

Causes and Prevention of Warmed-Over Flavor

Margaret Tims Younathan*

Oxidative rancidity is a major cause of flavor deterioration in thermally treated meats. Common methods of preserving food quality, such as refrigeration, do not prevent lipid oxidation and the reaction even takes place at freezing temperatures. Off-flavors due to oxidative changes in meat are autocatalytic type reactions which produce various by-products that contribute to the rancid odors and flavors.

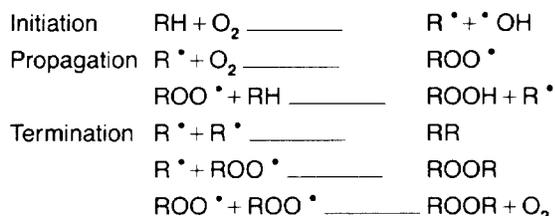
The rapid expansion of the meat processing industry in the area of precooked ready-to-eat meats has challenged the meat scientist to find methods of controlling rancidity. Growing demands of consumers for quick food service in the form of meats served through food production franchises or frozen entrees has prompted renewed interest in preventing oxidized flavor.

Several descriptive terms, such as "stale," or "rancid," have been used to characterize the oxidized flavor which develops in meats following thermal treatment, but perhaps the term "warmed-over" is one which most consumers have recognized in reheated leftover meats. The term was used by Tims and Watts (1958) to describe the rapid development of lipid oxidation in refrigerated cooked meats.

Autocatalysis of Lipids

Rancidity results in the autoxidation of meat lipid components. Unsaturated fatty acids, such as oleic, linoleic, linolenic and arachidonic, are the principal components involved in the oxidative process, (LaBuza, 1971, Keller and Kinsella, 1973). A free radical chain reaction involving three stages, namely, initiation, propagation, and termination, has been proposed to explain the autoxidation process (Uri, 1961; Lundberg 1962).

The mechanism is as follows:



RH represents the unsaturated fatty acid which reacts with

O_2 . A free radical $R \cdot$ is formed when a labile hydrogen is extracted from the carbon atom adjacent to the double bond. The free radical can react with oxygen to form a peroxy radical ($ROO \cdot$) which, in turn, may extract a hydrogen from another fatty acid, which propagates the chain reaction. Termination is achieved when two free radicals are joined together (RR), a peroxy radical ($ROO \cdot$) reacts with a free radical ($R \cdot$), or two peroxy radicals react ($ROOR$). Hydroperoxides ($ROOH$) are major initial oxidation products which decompose into hexanal, pentanal and malonaldehyde (Pearson et al., 1983), and may be responsible for rancid flavors that develop in meats (Gaddis et al., 1961).

Composition of Meat Lipids

In early studies on rancidity in meats, the research was concerned with the oxidation of adipose tissue (Watts, 1962), intermuscular lipids generally stored as large amounts of depot fat among connective tissues (Love and Pearson, 1971). When methods were perfected for measuring oxidation of lipid fractions not extracted by generally used non-polar solvents, it was possible to focus on phospholipids and proteolipids of muscle tissue. Hornstein et al., (1961) extracted tissue lipids from beef and pork and found similar amounts of phospholipids in both: Relatively large proportions of fatty acids with two, three or four double bonds were found in the phospholipid fraction (Table 1).

Table 1. Lipid Composition of Lean Meat

Lipid Fraction	Beef	Pork
Triglycerides (per cent of tissue)	2-4	5-7
Phospholipids	0.8-1.0	0.7-0.9
Fatty Acids with 2 and 3 double bonds		
Per cent of triglycerides	6.1	10.3
Per cent of phospholipids	25.2	31.9
Fatty acids with 4 or more double bonds		
Per cent of triglycerides	0.1	0.1
Per cent of phospholipids	19.2	16.3

From: Hornstein et al., (1961).

Role of Phospholipids

Muscle lipids are integral parts of the cellular structures, such as the cell wall, myofibrils, mitochondria and microsomes, and have a high degree of unsaturation, although composition can be influenced to some extent by diet (Watts, 1962). They consist primarily of phospholipids and lipoproteins which are integrated into and widely distributed throughout the muscle tissue at about a 1% level of tissue

*M.T. Younathan, School of Home Economics, Louisiana Agricultural Experiment Station, Louisiana State University Agricultural Center, Baton Rouge, Louisiana

Reciprocal Meat Conference Proceedings, Volume 38, 1985.

weight (Love and Pearson, 1971).

Protein bound phospholipids are important food constituents involved in reactions which affect food quality after storage (Lea, 1957). Their role in meat rancidity was postulated by Tims and Watts (1958). Using the 2-thiobarbituric acid (TBA) test (Turner et al., 1954), which could be performed directly on the meat tissue without the necessity of extracting the fat, meat samples were found to increase in rancidity a few hours after heating (Table 2). The TBA test can measure oxidation of highly unsaturated protein bound phospholipids which are not extractable by the ordinary fat solvents used for peroxide determinations.

Table 2. Effect of Cooking on TBA Values of Refrigerated Pork

Internal cooking temperature	Storage Intervals		
	0	2 hrs.	24 hrs.
Raw145	.109	.110
60° C372	1.70	2.47
70° C358	1.48	3.46
85° C259	2.00	3.68

From: Tims and Watts, 1958.

To explore further the role of phospholipids in rancidity of lean meat, fat extractions were done on samples of 3-day old cooked refrigerated pork. Phospho- and lipoproteins were separated from the neutral fat, then both phases tested for rancidity by the TBA method. The phospholipid and lipoproteins represented only a small proportion of the total lipid weight, yet had a TBA value approximately 30 times greater than the neutral fat portion (Table 3) (Younathan and Watts, 1960). These data led to the concept of "tissue rancidity" to differentiate it from the more familiar neutral or depot fat oxidation. Today, "warmed-over" flavor (WOF) is commonly used and is a term which elicits definite adverse flavor notes among meat scientists as well as consumers.

Table 3. Rancidity in Pork Lipid Fractions

Lipid fraction	Weight of lipid/ g tissue	TBA values/ g tissue
Total lipids272	0.65
Neutral fat246	0.20
Phospho- and proteolipids026	0.59

From: Younathan and Watts (1960).

Keller and Kinsella (1973) studied phospholipid changes and lipid oxidation in ground beef of three grades; round, chuck and hamburger (Table 4). The phospholipid content was highest in the leanest meat, i.e., ground round, suggesting that the phospholipids of meat muscles are in the bound state. Negligible amounts of phospholipids were found in drip losses, which further substantiates this fact. In analyzing the fatty acid content of phospholipids in ground round, these investigators found higher levels of arachidonic, a fatty acid with 4 double bonds, a phosphatidylethanolamine (PE) than in phosphatidylcholine (PC). With thermal treatment, arachidonic acid decreased, denoting lipid oxidation (Table 5).

Table 4. Total Lipid and Phospholipid in Three Grades of Fresh Hamburger Meat

	Total lipids (g/10g meat)	Phospholipids (mg/g meat)
Ground Round	1.038	5.46
Ground Chuck	1.852	4.69
Ground Beef	2.647	3.35

From: Keller and Kinsella (1973)

Table 5. Changes in the Fatty Acids of Phosphatidylethanolamine of Ground Round Beef During Cooking

Fatty acid	Carbon no.	Phosphatidylethanolamine	
		Fresh (moles %)	Teflon cooked
Palmitic	(C16:0)	2.9	6.7
Stearic	(C18:0)	26.2	28.8
Oleic	(C18:1)	16.6	18.4
Linoleic	(C18:2)	15.3	17.6
Linolenic	(C18:3)	—	—
Arachidonic	(C20:4)	39.0	28.5

From: Keller and Kinsella (1973).

Dugan (1971) noted an inverse relationship of phospholipid content to total lipids. As the percentage of fat declines, the proportion of tissue phospholipid increases. Fat content of the diet may vary, but the phospholipid fraction remains fairly constant, about 0.5% to 1.0% of total tissue or, as reported by Wilson et al., (1975), 0.5% for beef to 1.6% for chicken (dark meat).

The next step in delineating the role of phospholipids was to explore the effect of individual ones on WOF. Wilson et al., (1976) found that WOF became more apparent as the proportion of phospholipid to total lipids increased and suggested that the role of triglycerides in WOF development may be a minor one when compared to that of phospholipids. Igene and Pearson (1979) approached the problem by extracting total lipids, then adding back approximate composition portions to the protein residue, cooking, then testing for rancidity. Total lipids and total phospholipids significantly affected TBA numbers, although flavor rating of samples with the highest TBA numbers did not consistently have the lowest sensory scores (Table 6). A trained taste panel probably gives the

Table 6. TBA Numbers and Sensory Scores for Cooked Beef

Meat type	Composition of treatments	Mean TBA no.	Mean sensory scores ^c
Beef	B ₁ = 0.8% phospholipids	5.76	2.69
	B ₂ = 9.2% triglycerides	1.88	3.84
	B ₃ = 10% total lipids	6.81	2.64
	B ₄ = control	1.16	3.75

From: Igene and Pearson (1979).

most reliable measure of WOF while TBA indicates rancidity. Both tests should be used to follow WOF (Igene et al., 1985).

Evidence pointed to the importance of the phospholipid fraction, rather than the triglyceride fraction, as being responsible for WOF. Triglycerides, while not the sole agent, were thought to have an additive effect. In testing the influence of WOF of various phospholipid fractions, Igene and Pearson (1979) concluded that phosphatidylethanolamine (PE) is the most important phospholipid in the complex reaction which leads to WOF. Greater losses of polyunsaturated fatty acids (particularly arachidonic) were noted in PE than in PC, another observation to support the role of phospholipids in WOF. The importance of PUFAs in lipid oxidation was further emphasized in studies with meats held in frozen storage (Igene et al., 1980) when arachidonic acid (C 20:4) was found to decrease by 86% from the original amount.

Role of Heme and Nonheme Iron

Robinson (1924) first described the catalytic effect of iron porphyrins on oxidation of polyunsaturated fatty acids. Tappel (1955) demonstrated that oxidation of unsaturated fatty acids is catalyzed by heme compounds and is dependent on the presence of iron. Younathan and Watts (1959) observed that cured meats containing the pink ferrous nitric acid hemochromogen developed less rancidity than uncured cooked meats in which the pigment had been degraded to ferric denatured globin hemochromogen. They suggested that ferric iron was a more active catalyst in lipid oxidation than the ferrous form.

The activity of heme vs. nonheme iron in catalysis of lipid oxidation was the subject of several investigations in the decade which followed. Wills (1966) concluded that both heme and nonheme iron in tissues promoted oxidation of unsaturated fatty acids. Liu (1970 a, b) also reported catalytic activity of heme and nonheme iron in emulsions of linoleic acid. A nonheme iron complex (Fe^{+2} -EDTA, 1:1 ratio) accelerated oxidation at acid pH below 6.4. Metmyoglobin catalysis was more rapid and proceeded at a pH range of 5.6 to 7.8, with more activity at the more alkaline pH than acid. Chelating agents eliminated nonheme oxidation but were ineffective on heme oxidation. Using beef tissue homogenates, both heme and nonheme iron were found to increase catalysis. Ascorbic acid accelerated homogenate catalysis, pointing to the importance of nonheme iron in tissue hemogenate oxidation. Metal chelators, when added to the ascorbate-stimulated homogenate catalysis, reduced oxidation. Taking similar experiments to muscle tissue, Liu and Watts (1970) concluded that both heme and nonheme iron function in lipid oxidation. Destruction of hemes with H_2O_2 resulted in high malonaldehyde values. They noted that the destruction of heme increases the nonheme iron present, which is probably one source of nonheme iron in cooked meats. Other sources are transferrin, ferritin and various iron-containing enzymes.

Sato and Hegarty (1971) studied the role of heme and nonheme iron in meats which had been extracted with water to remove the pigment. Neither hemoglobin nor myoglobin had an effect on WOF. However, when iron salts such as $FeCl_3$ or $FeCl_2$ were added to extracted muscle, the ferrous iron seemed more important in WOF development than did

ferric iron.

Love and Pearson (1974) confirmed the observations of Sato and Hegarty (1971). Their data showed no acceleration of lipid oxidation when metmyoglobin was added to water-extracted beef muscle residues (conc. 1.0 to 10.0 mg/g). However, Fe^{2+} levels of 1.0 ppm catalyzed lipid oxidation when residues were thermally treated (Table 7).

Table 7. Effects of Various Concentrations of MetMb and Fe^{2+} on TBA Numbers of Beef Muscle Residue

<i>MetMb</i>		<i>Fe²⁺</i>	
<i>Concn. mg/g</i>	<i>TBA no.</i>	<i>Concn. ppm</i>	<i>TBA no.</i>
0	1.4	0	1.0
1.0	1.4	1.0	1.5
2.5	1.4	2.0	3.6
5.0	1.4	3.0	4.1
10.0	1.4	4.0	5.4

From: Love and Pearson (1974).

Using a meat residue remaining after meat pigment extraction, Igene et al., (1979) made further observations on the effect of heme and nonheme iron on WOF (Table 8). Their data showed that beef residues heated with total raw meat pigments gave very high TBA numbers; chelating the iron with EDTA, the value was reduced considerably. Precipitating the meat pigment by heating had little effect on TBA numbers, while chelating with EDTA lowered the lipid oxidation. Destruction of the pigment with H_2O_2 increased prooxidant activity of the extract, which could be decreased by adding EDTA. These investigators also measured the relative proportions of heme and nonheme iron in their meat pigment extracts. Heme iron accounted for approximately 90% of the total iron, nonheme, 8.7%. Heating the fresh meat pigment extract increased the nonheme iron to a level of 27.0%. They interpreted the increase to indicate that heating

Table 8. Role of Heme and Nonheme Iron on the Development of TBA Numbers in Cooked Beef

<i>treatment no.</i>	<i>preparation of experimental treatments</i>	<i>mean TBA no.</i>
1	residue + total raw meat pigments	5.00
2	residue + total raw meat pigment (chelated)	1.55
3	residue + total cooked free meat pigment	4.35
4	residue + total cooked free meat pigment (chelated)	1.46
5	residue + H_2O_2 treated total meat pigment	6.02
6	residue + H_2O_2 treated meat pigment (chelated)	1.54

From: Igene et al., (1979)

released significant amounts of bound (heme) iron to inorganic iron, which exhibited a prooxidant effect in cooked muscle tissue.

Chen et al., (1984) noted that rate of heating and finish temperature influenced the release of nonheme iron from meat pigment extracts. Temperatures in the range of 63° to 70°C were optimal; slow heating released more nonheme iron than fast heating.

Role of Nitrite

The effectiveness of sodium nitrite in preventing warmed-over flavor has been reviewed by Bailey et al., 1980, who designated it as the "most effective additive." Zipser et al., (1964) suggested that a catalytic inactive heme complex was formed with nitrite during heat treatment, thus inhibiting WOF development. Greene and Price (1975) also found that the cured meat pigment (Fe^{2+}) must be formed to achieve low TBA numbers. Igene and Pearson (1979) reported that nitrites may react with unsaturated fatty acids. Sato and Hegarty (1971) noted that nitrite can be reduced to nitric oxide, which is an effective free radical acceptor. In reviewing the action of nitrites on WOF, Pearson et al., (1977) postulated that nitrite probably complexes and stabilizes the lipids in the membrane components of muscle, or may inhibit the action of naturally-occurring prooxidants in meats.

Igene et al., (1979) heated beef tissue with and without pigment, with and without nitrite, then tested for rancidity. When meat pigments were present, nitrite decreased TBA numbers, increased panel scores (Table 9). Macdonald et al., (1980 a, b) concluded that nitrite significantly reduced rancidity in stored cooked pork and was more effective than butylated hydroxytoluene (BHT) and citric acid. In linoleic acid model systems, nitrite alone was a prooxidant, but substantially reduced oxidation in model systems containing such catalysts as Fe^{2+} or Fe^{2+} -EDTA. (Fig. 1) Similar effects were observed in an aqueous beef extract medium. These investigators agreed with Fooladi et al., (1979) that nitrite may, in some way, protect against phospholipid oxidation.

Table 9. Mean TBA Numbers and Sensory Scores in Cooked Beef

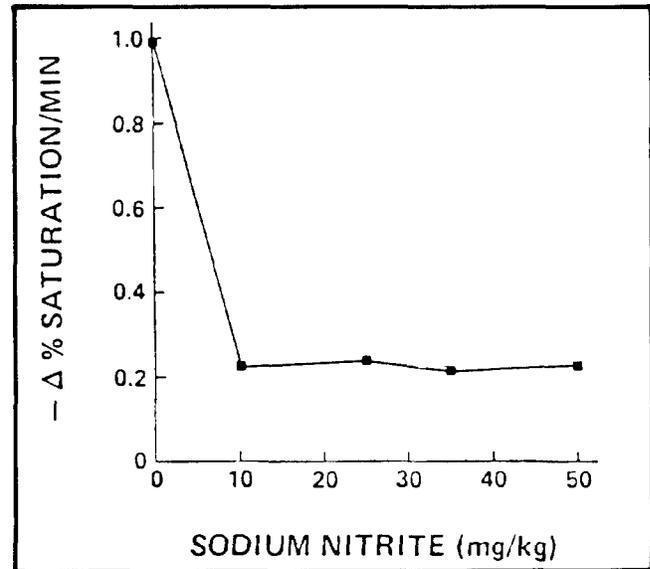
treatments	TBA no.	taste panel score
(A) cooked meat with pigment, no nitrite	1.93	2.42
(B) cooked meat with pigment plus nitrite	0.21	4.42
(C) cooked meat without pigment, no nitrite	0.61	3.50
(D) cooked meat without pigment plus nitrite	0.42	4.31

From: Igene et al., (1979)

Lipid Oxidation in Seafood and Poultry

The previous papers have dealt primarily with studies on beef or pork. WOF is as much a problem with poultry and seafoods as with large animals, such as cattle or hogs, and has been reviewed by Khayat and Schwall (1983) and Dawson and Gartner (1983). Both contain long chain fatty acids which are highly unsaturated. Heme and nonheme catalysts

Figure 1



Effects of Fe^{2+} -EDTA (1:1) and sodium nitrite on linoleate oxidation at 30°C and pH 5.6.

From: MacDonald et al., (1980)

occur in both tissues; copper may be an additional catalyst in seafood.

Thermal Treatment

Some type of heating is necessary to make meat palatable. Numerous methods have been used. Research is lacking on the effect of some of the newer heating techniques, such as use of microwave ovens and convection ovens. Johnston and Baldwin (1980) reheated 24-hour refrigerated roast beef slices by using a microwave oven and a conventional oven. No difference in TBA numbers, thiamin or riboflavin content was found between the two treatments.

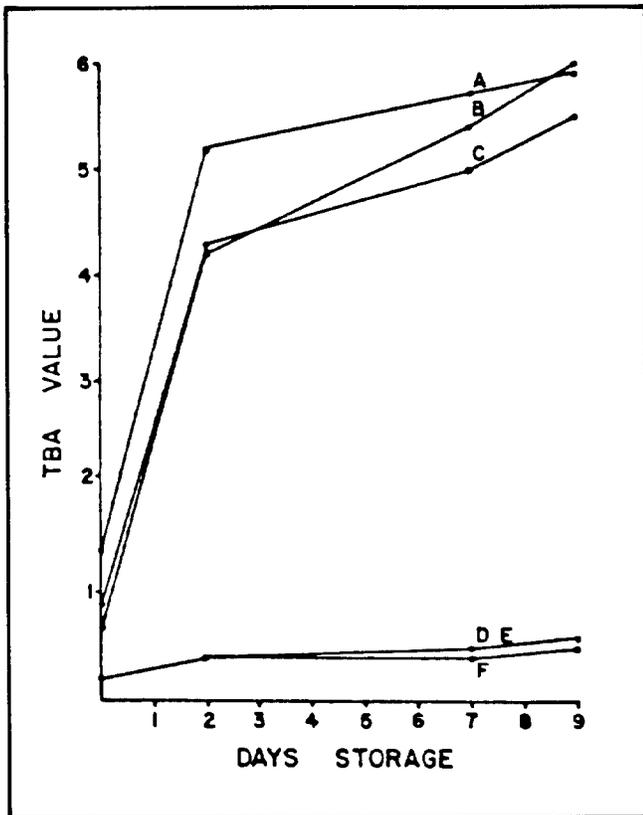
Gros (1984) studied the effect of microwave, microwave/convection combination, and oven broiling in retarding WOF in refrigerated ground beef semimembranous patties. Taste panelists did not detect flavor differences among the methods of heating: Aroma differences were found, however; Broiled patties rated highest, microwave patties rated lowest. Oven broiling also was found to reduce malonaldehyde production.

Control Measures for WOF

Phosphates have been shown to be effective inhibitors of autoxidation in cooked meats (Tims and Watts, 1958, Sato and Hegarty, 1971). Pyro-, tripoly-, and hexametaphosphate gave protection, but orthophosphates had no inhibitory properties (Fig. 2). Their role was assumed to be that of metal chelators, particularly for nonheme iron, but also possibly for copper contamination. Ascorbic acid acted synergistically with phosphates to protect against WOF. Low levels (100 ppm) catalyzed oxidation whereas high levels (1000 ppm) retarded oxidation (Sato and Hegarty, 1971).

Phenolic-type antioxidants [butylated hydroxyanisole, butylated hydroxytoluene, propyl gallate and tertiarybutylhydroquinone (TBNQ)] have been shown to improve flavor in

Figure 2



Effect of phosphates on TBA values of refrigerated cooked pork.

A: Control; B: Orthophosphate, no pH adjustment; C: Orthophosphate; D: Tripolyphosphate; E: Hexametaphosphate; F: Pyrophosphate.

beef and poultry (Van de Riet and Hard, 1979; Chang et al., 1961; Jakobsson and Bengtsson, 1972; Chen et al., 1984b). They probably would have applications to seafood.

A number of vegetable extracts have been found to be effective in preventing rancidity (Pratt and Watts, 1969, Younathan et al., 1980, 1983). They contain members of the flavonoid group, potential antioxidants due to the presence of active metal chelating sites, as well as orthodihydroxy groups on the same molecule. Plants that have been used are onions, green peppers, green pepper seeds and potato peelings. Many of these plants also contain sulphydryl groups and ascorbic acid. Oil seed proteins (glandless cottonseed, peanut and soybean) have been shown to retard oxidative rancidity in cooked refrigerated beef patties (Ziprin et al., 1981).

Summary

In summary, the available evidence seems to indicate that phospholipids are primary contributors to oxidative rancidity which develops when muscle tissues are subjected to thermal treatment. A major catalytic agent seems to be nonheme iron, although heme iron may play a role also. At present, there is no agreement in regard to the relative importance of each. Some of the most effective inhibitors are nitrites and metal chelators, such as phosphates, flavonoids and phenolic-type antioxidants. Ascorbic acid, a reducing agent, has been shown to enhance the effectiveness of many antioxidants.

References

- Bailey, M.E.; Dupuy, H.P.; Legendre, M.G. 1980. Undesirable meat flavor and its control in "The Analysis and Control of Less Desirable Flavors in Foods and Beverages" (G. Charalambous, ed.) Academic Press, New York, p. 43.
- Chang, P.Y.; Younathan, M.T.; Watts, B.M. 1961. Lipid oxidation in precooked beef preserved by refrigeration, freezing and irradiation. *Food Technol.* 15:168.
- Chen, C.C.; Pearson, A.M.; Gray, J.I.; Fooladi, M.N.; Ku, P.K. 1984a. Some factors influencing the nonheme iron contents of meat and its implication in oxidation. *J. Food Sci.* 49:581.
- Chen, C.C.; Pearson, A.M.; Gray, J.I.; Merkel, R.A. 1984b. Effects of salt and some antioxidants upon the TBA numbers of meat. *Food Chem.* 14:167.
- Dugan, L.R. 1971. Fats. In "The Science of Meat and Meat Products." J.F. Price and B.S. Schweigert (Eds.). 2nd ed., p. 113. Freeman, San Francisco, California.
- Fooladi, M.H.; Pearson, A.M.; Coleman, T.N.; Merkel, R.A. 1979. The role of nitrite in preventing development of warmed over flavor. *Food Chem.* 4:284.
- Gaddis, A.M.; Ellis, R.; Currie, G.T. 1961. Carbonyls in oxidizing fat. V. The composition of neutral volatile monocarbonyl compounds from autoxidized oleate, linoleate, linolenate esters and fats. *J. Am. Oil Chem. Soc.* 38:371-5.
- Greene, B.E.; Price, L.G. 1975. Oxidation-induced color and flavor changes in meat. *J. Agric. Food Chem.* 23:164.
- Gros, J.N. 1984. The effect of preparation on the development of warmed-over flavor in top round beef patties. M.S. thesis, Louisiana State University.
- Hornstein, I.; Crowe, P.F.; Heimberg, M.J. 1961. Fatty acid composition of meat tissue lipids. *J. Food Sci.* 26:581-6.
- Igene, J.O.; King, J.A.; Pearson, A.M.; 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.; 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.; Dugan, L.R.; Price, J.F. 1980. Role of triglycerides and phospholipids on development of rancidity in model meat systems during frozen storage. *Food Chem.* 5:263-76.
- Igene, J.O.; Yamauchi, K.; Pearson, A.M.; Gray, J.I.; Aust, S.D. 1985. Evaluation of 2-thiobarbituric acid reactive substances (TBRS) in relation to warmed-over flavor (WOF) development in cooked chicken. *J. Agric. Food Chem.* 33:364.
- Jakobsson, B.; Bengtsson, N. 1972. A quality comparison of frozen and refrigerated cooked sliced beef. 1. Influence of storage and processing variables. *J. Food Sci.* 37:230.
- Johnston, M.B.; Baldwin, R.E. 1980. Influence of microwave reheating on selected quality factors of roast beef. *J. Food Sci.* 45:1460.
- Keller, J.D.; Kinsella, J.E. 1973. Phospholipid changes and lipid oxidation during cooking and frozen storage of raw ground beef. *J. Food Sci.* 38:1200-04.
- Labuza, T.P. 1971. Kinetics of lipid oxidation in foods. *CRC Crit. Rev. Food Technol.* 2:355-8.

- Lea, C.H. 1957. Deteriorative reactions involving phospholipids and lipoproteins. *J. Sci. Food Agr.* 8:1.
- Liu, Hsiao-Ping. 1970a. Catalysts of lipid peroxidation in meats. 1. Linoleate peroxidation catalyzed by Met Mb or Fe(II)-EDTA. *J. Food Sci.* 35:590.
- Liu, Hsiao-Ping. 1970b. Catalysts of lipid peroxidation in meats. Linoleate oxidation catalyzed by tissue homogenates. *J. Food Sci.* 35:593.
- Liu, Hsiao-Ping; Watts, B.M. 1970. Catalysts of lipid peroxidation in meats. 3. Catalysts of oxidative rancidity in meats. *J. Food Sci.* 35:596.
- Love, J.D.; Pearson, A.M. 1971. Lipid oxidation in meat and meat products – a review. *J. Am. Oil Chem. Soc.* 48:547.
- Love, J.D.; Pearson, A.M. 1974. Metmyoglobin and nonheme iron as prooxidants in cooked meat. *J. Agric. Food Chem.* 22:1032.
- Lundberg, W.O. 1962. Mechanisms. In "Symposium on Foods: Lipids and their Oxidation," H.W. Schultz, E.A. Day, and R.O. Sinnhuber (Eds.) p. 33. AVI Publishing Co., Westport, CT.
- Macdonald, B.; Gray, J.I.; Gibbins, L.N. 1980a. Role of nitrite in cured meat flavor: antioxidant role of nitrite. *J. Food Sci.* 45:893.
- Macdonald, B.; Gray, J.I.; Kakuda, Y.; Lee, M.L. 1980b. Role of nitrite in cured meat flavor: chemical analysis. *J. Food Sci.* 45:889.
- Pearson, A.M.; Gray, J.I.; Wolyak, A.M.; Horenstein, N.A. 1983. Safety implications of oxidized lipids in muscle foods. *Food Technol.* 37 (7):121-9.
- Pearson, A.M.; Love, J.D.; Schorland, F.B. 1977. "Warmed-over" flavor in meat, poultry and fish. In "Advances in Food Research." Vol. 23 pp. 2-74. Academic Press, New York.
- Pratt, D.E.; Watts, B.M. 1964. The antioxidant activity of vegetable extracts. 1. Flavone aglycones. *J. Food Sci.* 29:27.
- Robinson, M.E. 1924. Haemoglobin and methaemoglobin as oxidative catalysts. *Biochem. J.* 18:225-264.
- Sato, K.; Hegarty, G.R. 1971. Warmed over flavor in cooked meats. *J. Food Sci.* 36:1098.
- Tappel, A.L. 1955. Unsaturated lipid oxidation catalyzed by heme compounds. *J. Biol. Chem.* 217:721.
- Tims, M.J.; Watts, B.M. 1958. Protection of cooked meats with phosphates. *Food Technol.* 12:240-3.
- Turner, E.W.; Poynter, W.D.; Montic, E.J.; Bessert, M.W.; Struck, G.M.; Olson, F.C. 1954. Use of the 2-thiobarbituric acid reagent to measure rancidity in frozen pork. *Food Technol.* 8:326.
- Uri, N. 1961. Physico-chemical aspects of autoxidation. In "Autoxidation and Antioxidants," W.O. Lundberg (Ed.) Vol. 1, p. 55. Interscience Publishers, New York.
- Van de Riet, Shirley; Hard, Margaret M. 1979. Flavor quality of antioxidant treated cooked ground beef patties. *J. Am. Dietet. Assoc.* 75:556.
- Watts, B.M. 1962. Meat Products. In "Symposium on Foods: Lipids and Their Oxidation," H.W. Schultz, E.A. Day, and R.O. Sinnhuber (Eds.) p. 202. AVI Publishing Co. Westport, CT.
- Wills, E.D. 1966. Mechanisms of lipid peroxide formation in animal tissues. *Biochem. J.* 99:667.
- Wilson, B.R.; Pearson, A.M.; Sharland, F.B. 1976. Effect of total lipids and phospholipids on warmed-over flavor in red and white muscles from several species as measured by TBA analysis. *J. Agr. Food Chem.* 24:7.
- Younathan, M.T.; Marjan, Z.M.; Arshad, F.B. 1980. Oxidative rancidity in stored ground turkey and beef. *J. Food Sci.* 45:274.
- Younathan, M.T.; Oon, J.K.; Yusof, R.B.M. 1983. Control of heat induced oxidative rancidity in refrigerated shark and mackerel. *J. Food Sci.* 48:176.
- Younathan, M.T.; Watts, B.M. 1959. Relationships of meat pigments to lipid oxidation. *Food Research* 24:728-734.
- Zipser, M.W.; Kiwon, T.W.; Watts, B.M. 1964. Oxidative changes in cured and uncured frozen cooked pork. *J. Agric. Food Chem.* 12:105.
- Ziprin, Y.A.; Rhee, K.S.; Carpenter, Z.L.; Hosteller, R.L.; Terrell, R.N.; Rhee, K.C. 1981. Glandless cottonseed, peanut and soy protein ingredients in ground beef patties: effect on rancidity and other quality factors. *J. Food Sci.* 46:58.

Discussion

Ing Peng: I am interested in your comments and thoughts on two questions. 1) Can warmed-over flavor also occur in fresh meat, and what is the importance of several enzymatic systems in the development of warmed-over flavor? 2) I'd like to know your thoughts on the possible involvement of singlet oxygen in the development of rancidity.

M.T. Younathan: I would like to comment on the first question. Of course, warmed-over flavor was originally the term introduced to indicate heat-induced lipid oxidation, but the same odor seems to develop in raw meat tissue, and there is a possibility that it may be related to the oxidation of the meat pigments. One of the studies on this has been by Green and Price, at the University of Georgia and also at Oregon State. They have presented information in this regard. There are some lipid peroxidases among the enzymatic systems present in muscle tissue and possibly, before the heat treatment, they would have an effect on development of off-odors and flavors in meats. I am sorry I am not knowledgeable about the singlet oxygen contribution and I cannot comment on that.

Joe Regenstein: I just wanted to make a comment on ascorbic acid in seafood, since you brought up poultry and seafood. One does have to be quite careful with ascorbic acid with many species of fish. It leads to some of the frozen-storage textural changes, that some of us are involved in preventing, so it may well work as an antioxidant, but is not a compound one should indiscriminately throw into fish. There

are proprietary materials out there which include ascorbate, and one does have to be quite careful with them.

Younathan: Yes, there are some texture changes that can take place in fish with the use of ascorbic acid. I think ascorbic acid, in conjunction with some of the phenolic antioxidants, will have more application to beef and pork than to seafood.

Bob Benedict: A comment on the former thing about grass-fed vs grain-fed. The grass-fed animals will have lower fat which would be recommended for the 21st century lower-fat diet. However, when one calculates the amount of polyunsaturated fats (fats containing linoleic, linolenic, arachidonic acid, etc.), as a percentage of total fat in the grass-fed, it's significantly higher in the phospholipid fraction. This would indicate that such meat would have a potential problem with the production of oxidized lipids, so although you are trading the benefit of a better nutritional factor with the lower calories, lower fat content of the grass-fed animals would be a potential problem with off-flavor.

Younathan: Thank you for that comment. It seems that as we move into the meats with less fat in them, we will be more involved in the warmed-over flavor problem.

Kevin Jones: I was wondering what influence pH would have on lipid oxidation, particularly when you get to lower pH products that have, perhaps, been fermented, but without nitrite.

Younathan: As you get into lower pH's, I think that there might be more effect of the non-heme iron on lipid oxidation.

John Forrest: In your list of mechanisms for controlling lipid oxidation, you mentioned several that are of the non-natural nature, and some that are natural. Because of the emphasis that Mike gave us on the content of natural materials in products, which of those natural components would you see that might have the most potential for helping control warmed-over flavor?

Younathan: I feel that of the natural products, probably the onions would have the most effect. Most people will accept onions in a meat dish, and the onions have sulfhydryl compounds and also a small amount of ascorbic acid, so I think the onions would be the best product to choose.

John Bursalatti: You, of course, were one of the original definers of the term "warmed-over flavor" and I think you have made a magnificent contribution to the whole area of meat science and processed meats. From your long experience, could you share with us what you consider to be the earmarks and the deteriorative characteristics of the warmed-over flavor on commercially-used meat products?

Younathan: On commercially-used products, I think the

main problem is the fact that they are stored, and, as we have shown, the reaction takes place within just a few hours. It takes place at freezing temperatures, but probably not at the same rate as at refrigerated temperatures. But it does need to be controlled and, as we have seen, the use of phosphates, the use of some of the phenolic-type antioxidants in the natural food products seems to be the route to go to produce pre-cooked meat for the market that does have an acceptable flavor and odor.

Gene Allen: I am trying to resolve in my mind if low-temperature cookery increases oxidative rancidity and the potential for warmed-over flavor. Why do we have a very successful industry in terms of pre-cooked beef, which goes to hotels and restaurants, that is initially cooked by long-time low-temperature cookery and then warmed up and served in the restaurant? What is the answer to this?

Younathan: Packaging may be one factor here that would prevent it. It may be packed in such a way that the oxygen is excluded, and there is little oxidation taking place. And also, perhaps the meat scientist is more aware of warmed-over flavor, the general public may not be as aware as those of us who work in meat every day.