.

Gill, J. L. 1978. *Design and Analysis of Experiments*. Vol. 1, Iowa State Univ. Press, Ames, IA.

Gokalp, H.Y., Ockerman, H. W., Plimpton, R. F. and Harper, J. 1983. Fatty acids of neutral and phospholipids, rancidity scores and TBA values as influenced by packaging and storage. J. Food Sci. 48:829.

Gray, J. I. 1978. Measurement of lipid oxidation: A review. J. Am. Oil Chem. Soc. 55:539.

Gray, J. I., and Weiss, G. M. 1988. Warmed-over flavor in meat. Research and Nutrition Information, National Live Stock and Meat Board. Chicago, IL.

Greene, B. E., and Price, L. G. 1975. Oxidation-induced color and flavor change in meat. J. Agric. Food Chem. 23(2):164.

Greene, B. E., and Cumuze, T. H. 1981. Relationship between TBA numbers and inexperienced panelist's assessment of oxidized flavor in cooked beef. J. Food Sci. 47:52.

Hanlon, J. F. 1984. Films and Foils. Chapter 3. Page 3-41 in *Package Engineering*. 2nd Ed. McGraw-Hill Book Co., New York, NY.

Harte, B. R. 1985. Packaging of restructured meat. Vol. 3. Restructured Meat and Poultry Products. Page 433 in Pearson, A. M., and Dutson, T. R., eds. *Advances in Meat Research*. van Nostrand Reinhold Co., New York, NY.

Hilditch, T. P., and Williams, P. N. 1964. *The Chemical Constitution of Natural Fats*. 4th ed. Chapman & Hall, London, Eng.

Hintlian, C. B., and Hotchkiss, J. H. 1986. The safety of modified atmosphere packaging: A review. Food Technol. 40(12):70.

Hornstein, I., Candrowe, P. F., and Heimberg, M. J. 1961. Fatty acid composition of meat tissue lipids. J. Food Sci. 26:581.

Hotchkiss, J. H. 1988. Experimental approaches to determining the safety of food packaged in modified atmospheres. Food Technol. 42(9):55.

Hwang, S. Y., Bowers, J. A., and Kropf. D. H. 1990. Flavor, texture, color, and hexanal and TBA values of frozen cooked beef packaged in modified atmosphere. J. Food Sci. 55:26.

Igene, J. O., King, J. A., Pearson, A. M. and Gray, J. I. 1979. Influence of heme pigments, nitrite, and non-heme iron on development of warmed-over flavor (WOF) in cooked meat. J. Agric. Food Chem. 27:838.

Igene, J. O., and Pearson, A. M. 1979. Role of phospholipids and triglycerides in warmed-over flavor development in meat model systems. J. Food Sci. 44:1285.

Igene, J. O., Pearson, A. M. and Gray, J. I. 1981. Effect of length of frozen storage, cooking and holding temperatures upon component phospholipid and the fatty acid composition of meat triglycerides and phospholipids. Food Chem. 7:289.

Igene, J. O., Yamauchi, K., Pearson, A. M., Gray, J. I., and Aust, S. D. 1985. Evaluation of 2-Thiobarbituric acid reactive substances (TBARS) in relation to warmed-over flavor (WOF) development in cooked chicken. J. Agric. Food Chem. 33:364.

Jantawat, P., and Dawson, L. E. 1980. Effects of inert gas and vacuum packaging on storage stability of mechanically deboned poultry meats. Poultry Sci. 59:1053.

Jeremiah, L. E. 1982. Influences of anatomical location and muscle quality on porcine lipid composition. Meat Sci. 7:1.

Jones, S. L., Carr, T. R., and McKeith, F. K. 1987. Palatability and storage characteristic of precooked pork roasts. J. Food Sci. 52:279.

Judge, M. D., and Aberle, E. D. 1980. Effect of prerigor processing on the oxidative rancidity of ground light and dark porcine muscle. J. Food Sci. 45:1736.

Kanner, J., and Mendel, H. 1977. Prooxidant and antioxidant effects of ascorbic acid and metal salts in a beta-carotene-linolene model system. J. Food Sci. 42:60.

Ke, P. J., Ackman, R. G., Linke, B. A., and Nash, D. M. 1977. Differential lipid oxidation in various parts of frozen mackerel. J. Food Technol. 12:37.

Kelley, G. G., and Watts, B. M. 1957. Effect of copper chelating agents on the pro-oxidant activity of ascorbic acid with unsaturated fats. Food Res. 22:308.

Kenny, M. 1962. Secondary degradation products. Page 79. In Schultz, H. W., Day, E. A., and Sinnhuber, R. O. eds. Symposium on foods: *Lipids and their oxidation*. AVI Publ., Westport, CT.

Keskinel, A., Ayres, J. C. and Synder, H. E. 1964. Determination of oxidative changes in raw meats by the 2-thiobarbituric acid method. Food Technol. 18:223.

King, A. D., and Nagel, C. W. 1967. Growth inhibition of a pseudomonas by carbon dioxide. J. Food Sci. 32:575.

Kreiger, R. A., Synder, O. P., and Pflug, I. J. 1983. Clostridium botulinum ionizing radiation D-value determination using a micro food sample system. J. Food Sci. 48:141.

Kuchmak, M., and Dugan, L. R. 1965. Composition and positional distribution of fatty acids in phospholipids isolated from pork muscle tissues. J. Am. Oil Chem. Soc. 42:45.

Labuza, T. P., and Breene, W. M. 1989. Applications of 'active packaging' for improvement of shelf-life and nutritional quality of fresh and extended shelf-life foods. (review). J. Food Process. Preserv. 13(1):1-69.

Lai, S. M. 1989. Effects of oleoresin rosemary on the oxidative quality of restructured chicken products. Master Thesis, Michigan State University, East Lansing, MI.

Laleye, L. C., Lee, B. H., Simard, R. E., Carmichael, L., and Holley, R. A. 1984a. Shelf life of vacuum- or nitrogen-packed pastrami: Effects of packaging atmospheres, temperature and duration of storage on physicochemical and sensory change. J. Food Sci. 49:832.

Laleye, L. C., Lee, B. H., Simard, R. E., Carmichael, L., and Holley, R. A. 1984b. Shelf life of vacuum- or nitrogen-packed pastrami: Effects of packaging atmospheres, temperature and duration of storage on microflora changes. J. Food Sci. 49:827.

Lea, C. H. 1939. *Rancidity in Edible Fats*. Chemical Publishing Company, New York, NY.

Lea, C. H. 1957. Deteriorative reactions involving phospholipids and lipoproteins. J. Sci. Food Agric. 8(1):1-13.

Lechowich, R. V. 1988. Microbiological challenges of refrigerated foods. Food Technol. 42(12):84.

Lin, H. S., and Sebranek, J. G. 1979. Effect of sodium nitrite concentration and packaging conditions on color stability and rancidity development in sliced bologna. J. Food Sci. 44:1451.

Lioutas, T. S. 1988. Challenges of controlled and modified atmosphere packaging: a food company's perspective. Food Technol. 42(9):78.

Love, J. D., and Pearson, A. M. 1971. Lipid oxidation in meat and meat products--A review. J. Am. Oil Chem. Soc. 48:547.

Love, J. D., and Pearson, A. M. 1974. Metmyoglobin and nonheme iron as prooxidants in cooked meat. J. Agric. Food Chem. 22:1033.

Love, J. D. 1983. The role of heme iron in the oxidation of lipid in red meats. Food Technol. 37(7):117.

Lundberg, W. O. 1962. Mechanisms. Page 31 in H. W. Schultz, E. A. Day, and R. O. Sinuhuber, eds. Symposium on food: *Lipids and their oxidation*. AVI Publ. Co., Inc., Westport, CT.

Martin, E. L. 1989. Multilayer flexible packaging. In *Encyclopedia of Packaging Technology* p. 451,

Mabrouk, A. F., and Dugan, L. R. Jr. 1960. A kinetic study of the autoxidation of methyl linoleate and linoleic acid emulsions in the presence of sodium chloride. J. Amer. Oil Chem. Soc. 37:486.

Mann, T. F., Reagan, J. O., Lillard, D. A., Campion, D. R., Lyon, C. E., and Miller, M. F. 1989. Effects of phosphate in combination with nitrite or maillard reaction products upon warmed-over flavor in precooked, restructured beef chuck roasts. J. Food Sci. 54:1431.

Manomaiwiboon, P. 1990. Effect of microbial load on vacuum packaged fresh ground beef patties subjected to "in package" thermal treatment. Master Thesis, Michigan State University, East Lansing, MI.

Marcuse, R., and Johansson, L. 1973. Studies on the TBA test for rancidity grading: II. TBA reactivity of different aldehyde classes. J. Amer. Oil Chem. Soc. 50:387.

Maron, S.H. and Lando, J.B. 1974. In Ch. 12. Solutions of Nonelectrolytes. *Fundamental of Physical Chemistry*. page 425. Macmillan Publishing Co. Inc., New York.

Martin, E. L. 1989. *Multilayer flexible packaging*. Page 451 in Encyclopedia of Packaging Technology.

Matlock, R. G., Terrell, R. N., Savell, J. W., Rhee, K. S., and Dutson, T. R. 1984a. Factors affecting properties of raw-frozen pork sausage patties made with various NaCl/phosphate combinations. J. Food Sci. 49:1363.

Matlock, R. G., Terrell, R. N., and Savell, J. W. 1984b. Factors affecting properties of precooked-frozen pork sausage patties made with various NaCl/phosphate combinations. J. Food Sci. 49:1372.

Melton, S. L. 1983. Methodology for following lipid oxidation in muscle foods. Food Technol. 37(7):105.

Miles, R. S., McKeith, F. K., Bechtel, P. J., and Novakofski, J. 1986. Effect of processing, packaging and various antioxidants on lipid oxidation of restructured pork. J. Food Protect. 49:222.

Moerck, K. E., and Ball Jr., H. R. 1974. Lipid autoxidation in mechanically deboned chicken meat. J. Food Sci. 39:876.

Mottram, D. S., Edwards, R. A., MacFie, H. J. H. 1982. a comparison of the flavor volatiles from cooked beef and pork meat systems. J. Sci. Food Agric. 33:934.

National Food Processors Association (NFPA). 1988a. Safety considerations for new generation refrigerated foods. Dairy & Food Sanitation 8(1):5.

National Food Processors Association (NFPA). 1988b. Factors to be considered in establishing good manufacturing practices for the production of refrigerated foods. Dairy & Food Sanitation 8(6):288.

Neff, W. E., Frankel, E. N., and Weisleder, D. 1982. Photosensitized oxidation of methyl linolenate, secondary products. Lipids 17:780.

Nolan, N. L., Bowers, J. A., and Kroph, D. H. 1989. Lipid oxidation and sensory analysis of cooked pork and turkey stored under modified atmospheres. J. Food Sci. 54:846.

O'Keefe, P. W., Wellington, G. H., Mattick, L. R., and Stouffer, J. R. 1968. Composition of Bovine muscle lipids at various carcass locations. J. Food Sci. 33:188.

Owen, J. E., and Lawrie, R. R. 1975. The effect of an artificially induced high pH on the susceptibility of minced porcine muscle to undergo oxidative rancidity under frozen storage. J. Food Technol. 10:169.

Palumbo, S. 1986. Is refrigeration enough to restore foodborne pathogens? J. Food Prot. 49:1003.

Pearson, A. M., Love, J. D., and Shorland, F. B. 1977. Warmed-Over Flavor in Meat, Poultry, and Fish. Vol. 23. Page 1. In C. O. Chichester, E. M. Mark and G. F. Stewart, eds. *Advances in Food Research*. Academic Press, New York, NY.

Pearson, A. M., Gray, J. I., Igene, J. O., and Yamauchi, K. 1983a. Status of warmed-over flavor research. Page 77 in Proc. Meat Ind. Res. Conf., American Meat Institute Foundation, Washington, D.C.

Pearson, A. M., Gray, J. I., Wolzak, A. M., and Horenstein, N. A. 1983b. Safety implications of oxidized lipids in muscle foods. Food Technol. 37(7):121.

Pikul, J., Leszczynski, E., and Kummerow, F. A. 1983. Elimination of sample autoxidation by butylated hydroxytoluene additions before thiobarbituric acid assay for molonaldehyde in fat from chicken meat. J. Agric. Food Chem. 31:1338.

Post, L. S., Lee, D., Solberg, M., Furgang, D., Specchio, J., and Graham, C. 1985. Development of botulinal toxin and sensory deterioration during storage of vacuum and modified atmosphere-packaged fish fillets. J. Food Sci. 50:990.

Prabhu, G. A., Molins, R. A., Kraft, A. A., Sebranek, J. G., and Walker, H. W. 1988. Effect of heat treatment and selected antimicrobials on the shelf-life and safety of cooked, vacuum-packaged, refrigerated pork chops. J. Food Sci. 53:1271.

Randall, C. H. 1988. Vacuum cooking -- the leading edge of a new frontier. Page 85 in Future-PAK '88, Sixth International Ryder Conference on Packaging Innovations, Ryder Associates, Inc., Whippany, NJ.

Rhee, K. S. 1978. Minimization of further lipid peroxidation in the distillation 2-thiobarbituric acid test of fish and meat. J. Food Sci. 43: 1776.

Rhee, K. S., Smith, G. C., and Terrell, R. N. 1983a. Effect of reduction and replacement of sodium chloride on rancidity development in raw and cooked ground pork. J. Food Prot. 46:578.

Rhee, K. S., Smith, G. C., and Rhee, K. C. 1983b. Retardation by glandless cottonseed flour of lipid oxidation and discoloration in raw ground beef containing salt. J. Food Sci. 48:351.

Rice, J. 1987. Mica-filled EVOH claim 3 to 5 times better barrier. Food Process. 1987(12):148.

Rizvi, S. S. H. 1981. Requirements for food packaged in polymeric films. CRC Crit. Rev. Fd. Sci. Nutr. 14:111.

Rodriguez, F., 1982. Physical States and Transitions. Ch. 3. In *Principles of Polymer System*. Page 33, 2nd Ed. Hemisphere Publishing Co. New York, NY.

Sacharow, S. 1988. EVOH and PVDC: rivals or allies? Prepared Foods 1988(2):82.

SAS Institute Inc. 1985. *SAS/STAT Guide for Personal Computers*. 6th ed. SAS Institute Inc., Gary, NC. 378pp.

Sato, K., and Hegarty, G. R. 1971. Warmed-over flavor in cooked meats. J. Food Sci. 36:1098.

Seiderman, S. C., Carpenter, Z. L., Smith, G. C., Dill, C. W., and Vanderzant, C. 1979a. Physical and sensory characteristics of pork packaged in various gas atmospheres. J. Food Prot. 42(4):317.

Seiderman, S. C., Carpenter, Z. L., Smith, G. C., Dill, C. W., and Vanderzant, C. 1979b. Physical and sensory characteristics of beef packaged in modified gas atmospheres. J. Food Prot. 42(3):233.

Sevanian, A., and Hochstein, P. 1985. Mechanisms and consequence of lipid peroxidation in biological systems. Ann. Rev. Nutr. 5:365.

Shahidi, F., Yun, J., Rubin, L. J., and Wood, D. F. 1987. The hexanal content as an indicator of oxidative stability and flavor acceptability in cooked ground pork. Can. Inst. Food Sci. Technol. J. 20(2):104.

Sheu, C. C. 1988. Effects of rosemary oleoresin and other antioxidants on the sensory and chemical properties of precooked pork patties. Master's Thesis, Iowa State University, Ames, IA.

Shorland, F. B. 1952. Evolution of animal fats. Nature (London) 170:924.

Siegel, D. G. 1982. Technical aspects of cooking ham in the package. Page 57, In Proc. Meat Ind. Res. Conf., American Meat Institute Foundation, Washington, D.C.

Silla, H., and Simonsen, B. 1985. Shelf-life of cured, cooked and sliced meat products. Fleischwirtsch 65(1):66.

Silliker, J. H., and Wolfe, S. K. 1980. Microbiological safety considerations in controlled atmosphere storage of meats. Food Technol. 34(3):59.

Simunovic, J., West, R. L., and Adams, J. P. 1985. Formulation of a pasteurized, restructured beef product. J. Food Sci. 50:693.

Smith, D. M., and Alvarez, V. B. 1988. Stability of vacuum cook-in-bag turkey breast rolls during refrigerated storage. J. Food Sci. 53:46.

Sofos, J. N. 1986a. Antimicrobial activity and functionality of reduced sodium chloride and potassium sorbate in uncured poultry products. J. Food Sci. 51:16.

Sofos, J. N. 1986b. Use of phosphates in low-sodium meat products. Food Technol. 40(9):52.

St. Angelo, A. J., Vercellotti, J. R., Legendre, M. G., Vinnett, C. H., Kuan, J. W., James Jr., C., and Dupuy, H. P. 1987. Chemical and instrumental analyses of warmed-over flavor in beef. J. Food Sci. 52:1163.

Stites, C. R., McKeith, F. K., Bechtel, P. J., and Carr, T. R. 1989. Palatability and storage characteristics of precooked beef roasts. J. Food Sci. 54:3.

Stoick, S. 1989. Oxidative Stability of Restructured Beef Steaks Processed with An Oleoresin Rosemary. Master's Thesis, Michigan State University, East Lansing, MI.

Tarladgis, B. G., Basil, G., Watts, B. M., and Younathan, M. T. 1960. A distillation method of the quantitative determination of malonaldehyde in rancid foods. J. Am. Oil Chem. Soc. 37:44.

Tarladgis, B. G., Pearson, A.M., and Dugan, L.R. Jr. 1964. Chemistry of The 2-Thiobarbituric acid test for determination of oxidative rancidity in foods. II. Formation of the TBA-malonaldehyde complex without acid-heat treatment. J. Sci. Food Agric. 15: 602.

Tay, H. C., Aberle, E. D., and Judge, M. D. 1983. Iron catalyzed oxidative rancidity in prerigor ground pork. J. Food Sci. 48:1328.

Terlizzi, F. M., Perdue, R. R., and Young, L. L. 1984. Processing and distributing cooked meats in flexible films. Food Technol. 38(3):67.

Tichivangana, J. Z. and Morrissey, P. A. 1985. The influence of pH on lipid oxidation in cooked meat from several species. Irish J. Food Sci. Technol. 9:99.

Tims, M. J., and Watts, B. M. 1958. Protection of cooked meats with phosphates. Food Technol. 12(5):240.

Uebersax, M. A., Dawson, L. E., and Uebersax, K. L. 1978. Evaluation of various mixing stresses on storage stability (TBA) and color of mechanically deboned turkey meat. Poultry Sci. 57:924.

Vercellotti, J. R., Kuan, J. W., Liu, R. H., Legendre, M. G., St. Angelo, A. J., and Dupuy, H. P. 1987. Analysis of volatile heteroatomic meat flavor principles by purge-and-trap/gas chromatography-mass spectrometry. J. Agric. Food Chem 35(6):1030.

Watanabe, H. 1988. The use of high barrier packaging films for extending shelf life of delicacies: Retortable and microwaveable. Page 435, In Future-Pak '88, Sixth International Ryder Conference on Packaging Innovation, Ryder Associates, Inc., Whippany, NJ.

Watts, B. 1962. Meat products. Page 202, In Schulz, H. W., Day, E. A., and Sinnhuber, R. O. eds. *Lipids and Their Oxidation in Foods*. AVI Pub. Co., Westport, CT.

Wilson, B. R., Pearson, A. M., and Shorland, F. B. 1976. Effect of total lipids and phospholipids on warmed-over flavor in red and white muscle from several species as measured by thiobarbituric acid analysis. J. Agric. Food Chem. 24:7.

Yamauchi, K., Nagai, Y., and Ohashi, T. 1980. Quantitative relationship between alpha-tocopherol and polyunsaturated fatty acid and its connection to development of oxidative rancidity in porcine skeletal muscle. Agric. Biol. Chem. 44:1061.

Yasuhara, A., and Shibamoto, T. 1989. Analysis of aldehydes and ketones in the headspace of heated pork fat. J. Food Sci. 54:1471.

Yen. J. R., Browm, R. B., Dick, R. L., Acton, J. C. 1988. Oxygen transmission rate of packaging films and light exposure effects on the color stability of vacuum-packaged dry salami. J. Food Sci. 53:1043.

Younathan, M. T., and Watts, B. M. 1960. Oxidation of tissue lipids in cooked pork. Food Res. 25:538.

Young, L. L., Reviere, R. D., and Cole, A. B. 1988. Fresh red meats: a place to apply modified atmosphere. Food Technol. 42(9):65.

Zipser, M. W., Kwon, T., and Watts, B. M. 1962. Oxidative change in cured and uncured frozen cooked pork. J Agric. Food chem. 12:105.