This is to certify that the

thesis entitled

EFFECTS OF OLEORESIN ROSEMARY ON THE OXIDATIVE QUALITY OF RESTRUCTURED CHICKEN PRODUCTS

presented by

SHU-MEI LAI

has been accepted towards fulfillment
of the requirements for

M.S. degree in FOOD SCIENCE

J. I. Gray
Major professor

Date 9-08-89

EFFECTS OF OLEORESIN ROSEMARY ON THE OXIDATIVE QUALITY OF RESTRUCTURED CHICKEN PRODUCTS

By

Shu-Mei Lai

A THESIS

Submitted to
Michigan State University
in partial fulfilment of the requirements
for the degree of

MASTER OF SCIENCE

Department of Food Science and Human Nutrition

1989

ABSTRACT

EFFECTS OF OLEORESIN ROSEMARY ON THE OXIDATIVE QUALITY OF RESTRUCTURED CHICKEN PRODUCTS

By

Shu-Mei Lai

The effects of oleoresin rosemary (OR), sodium tripolyphosphate (STPP) and TBHQ on the stability of lipids in restructured chicken nuggets were investigated. Lipid oxidation occurring during refrigerated and frozen storage was monitored by a modified thiobarbituric acid (TBA) test, sensory evaluation and hexanal analysis.

TBA-reactive substances (TBARS) values and sensory scores demonstrated that STPP/TBHQ exhibited the best protective effect. An additive antioxidant effect was obtained from the combination of OR and STPP, while no significant difference was found between STPP/OR and STPP/TBHQ. STPP/OR and STPP/TBHQ also effectively prevented loss of polyunsaturated fatty acids and formation of hexanal during frozen storage. Significant correlation coefficients between TBARS values and sensory scores were obtained in the refrigerated and frozen studies. Hexanal contents correlated better with sensory scores than with TBARS values in chicken nuggets after six months of frozen storage.

ACKNOWLEDGMENT

I wish to express my sincere gratitude to Dr. J. Ian Gray for his guidance and support as major professor, and for his encouragement throughout my graduate studies.

Appreciation is also extended to Dr. A.M. Booren, Dr. B. R. Harte, Dr. C.J. Flegal, and Dr. M.J. Zabik for serving on my guidance committee, and for their critical review of this manuscript. Further acknowledgement is due Dr. D.M. Smith and Ms. R.L. Crackel for their technical advice and assistance during the research program.

Special thanks are extended to Mr. Robert Evans and Ms Ariella Gastel of Kalsec Inc. for their interest in and support of my studies, and for supplying the oleoresin rosemary used in the study. I also would like to thank Diamond Crystal Salt Company for donating the antioxidant-coated salt used in the study.

In addition, I extend my appreciation to my friends and colleagues at Michigan State University who have offered their loving help and beyond understanding words of encouragement.

Finally, I wish to thank my parents for their support and understanding which enabled me to come to the United States to further my education.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

The term "warmed-over flavor" (WOF) describes an oxidized or rancid flavor which develops rapidly during the refrigerated or frozen storage of precooked or partially cooked meat products (Pearson and Gray, 1983). WOF is a major cause of quality deterioration in precooked meat products and limits the acceptability of these products (Love, 1983).

In recent years, mechanically deboned poultry meat (MDPM) has been utilized in the processing of precooked, restructured poultry products for adding value and providing uniform texture to the products. However, the stress and aeration to which MDPM is subjected during mechanical deboning and the compositional nature (bone marrow, heme and lipids) of MDPM contribute to its high oxidative potential (Dawson and Gartner, 1983). The inclusion of MDPM in processed products can further induce the oxidative deterioration of lipids and result in decreased product shelf life (McNeill et al., 1988).

Several synthetic phenolic antioxidants have been used successfully to prevent WOF development in restructured meat products (Chastain et al., 1982; Crackel et al., 1988a). However, consumers are becoming increasingly concerned about

the use of synthetic chemicals and are interested in the use of natural products. Oleoresin rosemary, a naturally occurring product, contains a number of compounds such as carnosol, rosmanol, rosmaridiphenol and rosmariquinone which possess antioxidant activity similar to or greater than butylated hydroxyanisole (BHA) (Houlihan and Ho, 1985).

Barbut et al. (1985) demonstrated that the incorporation of oleoresin rosemary in turkey breakfast sausages prepared from a combination of hand deboned and mechanically deboned turkey meat, can substantially suppress lipid oxidation and increase product shelf life at refrigerated temperatures. Korczak et al. (1988) also found a pronounced antioxidant effect of rosemary in precooked minced meat products.

The primary objective of this study was to assess the effectiveness of oleoresin rosemary as an antioxidant during refrigerated and frozen storage of chicken nuggets. A commercial oleoresin rosemary, tested at two levels with or without sodium tripolyphosphate, and TBHQ were evaluated. A secondary objective was to determine the correlation between thiobarbituric acid-reactive subatances (TBARS), sensory scores and hexanal contents as a means of assessing the efficacy of these procedures in following lipid oxidation in meat systems. Finally, the antioxidant activity of oleoresin rosemary in soybean oil during continuous heating was investigated.

REVIEW OF LITERATURE

Lipid oxidation in meat

Lipids are important to the physical and chemical characteristics of meat and poultry products. They contribute many desirable sensory properties to these products by enhancing their flavor and aroma (Herz and Chang, 1970) and by increasing their tenderness and juiciness (Blumer, 1963; Pearson, 1966). However, the potential for lipid oxidation or the development of rancid or warmed-over flavor (WOF) is one of the single greatest constraints in determining the processing and shelf-life characteristics of meat products. The nature, proportion, and degree of unsaturation of fatty acids present in a lipid system or food will indicate the approximate susceptibility of that product to oxidative deterioration.

Lipids in Muscle Tissues

Animal lipids are composed chiefly of neutral lipids and phospholipids. Phospholipids are present in small amounts but are essential to muscle because of their role as structural and functional components of cells and membranes. Unlike phospholipids which are mainly membrane-associated, neutral lipids are present as microscopic droplets within

the muscle cell, or in adipocytes (fat cells) primarily located within the perimysial connective tissue of the muscle. They provide fatty acids for energy metabolism in living muscle and contribute to the characteristic of the meat (Moody and Cassens, 1968).

Neutral lipids are principally triacylglycerols which are fatty acids esters. The fatty acids in the triacylglycerols may be the same or different at one, two, or all three positions on the glycerol molecule. Triacylglycerols mainly contain C_{14}, C_{16} and C_{18} fatty acids, which generally are either saturated or contain only one or two double bonds (Igene et al., 1981). Oleic acid ($C_{18:1}$) is the most abundant and widespread unsaturated fatty acid, whereas palmitic acid ($C_{16:0}$) is the most prominent saturated fatty acid in triacylglycerols. The polyunsaturated fatty acids are mostly confined to linoleic acid ($C_{18:2}$) with a lesser amount of linolenic acid and small quantities (< 1 %) of arachidonic acid (Pearson et al., 1977). Since fatty acids differ both in chain length and in degree of saturation, triacylglycerols vary greatly in composition and in their susceptibility to autoxidation.

Phospholipids normally occur in meat as phosphoglycerides. They generally comprise about 0.5 -1.0 % of the tissue content and contain not only $C_{18:2}$ and $C_{18:3}$ in significant amounts but also high concentrations of $C_{20:3}$ and other highly polyunsaturated fatty acids including $C_{20:4}$, $C_{22:4}$, $C_{22:5}$ and $C_{22:6}$ (Pearson et al., 1977; Igene

et al., 1981; Pikul et al., 1984b). The degree of unsaturation of the phospholipid fraction is about 15-fold higher than that of the triacylglycerol fraction (Igene and Pearson, 1979; Igene et al., 1981)

According to Allen and Foegeding (1981), the phospholipid content of meat varies inversely with total lipid content, increasing from 5 to 70% as the fat content decreases from 13 to 1% (Table 1). This reflects the constancy of the phospholipid fraction even though the total lipid content is highly variable.

Characteristics of Poultry Meat Lipids

(1) Lipid Composition

Even though animal lipids tend to be more uniform in composition than those from plants, they are still highly variable. The content and composition of muscle lipid differ among various species as well as within an animal depending on the muscle function and location (Gray and Macfarlane 1961; Dugan 1987). Allen and Foegeding (1981) summarized the lipid composition of different types of muscle from several species of animals (Table 1). These data illustrate that chicken and turkey have less total lipid but a higher proportion of phospholipid than fish, beef and pork. White meat of chicken and turkey also contains smaller amounts of total lipid compared to dark meat from the same species. In general, more aerobic metabolism of red or "dark" muscle compared to white or

"light" muscle is associated not only with high myoglobin concentration, but also with higher lipid concentration (Igene et al., 1979b). Wilson et al.(1976) concluded that the red oxidative muscles have more phospholipid than white glycolytic muscles taken from the same species. However, the proportion of phospholipids in the lipids of white poultry meat is greater than in dark meat (Katz et al., 1966; Wangen et al., 1971). On a weight-percentage basis, the white and dark meat of chicken contain about the same amount of phospholipid (Katz et al., 1966).

Table 1. Lipid composition of different muscle types[1].

Species	Muscle Type	Content		
		Lipid(%)	Neutral lipid(%)	Phospholipids
Chicken	White	1.0	52	48
	Dark	2.5	79	21
Turkey	White	1.0	29	71
	Dark	3.5	74	26
Fish(Sucker)	White	1.5	76	24
	Dark	6.2	93	7
Beef	Longissimus dorsi	12.7	95	5
Pork	L. dorsi	4.6	79	21

[1] Adapted from Allen and Foegeding (1981)

In addition, the lipid composition of mechanically deboned poultry meat (MDPM) also varies from that of hand deboned poultry meat (HDPM). Moerck and Ball (1974) reported that the total lipid content of mechanically

deboned chicken was 27% and was composed of approximately 98.6% neutral lipids and only 1.4% phospholipids.

The lipid content of MDPM is much higher than HDPM due to the stripping of fat from the skin during the deboning process (Janky and Froning, 1975). Table 2 shows the proximate composition of MDPM, and indicates considerable variability in fat composition. This variability is due to the variations in the initial source of meat and degree of skin and meat removal before deboning (Dawson and Gartner, 1983).

Table 2. Proximate composition of mechanically deboned poultry meat[1].

Poultry Source	Protein (%)	Fat (%)	Moisture (%)
Turkey Frame	14	16	68
Chicken Back, Neck[2]	14.5	17.6	66.6
Spent Layers	14.2	26.2	60.1

[1] Adapted from Dawson and Gartner (1983)
[2] Without skin

Grunden et al. (1972) determined the lipid content of MDPM from broiler backs and necks (with skin), spent layers as well as turkey racks. They found mechanically deboned turkey meat contained significantly less lipid (12.7%) than did meat from broiler backs and necks (27.2%) or spent layers (26.2%). This difference exists because the turkey

backs contained proportionally less skin than either the spent layer or broiler backs and necks. On the other hand, there were no significant differences in total lipids in mechanically deboned broiler meat and spent layers. With backs and necks there was a high percentage of skin which resulted in a higher fat percentage while the skin and heavy fat deposits of whole spent layer were diluted by the large percentage of meat on the whole carcass (Grunden et al., 1972).

(2) Fatty Acid Composition

Not only does the lipid composition vary with species and muscle type, but also the fatty acid composition in triacylglycerols and phospholipids varies according to species and muscle type.

Phospholipids are known to be more susceptible to oxidative deterioration than triacylglycerols because of their larger proportion of unsaturated fatty acids (Gray and Pearson, 1987). In lean chicken meat, about 41-46% of the fatty acids from phospholipids contain two or more double bonds compared to 22-24% in the triacylglycerols. Furthermore, phospholipids contain 24-30% of polyunsaturated fatty acids ($C_{18:3}$ to $C_{22:6}$) while very low levels (<2%) were found in the triacylglycerol fraction (Igene et al., 1981; Yamauchi et al., 1982; Pikul et al., 1984b; Wu and Sheldon, 1988). The phospholipids of chicken also contain a higher percentage (27%) of fatty acids with four or more

double bonds than do those (8% to 16%) of pork, beef and lamb (Allen and Foegeding, 1981).

Marion and Woodroof (1966), Katz et al. (1966), Moerck and Ball (1974) ,and Jantawat and Dawson (1980) analyzed the fatty acid distribution profiles of lipids from white meat, dark meat, skin and mechanically deboned meat from chicken and turkey. They reported that the neutral lipid fraction was composed of $C_{16:0}$, $C_{16:1}$, $C_{18:0}$, $C_{18:1}$ and $C_{18:2}$, and that there were no apparent variations in the triacylglycerol fatty acids among the four types of tissues. They also found that the phospholipid fraction for all the tissues analyzed contained high levels of $C_{16:0}$, $C_{18:0}$, $C_{18:1}$, $C_{18:2}$ and $C_{20:4}$ fatty acids. In addition, high levels of polyunsaturated 20 to 24 carbon fatty acids were also present in the phospholipids from most of the tissues in both turkey and chicken. Jantawat and Dawson (1980) reported the most obvious differences among phospholipids from different types of tissues are the quantities of $C_{20:4}$ and the levels of total polyunsaturated 20 to 24 carbon fatty acids. These groups of acids were lowest in chicken and turkey skin tissues. On the other hand, the quantities of pentaenes and hexanenes were highest in phospholipids of hand deboned tissues (Jantawat and Dawson, 1980).

(3) Phospholipid Component

Although the proportions of the individual phospholipids are relatively constant in fresh meat, there

is some variation in the proportion of the phospholipid components between species and between muscles within the same species. The major components of phospholipids from animal tissues include approximately 50% phosphatidyl choline (PC), 25% phosphatidyl ethanolamine (PE), and 5% sphingomyelin (SP), with lesser amounts of phosphatidyl serine (PS), phosphatidyl inositol, and other minor components (Pearson et al., 1977; Allen and Foegeding, 1981).

It has been reported that the white meat of chicken has more PC (60%) and less PE (15%), and both the dark and white meat of the turkey have higher SP (12%) than muscle from the other species (Peng and Dugan, 1965; Wangen et al., 1971; Allen and Foegeding , 1981). However, these results contradict the findings of Jantawat and Dawson (1980) who reported no significant difference in the proportion of PC and SP between dark and white meat of hand deboned poultry or among different species. On the other hand, the MDPM contained with higher levels of SP than HDPM, and this was corroborated by the finding that higher amounts of SP were present in the skin and bone tissue than from in poultry meat (Jantawat and Dawson, 1980).

Warmed-over Flavor Development in Meats

The term "warmed-over flavor (WOF)" was first introduced by Tims and Watts (1958) to describe the rapid onset of rancidity in refrigerated cooked meat. The rancid

or stale flavor becomes apparent within 48 hours in contrast to the more slowly developing rancidity which results from long term storage of frozen raw meat (Gray and Pearson, 1983). Although WOF is normally associated with cooked meat, it also develops rapidly in raw meat that has been ground and exposed to air (Greene, 1969; Sato and Hegarty, 1971; Benedict et al., 1975). Thus, WOF is the major cause of quality deterioration in precooked and/or restructured products and limits the acceptability of these products (Love, 1983).

Many studies over the past two decades have focused on mechanisms reponsible for development of WOF in meat systems. Many reports support the hypothesis that WOF is a result of lipid oxidation which is catalyzed nonenzymatically by metal ions (Love, 1983). The polyunsaturated fatty acids in phospholipids appear to be the primary substrate for the formation of oxidation products that contribute to WOF (Wilson et al., 1976; Pearson et al., 1977; Igene et al., 1985). Nevertheless, the precise mechanisms involving the formation of WOF have not been resolved.

The discussion in this section will focus on the role of lipids in oxidation with special emphasis on the phospholipids as well as membrane-bound lipids of poultry meat. The role of heme and nonheme iron also will be discussed.

(1) Role of Phospholipids and Triacylglycerols

Poultry meat is known to be more susceptible to oxidative rancidity than red meat due to the relatively high content of phospholipids (Wilson et al., 1976; Ang, 1988). Convincing evidence for the involvement of the phospholipids in oxidized off-flavor or WOF has been reported by several researchers. Acosta et al. (1966) isolated phospholipids from cooked turkey and found that this fraction consumed oxygen at a faster rate than non-phospholipid lipid fractions. Wilson et al. (1976) reported that phospholipids were the major contributor to WOF in turkey, chicken, beef and lamb.

Further studies by Igene and Pearson (1979) and Igene et al. (1981) involving model systems from beef and from chicken dark and white meat, demonstrated that both triacylglycerols and phospholipids contributed to WOF development, and that phospholipids made the greatest contribution. The triacylglycerols enhanced development of WOF only when combined with the phospholipids as total lipids. Pikul et al. (1984b) claimed that phospholipids contributed approximately 90% of the malonaldehyde measured in total lipid fractions from chicken meat. In addition, they pointed out that fat from chicken breast contained two times more malonaldehyde than fat from chicken leg due to a large phospholipid fraction in the fat from chicken breast (Pikul et al., 1984c).

The high susceptibility of phospholipids to autoxidation in poultry is attributed to its high concentration of polyunsaturated fatty acids (Pearson et al., 1977; Yamauchi et al., 1982; Wu and Sheldon, 1988). The rate of autoxidation of fatty acids increases significantly with an increase in the number of double bonds (Wu and Sheldon, 1988). Pearson et al. (1977) claimed not only the high content of polyunsaturated fatty acids in the phospholipid fraction, but also the high content of polyunsaturated fatty acids in the triacylglycerol fraction contributed to the marked susceptibility of poultry meat to oxidative breakdown.

However, the polyunsaturated fatty acids of triacylglycerols are almost confined to linoleic acid and small quantities (<1%) of arachidonic acid. Conversely, phospholipids contain not only $C_{18:2}$ and $C_{18:3}$ in significant amounts but also high concentrations of $C_{20:3}$ and and other highly polyunsaturated fatty acids including $C_{20:4}$, $C_{22:4}$, $C_{22:5}$ and $C_{22:6}$ which undergo the greatest oxidative deterioration during storage (Igene et al., 1981; Pikul et al., 1984b; Wu and Sheldon, 1988).

Of the phospholipids in poultry meat, Wu and Sheldon (1988) reported that PC was present in the highest concentration and contained the highest amount of highly polyunsaturated fatty acids. They found that the reduction of polyunsaturated fatty acids in PC during the storage of poultry meat was also greatest among the phospholipid

fractions, while only minor changes in the polyunsaturated fatty acids of PE were observed. These results contradict the findings of Igene and Pearson (1979) in which PE was considered a major contributor to the development of WOF, while PC was less influential.

Tsai and Smith (1971) previously suggested that both the nitrogen moiety of the phospholipids and the degree of unsaturation influenced the level of lipid autoxidation. Thus, the ethanolamine fraction of PE not only increased the oxidation of PE but also accelerated the oxidation of other phospholipids (Corliss and Dugan, 1970; Tsai and Smith, 1971).

(2) Contribution of Membrane-bound Lipids

The membrane-bound lipids consist primarily of phospholipids and perform essential roles in membrane structure and function, being components of mitochondria, muscle fiber sarcolemma, microsomes and sarcoplasmic reticulum (Gray and Pearson, 1987). With a relative high content of polyunsaturated fatty acids, membrane-bound lipids are essentially vulnerable to oxidative change (Igene et al., 1980). Asghar et al. (1988) reported that the rate of peroxidation of the membrane-bound lipids can be influenced by the fatty acid composition of the membrane. They found that dark-muscle microsomal lipids peroxidized faster than white-muscle microsomal lipids, an observation similar to that reported earlier by Kanner and Harel (1985).

In addition to the high amount of polyunsaturated fatty acids in the membrane, some prooxidants such as oxygen, peroxidase enzymes (Vladimirov et al., 1980), heme and nonheme iron (Igene et al., 1979a), hydrogen peroxide, and superoxide radical (Kanner and Harel, 1985) are normally present in muscle cells. Thus, any process that disrupts the integrity of the membranes exposes the phospholipids to oxygen, enzymes, heme pigments and metal ions which can accelerate the development of WOF even in raw meat (Sato and Hegarty, 1971).

(3) Role of Heme and Nonheme iron

The catalytic effects of iron prophyrins and metal ions in lipid oxidation have been studied intensively over the years. The ability of heme compounds to catalyze lipid oxidation was estabilished by Tappel (1962), while the role of heavy metals in increasing the rate of lipid oxidation was summarized by Ingold (1962). Metals such as copper, iron and cobalt possessing two or more valency states with a suitable oxidation-reduction potential between them, are particularly important catalysts and function by increasing the rate of formation of free radicals (Ingold, 1962).

A number of workers (Tappel, 1962; Greene et al., 1971; Love and Pearson, 1971) reported that myoglobin (Mb) served as a prooxidant in meat and was responsible for development of rancidity. Decker and Schanus (1986a,b) investigated linoleate oxidation catalyzed by an aqueous extract of

chicken drumstick muscle and reported that both heme and nonheme catalysts may be involved in catalysis of lipid oxidation in raw chicken dark meat. However, Sato and Hegarty (1971) reported that heme compounds had little effect on the rate of oxidation in cooked meats, while ferrous iron (Fe^{2+}) at concentrations as low as 1 mg/kg enhanced lipid oxidation in samples of water-extracted cooked meats.

A similar water-extracted meat system has been used by the other researchers to investigate the catalytic effect of nonheme Fe^{2+} (Igene et al., 1979a; Tichivangana and Morrissey, 1984, 1985). Moreover, Tichivangana and Morrissey (1984, 1985) indicated the order of catalytic activity was Fe^{2+}> Cu^{2+}> Fe^{3+}> Mb> MetMb and that the prooxidant effect of metal ions and MetMb was more pronounced in heated muscle than in raw muscle.

Increasing levels of nonheme ion in meat as a consequence of heating have been demonstrated by several investigators (Igene et al., 1979a; Schricker et al., 1982; Schricker and Miller, 1983; Apte and Morrissey, 1987). They concluded that heme pigments serve as a source of free iron, being readily broken down during the cooking process and catalyzing autoxidation. Chen et al. (1984a) found both the rate of heating and the final temperature influenced the release of nonheme iron from meat pigment extracts. Slow heating released more nonheme iron than fast heating (Schricker and Miller, 1983; Chen et al., 1984a).

Recently, Apte and Morrissey (1987) investigated the effect of hemoglobin and ferritin on lipid oxidation in raw and cooked muscle systems. Their results indicated that ferritin exerted very little catalytic activity in raw meat. However, heating probably denatured the ferritin molecule, as it does for myoglobin and hemoglobin, releasing Fe^{2+} which then catalyzed lipid oxidation. Fe^{2+} can also be released from ferritin by low levels of ascorbic acid, while nitrite inhibited both hemoglobin- and ferritin-induced oxidation (Apte and Morrissey, 1987).

Lipid Stability in Restructured Poultry Products

Restructured meat is meat from low value parts of a carcass that is sectioned, flaked or chunked into smaller pieces then shaped by mechanical means to form a new product. The technology for manufacturing restructured meats was developed in the 1970's as a means for adding value to underutilized cuts of meat as well as providing a uniform product by controlling the portion size, shape, texture and fat content (Booren et al., 1981; Huffman et al., 1981a).

There are three basic procedures that can be utilized in the production of restructured meats: (1) chunking and forming, (2) flaking and forming, and (3) tearing and forming. The first two procedures have been widely used and are increasing in importance. The third procedure has not

yet been fully developed and requires special equipment that tears the meat fibers apart and then reforms them into shapes resembling intact cuts (Pearson and Tauber, 1984).

The cost savings and the ability to maintain product consistency give the restructured product great potential in the market place. However, the widespread acceptance of these products has been limited due to changes in quality which occur as the result of lipid oxidation. The stability of lipids in restructured products is largely influenced by the procedures used in manufacturing, namely reduction in particle size, cooking and the addition of sodium chloride. Some studies that investigated the action of these catalyic factors occurring during processing and the lipid stability of restructured meats during storage will be briefly reviewed.

Catalytic Factors Occurring during Processing

Sato and Hegarty (1971) indicated that comminution of fresh meat and subsequent exposure to air can cause the development of rancidity within one hour. The operations in the manufacture of restructured meats such as grinding, chopping, flaking or emulsification also disrupt membranes and lead to incorporation of air or oxygen into the tissues. Both of the actions increase tissue susceptibility to oxidation and hasten development of WOF (Gray and Pearson, 1987).

As indicated earlier, membrane-bound lipids are high in phospholipids and, because of their high content of polyunsaturated fatty acids are especially vulnerable to oxidative deterioration (Igene et al., 1980). Poultry meat is known to have a higher content of phospholipids than red meat (Acosta et al., 1966; Igene et al., 1981). Restructuring of poultry meat exposes these labile phospholipids not only to oxygen but to other tissue catalysts such as enzymes, heme pigments and metal ions (Gray and Pearson, 1987).

In recent years, mechanically deboned poultry meat (MDPM) has been utilized in restructured poultry products. The extreme stress and aeration during the mechanical deboning processes and the compositional nature (bone marrow, heme, and lipids) of MDPM contribute to its high oxidative potential (Dawson and Gartner, 1983). Pearson and Tauber (1984) suggested that up to 10% of mechanically deboned meat in restructured products not only reduces the cost but also improves the texture. However, use of more than 20% of mechanically deboned meat causes an adverse effect on flavor and texture.

Although cooking is not necessary for the manufacture of restructured meats, it is the common process for commercial production of restructured poultry products such as turkey rolls and chicken nuggets. Heat could also disrupt the membranes and break lipoprotein complexes, which would expose lipids to oxygen and catalysts and make them

more susceptible to oxidative attack (Dawson and Gartner, 1983).

Yamauchi (1972) reported that the degree of WOF was affected by processing temperatures between 60 and 120°C. Smith et al. (1987) investigated the effect of processing temperature on WOF development in ground chicken breast meat. They indicated that WOF in chicken can be minimized by cooking at low temperature, such that no part of the product exceeds 74°C. Nevertheless, Zipser and Watts (1961), Sato et al. (1973) and Huang and Greene (1978) found that high temperature (>90°C) heating could reduce WOF development in cooked meat due to the formation of Maillard reaction products possessing antioxidant properties. Not only the final temperature but also the cooking rate was found to influence the WOF development (Huang and Greene, 1978). Chen et al. (1984a) reported that a slow heating rate increased the release of nonheme iron which is a major prooxidant in cooked meat from beef pigment extracts.

A small amount of added salt is necessary to assure proper binding of the meat particles in restructured meat products. It has been reported that increased salt levels in restructured pork products increase thiobarbituric acid (TBA) numbers and decrease raw color scores (Schwartz and Mandigo, 1976; Huffman et al., 1981b; Rhee et al., 1983). More recently, King and Earl (1988) evaluated the effect of selected sodium and potassium salts on TBA values of dark meat turkey patties. They found that in formulations

containing no added polyphosphates, TBA values increased with increasing amounts of added sodium chloride.

Although sodium chloride, or salt, is known to initiate color and flavor changes in meat, the processes involved in salt-catalyzed oxidation are not fully understood (Pearson et al., 1977). It has been postulated by some that salt-catalyzed rancidity may be related to metal impurities in the salt. Nevertheless, rancidity still developed in fat of dry cured hams, even though a purified low-metal salt was used (Olson and Rust, 1973).

Chang and Watts (1950) demonstrated that the effect of salt on oxidation of fat is dependent on the amount of moisture in the system. Pokorny and Janick (1971) suggested that heme derivatives have no effect on lipid oxidation in the absence of water but begin to exert prooxidant activity at a moisture content of 60%. This could be due to the release of nonheme iron, which could then catalyze oxidation. On the other hand, salt has been reported to inhibit lipid oxidation under certain conditions. Mabrouk and Dugan(1960) observed that autoxidation of aqueous emulsions of methyl linoleate was suppressed by increasing concentrations of dissolved salt in the system. They suggested that dissolved oxygen may be eliminated from the system as the salt concentration is increased. These findings implicated that the effect of salt could be both a prooxidant and an antioxidant, depending on the concentration dissolved.

Lipid Stability during Storage

Although WOF develops rapidly in meat stored at refrigerated temperatures, it also develops in frozen meat products during long-term storage (Gray and Pearson, 1987). The rate of WOF development and the changes in lipid content are different between refrigerated and frozen storage. Ang (1988) correlated the oxidative changes of the relevant constituents of cooked broiler meat during refrigerated storage. She reported that the oxidation rate during refrigerated storage was positively correlated with protein, water, phospholipids and nonheme iron content and negatively correlated with total lipids or neutral lipids.

Igene et al. (1980) found that the total lipid content of raw meat during frozen storage is affected mainly by losses in the triacylglycerol fraction while the phospholipid fraction remains relatively constant, irrespective of the length of frozen storage. In the same study, a decrease in $C_{18:2}$and $C_{18:3}$ fatty acids was observed in the triacylglycerol fraction of chicken dark meat and white meat during frozen storage. The dienoic acids (principally $C_{18:2}$) decreased from the original value in chicken dark and white meat by 43 and 14%, respectively (Igene et al., 1980). Frozen storage of fresh meat does not appear to cause serious oxidative deterioration of the membrane-bound lipids, but primarily acts on the triacylglycerols.

Igene et al. (1981) further studied the effect of length of frozen storage, cooking and holding temperaturature on component phospholipids of chicken meat. They reported that there was a significant decline in the amount of PE and PC during frozen storage, but the decline was much greater on cooking. PE and PC was found to be more stable following cooking on holding at $-18^{o}C$ than $4^{o}C$. The oxidation of triacylglycerols in poultry was also found to be related to the degree of unsaturation and the length of frozen storage (Igene et al., 1980; Pikul et al., 1984a).

Measurement of Lipid Oxidation

Lipid oxidation is a problem that has been associated with foods containing fat. The susceptibility and rate of oxidation of fatty acids vary according to their degree of unsaturation and the presence of activating agents such as heat, light, enzymes or metal catalysts (Nawar, 1985). The classical mechanism for lipid oxidation is via free-radical attack, and the initial substrate for oxidation is unsaturated lipids. The following simplified scheme shows the various steps in the free radical chain mechanism for lipid oxidation:

Initiation: $RH \text{ ----------> } R^{\bullet} + H^{\bullet}$

$RH + O_2 \text{ --------> } ROO^{\bullet} + H^{\bullet}$

Propagation: $R^{\bullet} + O_2 \text{ --------> } ROO^{\bullet}$

$ROO^{\bullet} + RH \text{ ------> } ROOH + R^{\bullet}$

Termination: $ROO^{\bullet} + R^{\bullet} \text{------} > ROOR$

$R^{\bullet} + R^{\bullet} \text{--------} > R\text{-}R$

$ROO^{\bullet} + ROO^{\bullet} \text{----} > ROOR + O_2$

$ROO^{\bullet}$ is a lipid peroxy radical, $R^{\bullet}$ is a lipid alkyl radical, and RH is an unsaturated lipid. Once the reaction has been initiated, the hydroperoxides (ROOH) which are formed subsequently decompose to free radicals, which in turn can accelerate the rate of lipid oxidation and/or form secondary products (Simic and Taylor, 1987). The secondary products and their decomposition products including aldehydes, ketones, acids and other carbonyl compounds may cause the off-flavor in food or react with the other substances in food (Khayat and Schwall, 1983).

Many chemical and physical methods have been developed to assess lipid oxidation. In this review, several methods, 2-thiobarbituric acid test, gas chromatographic analysis and sensory evaluation, commonly used for measuring lipid oxidation in meat systems will be addressed. A standardized physical method, the active oxygen method and its automated form--Rancimat, for evaluating the resistance of oils or fats to oxidation will be discussed briefly.

2-Thiobarbituric Acid Test

The most commonly used method for measuring oxidative changes in meat products is the 2-thiobarbituric acid (TBA) test. This procedure is based on the spectrophotometric quantitation of a red-violet complex formed between

malonaldehyde, a colorless end product of lipid oxidation, and TBA (Sinnhuber and Yu, 1958; Gray, 1978; Melton, 1983). There are three ways in which the TBA test can be performed: (1) directly on the meat products, followed by extraction of the colored complex; (2) on an extract of the meat; and (3) on an extract of the steam distillate of the meat (Melton, 1983). The method involving steam distillation is the most popular method for measuring the TBA number in muscle foods (Rhee,1978) and has been used extensively without modification to assess the oxidative deterioration of lipids in beef, pork and poultry products (Melton, 1983).

However, in recent years, some investigators have suggested that the method should be modified to include the addition of antioxidant at the blending or distillation stages of the procedure. Rhee (1978) reported that chilled blending and the addition of propyl gallate (PG) and EDTA at the blending and distillation stages substantially lowered the TBA number of catfish steaks. Pikul et al. (1983) evaluated the influence of butylated hydroxytotuene (BHT) on the TBA assay for malonaldehyde in fat extracted from chicken breast and leg meat. They recommended that an antioxidant be added during sample homogenization, distillation, or the extraction step of the TBA assay to prevent sample autoxidation and artificially high analytical results.

The TBA test principally determines both the malonaldehyde already formed naturally from hydroperoxide

cleavage and the secondary release due to the heating step in the reaction. However, some end products of lipid peroxide breakdown, other than malonaldehyde, will also react with TBA and may produce orange or yellow pigments, depending on the TBA test reagents (Tarladgis et al., 1964; Igene et al., 1985). Tarladgis et al. (1964) reported that malonaldehyde, the principal TBA-reactive substance (TBARS), not only can be released upon heating the sample in a acid medium, but also can be measured without the acid treatment. Crackel et al. (1988b) suggested that a modified TBA distillation method in which aqueous TBA reagent was used instead of the acid solution can decrease the formation of yellow pigments as well as improve the reproducibility regardless of the degree of oxidation in meats. Schmedes and Holmer (1989) described a new TBA method for selectively determining free malonaldehyde and lipid hydroperoxides by partitioning malonaldehyde and hydroperoxides in a Bligh and Dyer extraction before TBA reagent was added. This method is suitable for samples with a low fat content such as minced cod.

Recently, a systematic study of the influence of peroxidation conditions on lipid hydroperoxide decomposition to TBA-reactive substances (TBARS) was reported by Janero and Burghardt (1989). They found that malonaldehyde represented the major TBARS in purified lipid but only 20-60% of total TBARS in the membrane/tissue systems. Therefore, the presence of nonlipid-related, non-

malonaldehyde TBARS in biological samples represents a major limitation to the quantitative use of the TBA test. Many investigators have come to the same conclusion and suggested that the results of the TBA test need to be considered with sensory evaluation or with results of other suitable chemical tests (Gray, 1978; Csallany et al.,1984; Igene et al.,1985).

Gas Chromatographic Analysis

Hexanal, one of the major secondary products formed during oxidation of linoleic acid (Frankel et al., 1981), and other aldehydes have been used sucessfully to follow lipid oxidation in foods (Melton, 1983). These volatile compounds have been identified in oxidizing meat systems including cooked, uncured pork (Cross and Ziegler, 1965) and hams (Lillard and Ayres, 1969; MacDonald et al., 1980), and have been associated with WOF development in cooked turkey (Ruenger et al., 1978) as well as cooked ground pork (Shahidi et al., 1987).

Several gas chromatographic (GC) techniques have been applied to the analysis of volatile compounds in foods. Fritsch and Gale (1977) utilized a headspace gas-sampling technique followed by GC analysis to measure hexanal in cereal foods. Volatiles were released from the cereal by the addition of boiling water, the system was sealed and a sample of headspace gas was withdrawn for GC analysis. Another approach for the collection of volatiles was

described by Bailey et al. (1980). Volatiles released by boiling an aqueous homogenate of a meat sample were extracted into a suitable organic solvent in a Likens-Nickerson extraction apparatus, a closed continuous extraction system. The solvent containing the extracted volatiles was concentrated and analyzed using conventional GC methods.

Legendre et al. (1979) developed a direct GC method that can be applied to the analysis of aqueous and non-aqueous foods and reported that this procedure was rapid, efficient and sensitive enough that sample volatiles can be analyzed without the need for prior enrichment.

The direct GC method has been modified with a purge-and-trap system and efficiently used for the analysis of volatiles in fish, beef and pork (Josephson et al., 1983; Vercellottli et al., 1987; Shahidi et al., 1987). The volatiles stripped from the meat samples were trapped in a Tenax trap and injected directly into the GC. Moisture-free samples and subambient oven temperature programming allowed effective resolution of low molecular weight volatiles as well as those of higher molecular weight (Vercellotti et al., 1987).

In addition, an external closed inlet device (ECID) can be adapted to the GC. The meat to be analyzed is placed in an external inlet assembly where the volatiles are stripped from the sample by heat and an inert carrier gas. The volatiles condense onto a GC column and subsequently are

resolved by temperature programming (St. Angelo et al., 1988). With the ECID and a capillary column, the resolution and sensitivity were greatly increased and more than 150 compounds were identified in the volatiles of the WOF roast beef (St. Angelo et al., 1987; St. Angelo et al., 1988).

Sensory Evaluation

Sensory analysis has been used in the study of WOF for over 30 years. Tims and Watts (1958) noted that meat flavor can deteriorate after only a few hours of refrigerated storage. They called the flavors developed during storage "warmed-over", "rancid" or "stale".

Sensory evaluation was not only used to elucidate the mechanisms involved in WOF development, but also to evaluate the efficacy of other analytical methods for measuring WOF. The relationships of TBARS numbers, peroxide values and hexanal contents of muscle foods to sensory scores have been studied by many researchers (Tarladgis et al., 1960; Zipser et al, 1964; Igene and Pearson 1979; Igene et al., 1979b, 1985; Greene and Cumuze, 1981; St. Angelo et al., 1987). Although these studies showed significant correlations between sensory scores and chemical tests, there are some variations among these reports. The inconsistent data may be due to the use of different category scales, use of trained or untrained panelists, and use of different descriptors of WOF (Poste et al., 1986; Love, 1988).

Addis and Pearson (1986) stated that in studying WOF

one must first select a sample with WOF and train panelists to recognize it and then screen out those who cannot detect WOF. The numbers of panelists used for sensory evaluation of off-flavors in meat varies, but it is desirable to have 6-12 people on a trained panel (Melton et al., 1987).

Rancimat Method

The Active Oxygen Method (AOM) or Swift test has proved very popular for evaluating the stability of fats and oils and has been standardized in the Official and Tentative Methods of the American oil Chemist's society (Method cd. 15-27, AOAC, 1975). In order to determine oxidative stability, a fat is exposed to a stream of dry air at a temperature of 100-140^{o}C. The progress of the oxidation curves can be followed by periodic determination of the peroxide value (PV). The curves comprise an induction phase, in which practically no secondary products are formed, followed by an oxidation phase, during which a large increase in PV and volatile products is detected (Laubli and Bruttel, 1986). The induction time obtained from the curve is a good indication of resistance of fats or oils to oxidative rancidity. Older fats generally have shorter induction times (Frank et al., 1982).

Because of the repeated PV determinations involved, the measurement of this parameter with AOM is very costly and labor intensive. An alternative method, the Rancimat procedure, for the automatic analysis of oxidative stability

was developed by Hadorn and Zürcher (1974). This method is based on the conductometric determination of volatile degradation products (mainly formic acid) and features automatic plotting of conductivity against time (Frank et al., 1982). The progress of the oxidation curves determined in this manner is virtually parallel to the development of the PV (Hadorn and Zürcher, 1974). The results obtained with the Rancimat method also showed extremely high correlation with those of the AOM test (Laubli and Bruttel, 1986).

Prevention of Warmed-over Flavor Development

Although WOF is recognized as a major problem in precooked or restructured meat products, it can be effectively controlled or minimized by the use of antioxidants or curing with nitrite. Nitrite acts as an effective inhibitor of WOF by stabilizing the prophyrin ring of heme pigments and preventing the release of iron during the cooking process (Asghar et al., 1988). Thus, WOF is not as serious a problem in cured meats as it is in noncured meat products.

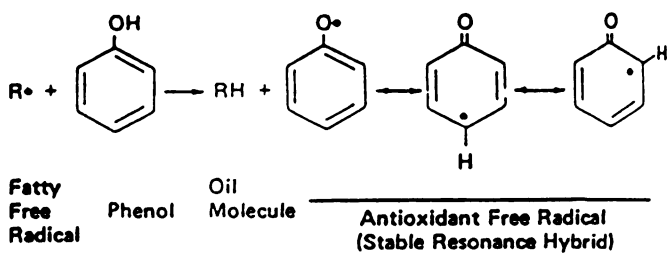
Other compounds used to prevent lipid oxidation or WOF in meats include polyphosphates and ascorbate, as well as synthetic and natural antioxidants (Pearson and Gray, 1983). Labuza (1971) classified antioxidants into three categories: Type I, free radical terminators, primarily phenolic

compounds that donate an electron to a free radical; Type II, free radical production preventers, mostly chelating agents that tie up transition metals; and Type III, environmental factors such as redox compounds and water activity regulators. Roozen (1987) used a meat model system to test the effectiveness of these three types of antioxidants in preventing peroxidation. These results showed that Type I and Type II antioxidants decreased the amount of TBARS in meat, while many of Type III antioxidants gave an increase, depending on their concentration.

In this section, the antioxidant activity and application to meat products of Type I antioxidants including synthetic and natural phenolic antioxidants, will receive major emphasis. The role of phosphates, which are Type II antioxidants, in the prevention of WOF development in meats will also be addressed.

Synthetic Phenolic Antioxidants

Phenolic antioxidants are Type I antioxidants because they function by interfering with the free radical mechanism of autoxidation. Their antioxidant action may be simply described as follows:



where the phenolic substance accepts the free radical and forms an antioxidant free radical. This radical intermediate is relatively stable due to resonance delocalization and lack of positions suitable for the attack by molecular oxygen, and thus is incapable of initiating or propagating further oxidation (Sherwin, 1978; Nawar, 1985).

Phenolic compounds perform differently from one another as antioxidants, their activity depending on the types and positions of substituent groups affixed to the benzene ring nucleus of the compound (Pokorny, 1971; Sherwin, 1985). Butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG), and tertiary butylhydroquinone (TBHQ) are the most widely used synthetic phenolic antioxidants and have been broadly studied in meat systems. These compounds can be used alone or in combination. Synergism occurs when a mixture of antioxidants produces a more pronounced activity than the sum of the activities of the individual antioxidants when used separately (Nawar, 1985). Other chemical properties, such as melting point, carry-through properties, and solubility, also need to be considered when selecting an antioxidant or deciding on a medium which can achieve the best dispersion of antioxidant in meats.

Conflicting data have been published in the literature concerning the effectiveness of lipid-soluble antioxidants in preventing WOF. One problem appears to be the distribution of the antioxidant in the appropriate tissue

(Bailey, 1980). Watt (1961) concluded that phenolic antioxidants were of little value in intact meat cuts.

However, many studies have shown that phenolic antioxidants are effective in preventing WOF in comminuted meat systems. Moerck and Ball (1974) reported that an antioxidant mixture containing 20% BHA, 6% PG, and 4% citric acid in propylene glycol (Tenox IV^{TM}) added at 0.01% by weight was effective in preventing oxidation in mechanically deboned chicken. Barbut et al. (1985) found that the addition of BHA/BHT to turkey sausages substantially reduced measurable TBARS formation during refrigerated storage for up to 16 days. BHA and PG were also demonstrated to be effective in retarding lipid and pigment oxidation in ground beef during refrigerated storage for more than one week (Greene, 1969; Greene et al., 1971). Similar results were obtained in cooked ground beef using BHA and/or BHT (Sato and Hegarty, 1971; Chen et al., 1984b).

Of more direct relevance to restructured meats is the work of Chastain et al. (1982), who studied the effects of BHA, TBHQ, and a combination of these two antioxidants on the quality of restructured (50:50) beef-pork steaks. They found that the antioxidants when used alone or in combination lowered TBARS values and decreased discoloration in the steaks. BHA was more effective in inhibiting discoloration while TBHQ was slightly superior in retarding off-flavor development during 20 weeks of frozen storage. Miles et al. (1986) evaluated the effect of BHA/BHT/citric

acid (1:1:1) on the oxidation of restructured pork chops in refrigerated storage. Their results indicated that antioxidants were effective in controlling discoloration and oxidation of restructured pork stored at refrigerated temperature. In addition, the protective effect of TBHQ on lipid stability in restructured beef steaks over 12 months of frozen storage was also confirmed by Crackel et al. (1988a).

Natural antioxidants

Although synthetic phenolic antioxidants can be used successfully to prevent the development of WOF, consumers are becoming increasingly concerned about the use of synthetic chemicals, in contrast to the use of natural products. The present trend in food processing is to use natural ingredients, even though they have not been shown to be safer than synthetic additives used for the same purpose (Chang et al., 1977).

There have been extensive investigations of the use of antioxidants from naturally occurring substances in food products. However, it has not yet been specified what is meant by "natural antioxidant". There are a great variety of natural products which possess antioxidants activity. Such antioxidants can be found in vegetable oils, mostly as tocopherols (Dougherty, 1988). Some researchers also described Maillard reaction products and ascorbic acid as natural antioxidants. Many of these antioxidants

investigated were reported to be successful in retarding or inhibiting the formation of WOF in meats (Pearson and Gray, 1983; Rhee, 1987).

Another group of frequently used antioxidants which have potential for even more application in the future are extracts from vegetables, spices, and herbs. Watts (1961) found that hot-water extracts of many vegetables, including celery leaves, turnip greens, mint leaves, carrots, white potato skins, beet stems, and mushrooms, were effective in retarding oxidation of lipids in cooked meats. Pratt and Watts (1964) later demonstrated that a number of plant extracts possessed potent antioxidant properties when added to sliced roast beef. Younathan et al. (1980) also reported that rancidity in turkey was effectively controlled by hot-water extracts of eggplant tissues and peelings of yellow onions, potatoes, and sweet potatoes. Recently, Jurdi-Haldeman et al. (1987) reported that onion juice was a more effective antioxidant than garlic juice in reducing development of rancidity in meat.

All protective effects found in these researches were attributed to the flavone and flavonoid contents of the vegetable extracts. The antioxidant activity of these compounds is apparently related to the ketone in the 4 position of the pyrone ring and the 2,3-olefin group. The 3,4- and 5,7-dihydroxy groups (shown in Figure 1) apparently contributed to the antioxidant role (Crawford et al., 1961).

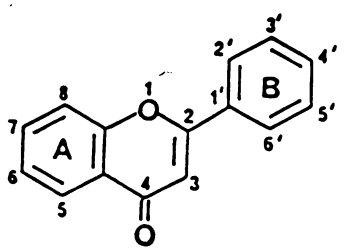


Figure 1. Parent flavone structure.

Spices and herbs have also been studied as antioxidants. Early observations that most common spices have antioxidant properties in lard were made by Chipault et al. (1952). Gerhardt and Blat (1984) comprehensively evaluated the antioxidant activity of spices in pork fat, and the stability of the fat was determined by a dynamic test involving Rancimat methodology. Results of this study indicated clearly that the majority of the spices tested possessed some antioxidant activity (Table 3), with rosemary, sage, thyme, and mace being the most potent.

The antioxidant properties of spices are apparently related to their phenolic contents, thus their antioxidant action is similar to synthetic phenolic antioxidants. However, the applicability of spices and herbs to meat products is determined by their sensory compatibility with each specific meat product concerned. Barbut et al. (1985) reported that a low flavor, organic-solvent extracted oleoresin rosemary possessed antioxidant activity comparable to a commercial blend of BHA/BHT/citric acid in suppressing lipid oxidation and did not adversely affect the overall

Table 3. Antioxidant activity of various spices added to pork fat and subjected to an accelerated oxidation test[1].

Spice	Protection Factor[2]	Antioxidant Activity Group Rating[3]
Mace	4.10	Optimal
Rosemary	4.30	Optimal
Sage	4.00	Optimal
Thyme	4.60	Optimal
Nutmeg	2.59	Very good
Ginger	2.11	Good
Clove	1.95	Good
Marjoram	1.72	Good
Turmeric	2.16	Good
Paprika	1.51	Good
Garlic	1.11	Neutral to slight
White pepper	1.15	Neutral to slight
Cumin	1.19	Neutral to slight
Coriander	1.10	Neutral to slight

[1] Adapted from Gerhardt and Blat (1984).
[2] Protection factor is ratio of induction period of sample (pork fat + spice) to the induction period of the blank (pork fat).
[3] Optimal antioxidant effect, protection factor > 3.5; very good, 2.5-3.5; good,1.5-2.5; neutral, 1.0-1.5.

palatability of the product. Because rosemary extracts have been shown to possesse superior antioxidant activity in several fat substances, many researchers have devoted their time to examining its unique properties. The chemistry and application of rosemary will be discussed later in detail.

Phosphates and Other Chelating Agents

Tim and Watts (1958) not only coined the expression "WOF" in cooked meat, they also demonstrated that phosphates effectively reduced WOF in meats. They reported that pyro-, tripoly- and hexametaphosphates had a protective effect, whereas orthophosphate did not. Sato and Hegarty (1973) further verified the effects of these phosphates in preventing rancidity in precooked ground beef, with tripolyphosphate being the most effective.

Polyphosphates have been observed to reduce oxidative changes in cooked chicken meat (Thomson, 1964; Farr and May, 1970). Marion and Forsythe (1962) also reported that polyphosphates had a protective effect on oxidation of uncooked turkey meat during storage. The antioxidant effect of polyphosphates in mechanically deboned poultry was confirmed by MacNeil et al. (1973). Furthermore, Froning (1973) found that polyphosphate-treated mechanically deboned chicken meat had significantly lower TBA values than those found in the controls at all storage periods. According to Watts (1962), polyphosphates effectively protect cooked

meats against oxidation at concentrations as low as 0.01 - 0.05%.

The phosphates appear to prevent autoxidation by chelating metal ions, specially ferrous ion which has been shown to be the major catalyst of lipid oxidation in cooked meats (Love and Pearson, 1974; Igene et al., 1979a). A number of other metal ion chelators have also been shown to be effective as inhibiters of oxidation in meat system. Benedict et al. (1975) reported that both citric acid and 1-ascorbyl stearate showed a limited effect in decreasing lipid and heme oxidation in lean beef ground with beef fat. MacDonald et al. (1980) demonstrated that citric acid reduced TBA values in refrigerated hams when used at the 1000 ppm level but was not as effective as 50 ppm of nitrite. Besides, EDTA has been shown to be an effective inhibitor for development of WOF in cooked meat by Igene et al. (1979a). However, EDTA has limited use in meat products because it has not been approved for use in these products.

Rosemary as an Antioxidant

Rosmarimus officinalis L. has been shown to be an effective antioxidant in a variety of fats (Chipault et al., 1952, 1955; Gerhardt and Blat, 1984). However, the applicability of whole spices in foods may be limited by the specific product. To obtain an active extract with lower color and odor from the leaf of rosemary, scientists have

developed several processes involving a variety of solvents and distillation stages. In addition, much effort has been directed toward the identification of the active principles in the extracts.

Studies involved in the preparation of active extracts from rosemary as well as the isolation and identification of active antioxidant compounds in rosemary will be discussed. Several recent studies on the antioxidant effect of the rosemary extract in meat products will also be reviewed.

Preparation of Rosemary Extracts

It has been found that a somewhat concentrated but crude preparation of a rosemary extract which has considerable antioxidant properties may be obtained by extracting the leaves of rosemary with alcohol or other appropriate solvents (Löliger, 1983). Although such crude extracts have been described as purified and of substantially less taste or odor than the natural spice, they are not fully free of the intrinsic characteristics of the spice. To obtain a particular product form of the rosemary extracts that does not have an objectionable odor or color, Chang et al. (1977) developed a process involving alcoholic extraction, followed by vacuum steam distillation or molecular distillation. The extract at 0.02% was found

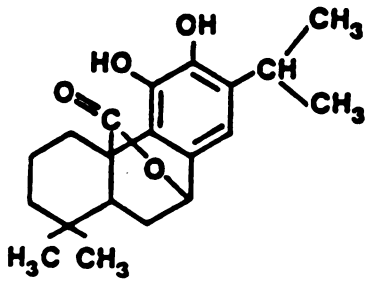
to be as effective as Tenox VI^{TM} (BHA/BHT/PG and citric acid) in lard and vegetable oil.

Bracco et al. (1981) later developed a new industrial process for obtaining natural antioxidants from rosemary and cocoa shells. The micronization of spices in vegetable oil is the main step in the process and results in the rupture of the cell walls, releasing the antioxidant compounds and yielding a lipid suspension for the molecular distillation steps. Through the two-stage falling film molecular distillation, the lipid phase is separated and a distillate which has essentially no taste and odor, with a light red/orange color is obtained. The antioxidant activity of this distillate is comparable to that of BHA or BHT when tested in chicken fat (Bracco et al., 1981).

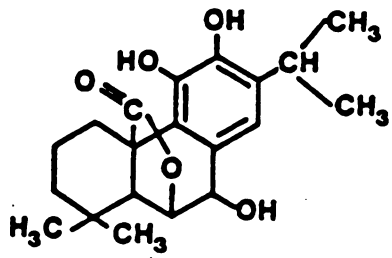
Composition and Structure of Rosemary Extracts

The molecular distillate and the crude extract of Rosemarinus officinalis L. are both composed of at least 45 compounds as analyzed by reversed phase HPLC with a UV detector; about half of these compounds have been identified. Such analytical work is very complicated, due in part, to the instability of many of the components in pure form (Loliger, 1983).

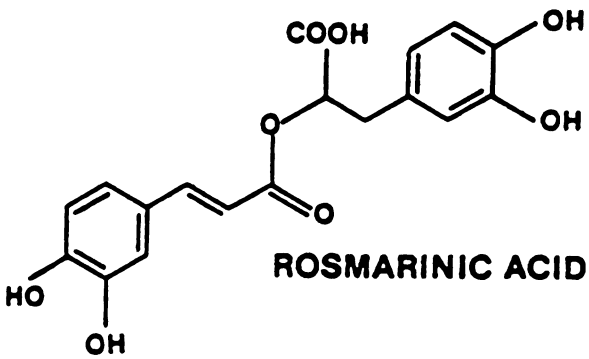
The first important antioxidant compound isolated from rosemary leaves was a phenolic diterpene named carnosol (Figure 2), the structure of which was determined by Brieskorn et al. (1964). Wu et al. (1982) also identified



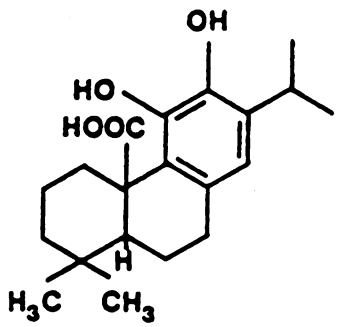
CARNOSOL



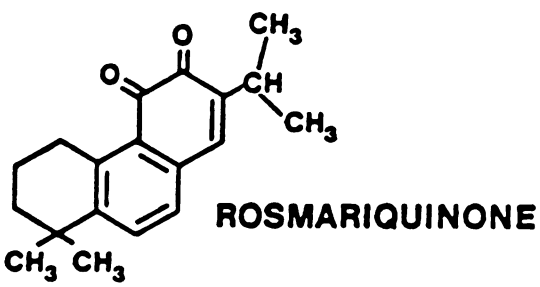
ROSMANOL



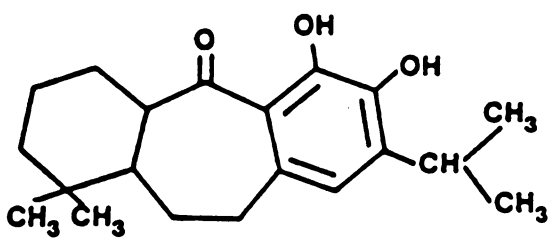
ROSMARINIC ACID



CARNOSIC ACID



ROSMARIQUINONE



ROSMARIDIPHENOL

Figure 2. Structures of some antioxidant compounds in rosemary extracts.

carnosol from rosemary leaves and found that its antioxidant activity was similar to BHT, when added to lard at the 0.02% level.

Inatani et al. (1982) isolated and characterized another antioxidant compound, rosmanol, from rosemary leaves. Later, Inatani et al. (1983) reported that rosmanol at a 0.02% level was a better antioxidant than BHA or BHT, using the active oxygen method (AOM). Several derivatives of rosmanol were also identified and reported to be as effective as rosmanol by the same research group. Isoromanol and epirosmanol, in particular, were found to be more effective at the 0.005% than were BHA and BHT at the 0.02% level in lard (Nakatani and Inatani, 1984).

Meanwhile, Houlihan et al. (1984) isolated a novel antioxidant from rosemary leaves, named rosmaridiphenol (Figure 2). They also found that the antioxidant activity of this diphenolic diterpene surpassed BHA and approached the effectiveness of BHT when tested at the 0.02% level in prime steam lard. Later, another active compound was isolated from rosemary leaves, named rosmariquinone (Figure 2), with similar antioxidant activity to rosmaridiphenol(Houlihan et al., 1985). The antioxidant activity of rosmariquinone was superior to BHA, but slightly less than BHT when tested in prime steam lard at a concentration of 0.02% (Houlihan et al., 1985).

Antioxidant Effect of Rosemary in Meat Products

Although many studies have demonstrated the antioxidant properties of rosemary or rosemary extract in fat and oil systems, very little research has been done to incorporate these compounds into meat products. MacNeil et al. (1973) compared the antioxidant effect of a rosemary spice extract (RSE) to that of polyphosphates and BHA/citric acid in cooked, simulated mechanically debonded turkey meat (MDTM) at refrigerated storage. They found that RSE (0.05%) and BHA/citric acid treatments exhibited substantially lower TBA values than did polyphosphates and RSE (0.01%). However, the sensory score of RSE (0.05%) was lower than that of the control, whereas the RSE (0.01%) had a better flavor rating than any of the other treatments. Recently, Barbut et al. (1985) used rosemary oleoresin (20ppm) as an antioxidant in turkey breakfast sausages prepared with 75% hand deboned turkey meat and 25% MDTM. Chemical and sensory analyses showed that rosemary oleoresin was comparable to a commercial blend of BHA/BHT/citric acid in suppressing lipid oxidation and extending of the shelf life at refrigerated storage.

More recently, Korczak et al. (1988) investigated the effects of several spices on the oxidative stability of minced meat products under refrigerated and frozen conditions. Rosemary, when added at 0.1% and 0.5% levels, exhibited a pronounced antioxidant effect in frozen storage. However, rosemary did not have a significant protective

effect in refrigerated storage. Sheu (1988) also evaluated the effects of oleoresin rosemary and other antioxidants in the prerigor, precooked, vacuum packaged pork patties during refrigerated storage. He reported that oleoresin rosemary, ascorbic acid and sodium tripolyphosphate (STPP) significantly retarded the lipid oxidation thoughout the storage period. Strong synergism was also found when oleoresin rosemary was combined with ascorbic acid as well as STPP.

MATERIAL AND METHODS

Experimental Description

The experiment was designed to evaluate the efficacy of oleoresin rosemary (OR) as an antioxidant in precooked restructured chicken nuggets by incorporating OR into chicken meat and/or frying oil. The effect of OR on resistance of oil to oxidation during the heating process was also included in the study.

Restructured chicken nuggets formulated to 7% fat were prepared from a combination of white (50%) and dark (30%) meat and mechanically deboned meat (20%) using a standard commercial procedure. Seven treatments with different ingredients which were added to the meat batter during blending were formulated as:

1. salt 0.75%
2. salt 0.75% + OR 0.05%
3. salt 0.75% + OR 0.1%
4. salt 0.75% + sodium tripolyphosphate (STPP)
5. salt 0.75% + STPP 0.3% + OR 0.05%
6. salt 0.75% + STPP 0.3% + OR 0.1%
7. salt 0.75% + STPP 0.3% + TBHQ 0.02%

where the percentage of all ingredients were based on total weight of the meat, except that OR and TBHQ were based

on the lipid content of the meat. All treatments of chicken nuggets were fried in a commercial soybean oil without any antioxidant added. In addition to frying in soybean oil without antioxidant, formula 4 was also fried in soybean oil cnotaining one of two levels of OR (0.025% and 0.05%), thus resulting in treatments 8 and 9, respectively.

The nuggets were refrigerated at 4^{o}C for six days and frozen at -20^{o}C for six months. TBA test and sensory evaluation were carried out after 4 even periods of storage time (0, 2, 4, 6 days/months). In the frozen study, the changes in fatty acids in the triacylglycerol and phospholipid fractions were monitored at the 2nd, 4th, and 6th month. The hexanal content was only measured after six months of frozen storage.

Three formulations of soybean oil with 0%, 0.025% and 0.05% OR were heated at 176^{o}C for 0, 12, 24, 48, 96 hours in the open atmosphere. Rancimat and fatty acid analyses were taken to evaluate lipid stability as influenced by the different heating times.

Three replications were carried out for all experiments except the study of the heated oil which was only replicated twice. Chemical analyses for all studies were done in duplicate.

Preparation of Chicken Nuggets

Ingredients

Hand deboned chicken white and, dark meat and mechanically deboned chicken (MDC) without skin were obtained from a local processing plant (Nottwa Gardens Co., Athens, MI) and processed within 24 hours of purchase.

The other ingredients included salt (Cargill Inc., Minneapolis, MN), sodium tripolyphosphate (Stauffer Chemical Co., Westport, CT) and TBHQ (Tenox TBHQ, Eastman Chemical Products Inc., Kingsport, TN). The oleoresin rosemary-coated salt (OR 100) was prepared by Diamond Crystal Salt Company (St. Clair, MI) using an oleoresin rosemary preparation (Herbalox^{TM} Seasoning, O Type) which was supplied by Kalsec Inc. (Kalamazoo, MI). This oleoresin rosemary was also directly added to the soybean oil. The coating for the chicken nuggets was made from an all purpose white batter mix and standard breading which were produced by Newly Weds Food Inc. (Chicago, IL). Soybean oil (Draft Inc., Glenview, IL), without any antioxidant, was used in the processing of the chicken nuggets and the heated oil study.

Processing of Chicken Nuggets

Precooked, restructured chicken nuggets were manufactured using a commercial procedure under Regulation 9CFR 381 (USDA-AHS 1986 Purchase Specs).

The TBA test and lipid analysis were carried out prior to processing to ascertain the freshness of the meat and to determine the amount of antioxidant to be added.

After all visible fat was removed, the white and dark meat was cut into $5cm^2$ pieces and ground once through a 95mm plate using a Hobart meat grinder (Model A-200, Kitchenaid Division, Hobart Co. Troy, OH). The ground meat containing 50% white, 30% dark and 20% MDC was then blended with 5% water, 0.75% salt and/or antioxidants at speed 1 for 30 seconds to thoroughly mix all the ingredients, then increased to speed 2 for another 2 minutes in a mixer with the paddle attachment (Model A-200, Kitchenaid Division, Hobart Co., Troy, OH).

The meat was then stuffed into 4.5cm diameter fibrous casings (Viskase Co., Chicago, IL) and frozen at $-20^{o}C$ for 4 hours followed by slicing with a meat slicer (Model 512, Hobart Co., Troy, OH). The sliced patties, averaging 9.5mm in thickness were dipped in the batter, immediately drained and immersed in the breading (bread crumb). The breaded frozen patties, averaging 21g in weight, were tempered at $4^{o}C$ for approximately one hour. After tempering, the patties were fried at $176^{o}C$ in a Hotpoint fryer (model HK 3, General Electric, Chicago, IL) until an internal temperature of $76^{o}C$ was reached. Temperature was monitored by copper constantan thermocouples placed in the center of individual nuggets in the first batch. Using this batch, the average frying time for nuggets to reach the desired internal

temperature (76°C) was calculated. The calculated frying time was then used for subsequent batches. It was assumed that the size and thickness of each nugget did not differ enought to effect the frying time.

The cooked nuggets were immediately cooled in a freezer (-20°C) and packaged (without vacuum) in polyethylene-laminated nylon pouches (Koch, Kansas City, MO). These pouches (3.5 mil) have an water-vapor transmission rate of 0.041 $ml/m^2 \cdot day \cdot mmHg$ and an oxygen transmission rate of 0.124 $ml/m^2 \cdot day \cdot mmHg$ at 22.7°C. 50% RH. The packed samples were then divided into two batches for refrigerated (4°C) and frozen (-20°C) studies.

Preparation of Heated Oil for Rancimat Study

Soybean oil, similar to that used for frying the chicken nuggets, was used for the heated oil study. The oil samples containing one of three levels of oleoresin rosemary, 0%, 0.025% and 0.05%, were heated at 176°C for 96 hours in a 1.8L fryer (Fry Daddy, National Presto Industries Inc., Eau Claire, WI) with an autotransformer (Staco Energy products Co., Payton, OH) to control the temperature. Heated oil samples were taken from the fryers periodically, cooled and put in the freezer (-20°C) until analyzed.

Methods of Analysis

All chemical reagents and solvents utilized in this study were analytical grade and/or HPLC grade. Individual esters and standard mixtures for fatty acid analysis were purchased from Supelco Inc. (Bellefonte, PA) and Alltech Associates, Inc. (Beerfield, IL). 2-Heptanone and n-hexanal standards for quantitative measurement of hexanal in the chicken nuggets were obtained from Sigma Chemical Co. (St. Louis, MO).

2-Thiobarbituric Acid (TBA) Test

TBARS values (mg malonaldehyde per kg tissue) were measured in duplicate on whole chicken nuggets using the distillation method of Tarladgis et al. (1964), as modified by Crackel et al. (1988b). In the modified method, an aqueous TBA solution was used instead of the acetic acid/TBA reagent and the TBA factor for this procedure was determined to be 6.2 (Crackel et al., 1988b).

Lipid Extraction and Fractionation

The dry column method of Marmer and Maxwell (1981) was used for simultaneous lipid extraction and class separation. The breading from the chicken nuggets was removed carefully prior to analysis. Ground meat (5g), anhydrous sodium sulfate (20g) and Celite 545 (15g) were ground with a mortar and pestle to a uniform, free flowing powder. The powder was quantitatively transferred to a glass column (35 x

300mm) and lightly tamped to obtain a uniform bed. The neutral lipids were eluted with 150ml dichloromethane . After the neutral lipids were eluted, 150ml dichloromethane/methanol (9:1) were added to the column to elute the phospholipids. Each lipid fraction was collected and concentrated to 25ml, then stored at -20^{o}C until further analysis.

Fatty Acid Analysis

Fatty acid methyl esters of total lipid and neutral lipid fractions were prepared following the boron trifluoride-methanol procedure of Morrison and Smith (1964). The phospholipid fraction was methylated according to the direct transesterification method of Maxwell and Marmer (1983).

Fatty acid methyl esters were analyzed using a gas chromatograph (Model 5890A, Hewlett Packard, Avondale, PA) equipped with a flame ionization detector. A 30 x 0.25mm i.d. fused silica capillary column (DB-225, J & W Scientific, Folsom, CA) operated with a helium carrier gas (linear velocity: 25 cm/sec, split ratio 20:1) was used for separation of the fatty acid methyl esters. The GC oven temperature was initially held at 175^{o}C for 10 min, then increased at a rate 1.5^{o}C/min to a final temperature of 200^{o}C and held for 40 min. The injector and detector temperature were held at 275^{o}C and 300^{o}C, respectively.

Identification of the fatty acid methyl esters was

based on comparison of retention times of samples to those of fatty acid methyl ester standards. Fatty acid distribution for each fraction was based on integration of peak areas (HP 3392A Integrator) and reported as weight percent of total methyl esters.

Hexanal Analysis

Determination of hexanal in chicken nuggets was achieved by purge and trap gas chromatography. A dynamic headspace concentrator (Model 4000, Tekmar Co. Cincinnati, OH) with a capillary interface (Model 1000) was used for the isolation of direct vapor or headspace volatile constituents.

A 1g meat slurry (prepared from 10g ground whole chicken nugget and 90g H_2O), 100ng 2-heptanone (internal standard) and 4ml H_2O were transferred to a 25ml sample holder and placed in a 80^{o}C water bath. The volatiles were removed from the sample (0.1g solids) by purging with purified helium gas at a flow rate of 40ml/min for 13 min and trapped on a Tenax trap (12 x 1/8 in) held at 30^{o}C.

To desorb the volatiles, the Tenax trap was heated to 180^{o}C and held for 6 min. The desorbed volatiles were concentrated at the top of the capillary column at the capillary interface, which was cooled to -150^{o}C with pressurized liquid nitrogen. The interface was then rapidly heated and backflushed to inject the volatiles directly onto a fluid methyl silicone capillary column (25m x 0.2mm i.d.,

HP 101, Hewlett Packard) installed in the gas chromatograph The GC was operated using a temperature program which started at 15°C for 15 min and increased to 40°C at the rate of 2.5°C/min, then increased at 10°C/min to a final temperature 220°C and held for 30 min. Injector and detector temperature were set at 115°C and 300°C, respectively. The flow rate of the helium carrier gas was 0.5ml/min. Identification and quantitation of hexanal in the meat sample was based on comparisons of retention times and peak areas of a hexanal standard.

Sensory Evaluation

Precooked, restructured chicken nuggets were evaluated for warmed-over flavor (WOF) after refrigerated and frozen storage by a ten-member sensory panel. All judges had previous training and experience in evaluating warmed-over flavor in meat. Testing sessions were held using partitioned booths equipped with standard indoor fluorescent lighting.

Chicken nuggets were portioned (ca 12g each) and served to the panelists in covered petri dishes coded with 3-digit random numbers. The samples was heated for 15 sec to 70°C in a microwave oven immediately before serving. A six point scale for degree of WOF (0 = no WOF and 5 = very strong WOF) was used (Appendix A). Vacuum packaged chicken nuggets containing STPP and TBHQ were served as reference samples and were considered as "no WOF".

Rancimat Method

The induction times of heated soybean oil samples were obtained with a 617 Rancimat (Metrohm AG, CH-9100 Heriaus, Switzerland). The sample (2.5g) was weighed out in the reaction vessel, which was then placed in the heating block for 10 min to preheat the sample. A 18-L/hr steam of air was bubbled through the sample which was held at 120^{o}C, and the volatile components were trapped in 50ml of distilled water, where the conductometric measurement of the induction times was made.

Statistical Analysis

The experiments were designed as a three factor (treatment x time x replication) complete randomized model with balanced data. Means, standard errors, sum of square, mean square error and correlation coefficient of data from all tests were calculated using the MSTATC microcomputer statistical program (Michigan State University, 1989).

Tukey's test was used to determine signification of means for multiple comparisons and Bonferroni t statistics were performed to analyze specific contrasts among treatments. Interactions of main effects and correlation between sensory scores and results from chemical analyses were interpreted according to Gill (1978).

RESULTS AND DISCUSSION

Evaluation of Lipid Quality in Chicken Meat

Preliminary tests were conducted to determine the lipid content and initial TBARS values of chicken meat. The lipid contents of different chicken tissues vary considerably and influence the oxidative potential of the tissues. TBARS values are the most commonly used index for quantitating the extent of oxidative rancidity in meats (Melton, 1983). To determine the quality (freshness) of the raw materials and to ascertain the degree of variation in initial source of meats, the TBA test was performed on the raw meat prior to processing.

The total lipid contents of chicken white meat, dark meat and mechanically deboned chicken (MDC) are presented in Table 4. The average lipid content of the MDC was 17.6% and was similar to previously reported values (Froning 1970; Uebersax et al., 1978). The lipid contents of the hand deboned white and dark meat were 2.6% and 7.2%, respectively. These values were higher than those found in the studies of Katz et al. (1966) and Pikul et al. (1985), who reported lipid contents of only 1% in white meat and 2.5% in dark meat.

This variation in results could be due to the different

Table 4. Total lipid contents of raw chicken meat.

Source of Meat	% Total Lipid[1]			
	Rep 1	Rep 2	Rep 3	Average
White Meat	2.8	2.5	2.4	2.6
Dark Meat	7.3	7.0	7.1	7.2
MDCM[2]	17.2	17.6	17.9	17.6

[1] Sampls were taken from each of three replications and analyzed in duplicate.
[2] MDCM: Mechanically deboned chicken meat.

Table 5. TBARS values of raw chicken meat prior to processing.

Source of Meat	TBARS Values[1,2]			
	Rep 1	Rep 2	Rep 3	Average
White Meat	0.15	0.15	0.14	0.15
Dark Meat	0.24	0.21	0.22	0.22
MDCM[3]	0.27	0.31	0.32	0.30

[1] Sampls were taken from each of three replications and analyzed in duplicate.
[2] mg malonaldehyde/kg sample.
[3] MDCM: Mechanically deboned chicken meat.

methods used to extract the lipids from the meat samples. Maxwell et al. (1980) reported more complete extraction of phospholipids by the dry column method. Crackel (1988a) also found that the lipid contents of restructured beef steaks determined by the dry column procedure were higher than those obtained by the AOAC method (procedure 24.005) Lin (1988) reported 2.2% fat in white meat and 7.9% fat in dark meat using the dry column method, thus confirming that the higher lipid contents of the chicken meats obtained in the present study were due to the more complete lipid extraction by the dry column procedure.

The TBARS values of the raw chicken meat from each replicated experiment are summarized in Table 5. TBARS values of dark meat were higher than those obtained for white meat. This observation is due to the higher lipid content in dark meat and is compatible with the findings of other researchers (Igene et al., 1980; Pikul et al., 1985). Among all types of meat, the TBARS values of the MDC were the highest. The high oxidative potential of the MDC results from not only its compositional nature (lipids, bone marrow and heme), but also from the extreme stress and aeration to which it is subjected during its processing (Dawson and Gartner, 1983).

On the other hand, the TBARS values for all raw samples from all replicates were lower than 0.32, indicating no substantial oxidation occurred after slaughtering and subsequent handling. It is essential to assure that the

meat is as fresh as possible in order to evaluate the effects of processing variables on the quality of the meat products. In addition, very little variation in TBARS values within replicates was observed which represented consistency in the sources of meats.

Effects of Antioxidants on Chicken Nuggets

Restructured chicken nuggets were used to evaluate the antioxidant effects of oleoresin rosemary (OR), sodium tripolyphosphate (STPP) and tertiary butylhydroquinone (TBHQ). Chicken nuggets formulated to 7% fat were prepared from a combination of white (50%) and dark (30%) meat and mechanically deboned chicken meat (20%) as described previously. The effects of these antioxidants on lipid quality of the chicken nuggets during the cooking process and storage will be discussed, respectively.

Changes by Cooking Process

The breaded chicken nuggets were fried to an internal temperature of 76^{o}C in soybean oil. Measurements of total weight and TBARS values of the chicken nuggets were performed before and after frying to evaluate cooking yields and degrees of oxidation arising from the cooking process.

Results of total weight changes and cooking yields of chicken nuggets are presented in Table 6. There were no significant differences in total weight of chicken nuggets

Table 6. Total weight changes and cooking yields of chicken nuggets after frying.

Treatment	Total Weight[1](g) Before frying	After frying	Cooking Yields[2]
Salt	21.3	20.9	98.1[a]
Salt/OR (0.05%)	21.6	21.4	99.1[ab]
Salt/OR (0.1%)	21.1	20.8	98.6[a]
Salt/STPP	21.4	21.5	100.5[b]
Salt/STPP/OR (0.05%)	20.8	21.0	101.0[b]
Salt/STPP/OR (0.1%)	21.1	21.3	101.0[b]
Salt/STPP/TBHQ	21.0	21.3	101.4[b]
Average	21.1	21.2	99.96

[1] All values represent the average of three replicated experiments.
[2] All numbers in the same column bearing the same superscript do not differ significantly at $p < 0.05$.

Table 7. Changes in TBARS values of chicken nuggets during frying.

Treatment	TBARS Values[1,2,3] Before frying	After frying
Salt	0.39[a]	1.67[a]
Salt/OR (0.05%)	0.31[ab]	1.28[ab]
Salt/OR (0.1%)	0.33[ab]	0.94[ab]
Salt/STPP	0.26[ab]	0.64[ab]
Salt/STPP/OR (0.05%)	0.24[ab]	0.50[b]
Salt/STPP/OR (0.1%)	0.26[ab]	0.46[b]
Salt/STPP/TBHQ	0.17[b]	0.38[b]

[1] All values represent the average of three replicated experiments.
[2] mg malonaldehyde/kg sample.
[3] All numbers in the same column bearing the same superscript do not differ significantly at $p < 0.05$.

among treatments before frying. However, chicken nuggets prepared with STPP showed higher cooking yields than those processed without STPP. It has been reported that alkaline phosphates can improve processing yield, increase juiciness and enhance texture and palatability scores of restructured meat (Shults et al., 1972; Shults and Wierbicki, 1973; Theno et al., 1978; King et al., 1986). Phosphates not only cleave the actomyosin complex formed at rigor into components with increased water holding capacity but also alter the ionic strength of the sarcoplasm. This increases the electrostatic repulsions between muscle filaments, thus increasing the amount of space available for water binding (Shults and Wierbicki, 1973; Neer and Mandigo, 1977; Huffman et al., 1981a).

Phosphates also were effective in retarding oxidation of the chicken nuggets during the frying process (Table 7). Before frying, the TBARS values of all samples were approximately the same level, except those for the control (salt alone) and TBHQ samples. After frying, TBARS values for all samples were greatly enhanced. The samples without STPP treatment showed 3-4 fold increases, while those chicken nuggets prepared with STPP exhibited only a 2 fold increase after frying.

Several model system studies have demonstrated that nonheme iron, rather than heme iron, is the active catalyst responsible for the rapid oxidation of cooked meat (Sato and Hegarty, 1971; Love and Pearson, 1974; Tichivangana and

Morrissey, 1985). Igene et al. (1979a) further concluded that heme pigments serve as a source of free iron, being readily broken down during the cooking process and can catalyze autoxidation.

Phosphates function as antioxidants by chelating metal ions. However, most chelates are unstable at high temperature (Labuza, 1971). Roozen (1987) also found that the effect of chelating agents was restricted to the oxidation reaction during cold storage. The results from the current study confirmed that there was no significant protective effect of phosphates during heating.

Some investigators have found that phosphates are effective in retarding lipid oxidation of poultry meat during the cooking process (Thomson, 1964; Ang and Hamm, 1986; King and Earl, 1988). However, the amount of phosphates used in their studies (0.5 - 3%) were higher than that used (0.3%) in the present investigation. It should be pointed out that the relatively large amount of STPP affected the pH of the model system and this can influence the results obtained (Chen and Waimaleongora-ek, 1981; Trout and Schimidt, 1984). A more direct influence of phosphate concentration may be the increase in the water holding capacity, thus diluting the lipid content in meat.

As mentioned previously, samples containing STPP had slightly higher cooking yields than those processed without STPP. However, there was no significant difference in lipid contents among processing treatments in the chicken nuggets

prepared with or without breading (Table 8). This indicated that the effect of phosphates in slowing the rate of lipid oxidation during heating is by chelating metal ions rather than by holding water and diluting the fat content of the meat.

Table 8. Total lipid contents[1] in chicken nuggets after frying.

Treatment	Total Lipid Content (%) With breading[2]	Without breading[3]
Salt	12.8	9.4
Salt/OR(0.05%)	12.7	9.1
Salt/OR (0.1%)	12.7	9.2
Salt/STPP	13.0	8.3
Salt/STPP/OR (0.05%)	13.1	8.5
Salt/STPP/OR (0.1%)	13.2	8.8
Salt/STPP/TBHQ	13.1	8.6
Average	12.9	8.8

[1] All values represent the average of three replicated experiments.
[2] Total lipids taken from whole chicken nuggets.
[3] Total lipids taken from chicken nuggets after removing breading.

Although STPP did not differ significantly from the control, when combined with other antioxidants substantial lowering of TBARS values ($P < 0.05$) was obtained (Table 7). Oleoresin rosemary (OR) produced results similar to those obtained with STPP. The antioxidant effect of OR, when used

alone, was minimal during the cooking process until combined with STPP.

Lipid Stability During Refrigerated Storage

A modified TBA distillation method in which an aqueous TBA reagent was used instead of an acid solution was utilized in this study to monitor the oxidative changes of chicken nuggets during storage. Some investigators have recommended the addition of antioxidants to the meats during sample preparation in order to minimize artifactual results arising from sample oxidation (Rhee, 1978; Pikul, et al., 1983). In a preliminary study, the ability of PG and EDTA to eliminate further oxidation of the chicken meat during the TBA test was demonstrated. Thus, a 0.5% PG/EDTA (1:1) solution was added to the chicken nuggets before the blending process.

In the refrigerated study, analysis of variance (ANOVA) of the TBARS values and sensory scores was performed based on a three factor model with balanced data. The ANOVAs (Tables 9 and 10) of the results from the TBA tests and sensory evaluation both showed significant effects ($P < 0.01$) of treatments, days (storage times) and replications as well as significant interactions ($P < 0.01$) of each combination from these factors. Because only the fixed effects (treatments and times) received the major emphasis and interacted significantly, the tests of the treatment effect and the time effect were performed conditionly, i.e.,

Table 9. Analysis of variance for TBARS values of chicken nuggets duting refrigerated study.

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
Treatment (T)	8	100.86	12.61	107.22	< 0.01
Day (D)	3	62.22	20.74	176.41	< 0.01
Replication (R)	2	28.39	14.20	120.74	< 0.01
TD	24	16.24	0.68	5.76	< 0.01
TR	16	6.27	0.39	3.33	< 0.01
DR	6	7.20	1.20	10.20	< 0.01
Error (TDR)	48	5.64	0.12		
Total	107	226.83			

Table 10. Analysis of variance for sensory scores of chicken nuggets during refrigerated study.

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
Treatment (T)	8	46.46	5.81	47.39	< 0.05
Day (D)	3	53.24	17.75	144.82	< 0.01
Replication (R)	2	0.85	0.42	3.45	< 0.01
TD	24	5.20	0.22	1.77	< 0.01
TR	16	4.83	0.30	2.46	< 0.01
DR	6	6.07	1.01	8.26	< 0.05
Error (TDR)	48	5.88	0.12		
Total	107	122.53			

the contrasts for the treatments were calculated separately within each level of storage times.

As shown in Table 11 and Figures 3 and 4, the TBARS values for all samples increased over time, with the control (salt only) oxidizing most rapidly and to the greatest extent. The TBARS values for all antioxidant-treated samples were lower than the control values throughout the entire storage period. Although each antioxidant offered protection, the STPP/TBHQ treatment was most effective in retarding salt-catalyzed oxidation as shown by the nearly horizontal lines in Figure 4.

There were no significant differences in TBARS values over the storage times for the STPP, STPP/OR (0.05%), STPP/OR (0.1%) and STPP/TBHQ-treated samples, indicating that all four treatments with STPP, when used alone or combined with other antioxidants, effectively prevented lipid oxidation over six days of storage at 4^{o}C. However, samples processed with OR (0.05% and 0.1%) but without STPP showed significant increases in TBARS values ($P < 0.05$) after 2 and 4 days of storage, respectively. These results showed that STPP provided better protection against autoxidation than OR after 2 days refrigerated storage.

The TBARS values of the samples containing STPP and fried in soybean oil containing OR (0.025% and 0.05%) were relatively constant at the begining of the storage period. However, both of these two treatments had a significant

Table 11. TBARS values[1,2,3] of chicken nuggets during refrigerated storge.

Treatment	Day 0	Day 2	Day 4	Day 6
Salt	1.67^{c}	3.29^{b}	4.36^{ab}	5.13^{a}
Salt/OR (0.05%)	1.28^{c}	2.76^{b}	4.05^{a}	4.57^{a}
Salt/OR (0.1%)	0.94^{c}	2.15^{bc}	3.19^{ab}	3.75^{a}
Salt/STPP	0.64^{a}	1.26^{a}	2.36^{a}	2.65^{a}
Salt/STPP/OR (0.05%)	0.50^{a}	0.97^{a}	1.65^{a}	1.78^{a}
Salt/STPP/OR (0.1%)	0.46^{a}	0.75^{a}	1.20^{a}	1.47^{a}
Salt/STPP/TBHQ	0.38^{a}	0.47^{a}	0.62^{a}	0.59^{a}
Oil + OR[4] (0.025%)	0.58^{b}	1.11^{ab}	1.66^{ab}	2.43^{a}
Oil + OR (0.05%)	0.64^{b}	1.40^{ab}	2.13^{a}	2.56^{a}

[1] All values represent the average of three replicated experiments.
[2] mg malonaldehyde/kg sample.
[3] All numbers in the same row bearing the same superscript do not differ significantly at $p < 0.05$.
[4] Samples fried in oil containing OR contained salt and STPP.

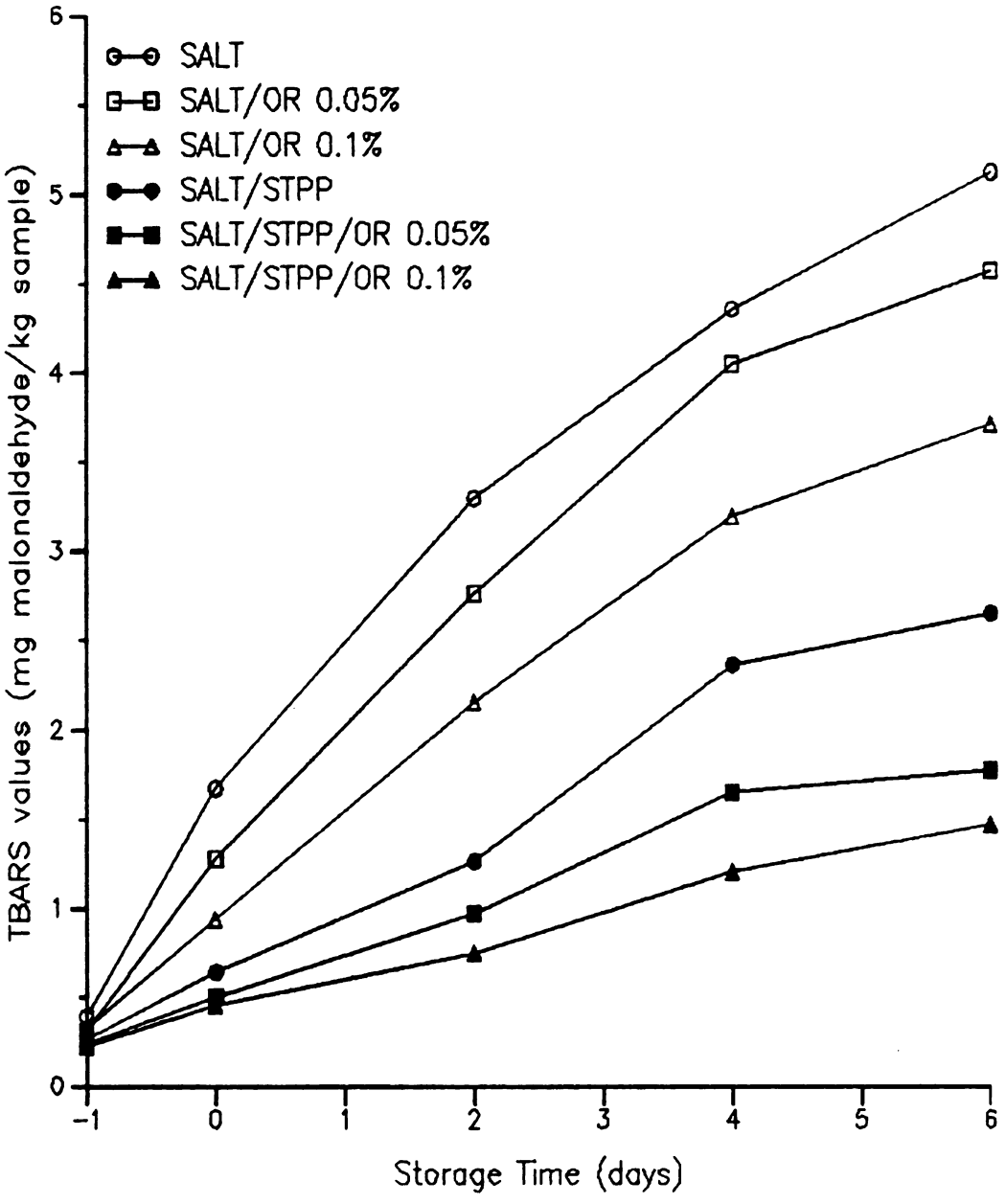


Figure 3. Effects of oleoresin rosemary (OR) and sodium tripolyphosphate (STPP) on TBARS values of chicken nuggets during refrigerated storage.

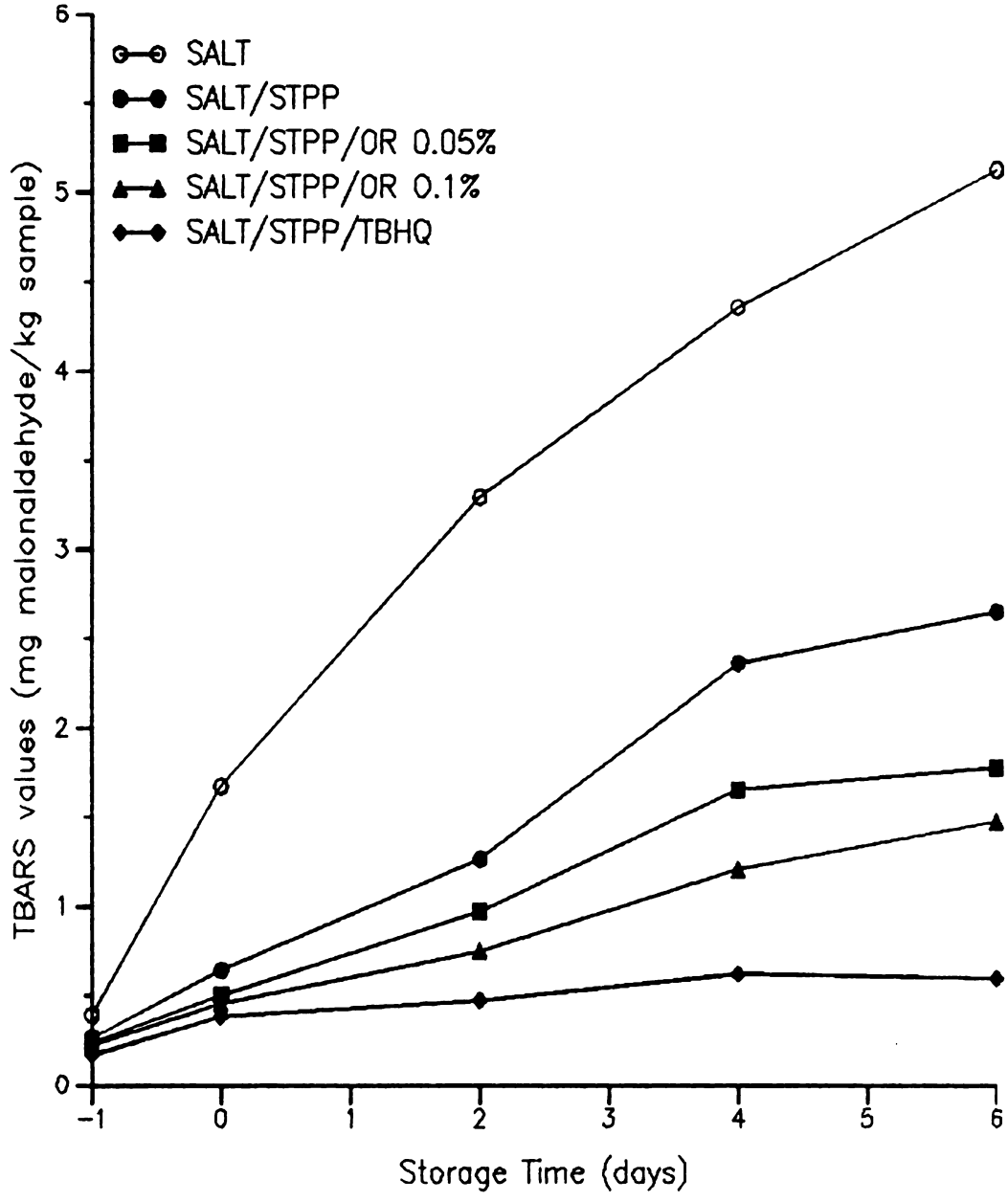


Figure 4. Effects of oleoresin rosemary (OR) and tertiary butylhydroquinone (TBHQ) on TBARS values of chicken nuggets during refrigerated storage.

increase ($P < 0.05$) in TBARS values after 6 days of refrigerated storage.

The corresponding sensory scores are presented in Table 12. As for the TBARS values, sensory scores for all treatments increased over time. The most intense WOF was detected in the control samples (salt alone) at each test day by the sensory panel. Significant increases ($P < 0.05$) in WOF was found in both the control samples and those treated with 0.05% OR after 2 days. There was no difference in the degree of WOF in the nuggets after 2 days and 6 days of storage, perhaps due to the rapidly developed WOF which was already very strong after 2 days. This strong off-flavor prevented the panel from accurately assessing the varying degrees of oxidation among storage periods.

On the other hand, the sensory panel seemed to be more sensitive to the varying degrees of WOF in the less oxidized samples. Samples containing STPP, alone or in combination with other antioxidants, showed no significant differences in TBARS values after 6 days storage. However, significant increases ($P < 0.05$) in the intensity of WOF were detected by sensory panelists after 4 days at $4^{o}C$. Another difference between TBARS and sensory data was that there was no significant difference in the sensory scores throughout the entire storage period in the samples containing STPP and which were fried in oil containing OR (0.025% and 0.05%). It is possible that flavor from OR was carried through the oil to the nuggets and masked the oxidized flavor.

Table 12. Sensory scores[1,2,3] of chicken nuggets during refrigerated storge.

Treatment	Month 0	Month 2	Month 4	Month 6
Salt	1.17^{b}	2.92^{a}	3.53^{a}	3.77^{a}
Salt/OR (0.05%)	1.38^{b}	2.98^{a}	3.37^{a}	3.69^{a}
Salt/OR (0.1%)	1.50^{c}	2.43^{bc}	3.65^{ab}	3.93^{a}
Salt/STPP	0.93^{b}	2.00^{ab}	2.37^{a}	2.31^{a}
Salt/STPP/OR (0.05%)	0.32^{b}	1.31^{ab}	1.70^{a}	2.23^{a}
Salt/STPP/OR (0.1%)	0.63^{b}	1.36^{ab}	1.80^{a}	2.06^{a}
Salt/STPP/TBHQ	0.30^{b}	1.08^{ab}	1.27^{a}	1.73^{a}
Oil + OR[4] (0.025%)	0.93^{a}	1.54^{a}	1.97^{a}	2.37^{a}
Oil + OR (0.05%)	0.73^{a}	2.00^{a}	2.17^{a}	2.60^{a}

[1] All values represent average of three replicated experiments.
[2] Based on a scale of 0 to 5; 0 corresponds to no WOF and 5 to a very strong WOF.
[3] All numbers in the same row bearing the same superscript do not differ significantly at $p < 0.05$.
[4] Samples fried in oil containing OR contained salt and STPP.

To deterimine if the aroma or flavor from rosemary was detectable in the samples, panelists were asked if they could detect any "non-meat" flavor in the samples while evaluating the extent of WOF in the samples. However, there was no evidence of "non-meat" flavor in any sample at each sampling period.

To evaluate the treatment effect in the TBARS values and sensory scores, specific contrasts were performed at each storage time and are summarized in Table 13. The result of contrast 1 from both the TBA test and sensory evaluation again demonstrated that STPP had a protective effect on chicken nuggets during refrigerated storage. This result is in agreement with the findings of Roozen (1987), who reported that the strong antioxidant effect of STPP in meat systems was mainly exhibited in low temperature storage rather than in the heating process.

STPP functions as an antioxidant in muscle foods by chelating prooxidant metals such as ferrous iron (Tims and Watts, 1958; Love and Pearson, 1974; Igene et al., 1979a). Labuza (1971) reported that such complexes are less stable at higher temperatures. As discussed previously, STPP, when used alone, did not show a significant protective effect in chicken nuggets during the frying process. The different effects of STPP in retarding lipid oxidation on heating and during refrigerated storage were demonstrated in this study.

Contrast 2 (Table 13) verified that the STPP/TBHQ treatment provided more effective protection compared to

Table 13. Significance levels of designed contrasts for treatment effects on TBARS values and sensory scores of chicken nuggets in the refrigerated study.

Contrast	Test	Test Day 0	2	4	6
(1) STPP vs No STPP	TBA	0.01	0.01	0.01	0.01
	Sensory	0.01	0.01	0.01	0.01
(2) STPP vs STPP/TBHQ	TBA	ns	0.01	0.01	0.01
	Sensory	ns	0.05	0.05	ns
(3) STPP/OR 0.05% vs STPP/TBHQ	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(4) STPP/OR 0.01% vs STPP/TBHQ	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(5) OR Linear Effect (OR_L)	TBA	0.01	0.01	0.01	0.01
	Sensory	ns	ns	ns	ns
(6) OR Quadratic Effect (OR_Q)	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(7) Interaction STPP & OR_L	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(8) Interaction STPP & OR_Q	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(9) Oil with OR Linear Effect	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(10) Oil with OR Quadratic Effect	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns

ns: Not significant

STPP alone. Significant differences ($P < 0.01$) in the TBARS values were found after 2 days of storage. The differences in the sensory scores also became significant ($P < 0.05$) after the 2nd day, although no apparent differences were detected at the 6th day.

TBARS and sensory data both indicated that TBHQ has a excellent antioxidant effect when used by itself. The effectiveness of TBHQ in meat products has been investigated by Chastain et al. (1982) and Crackel et al. (1988a). Its activity has been attributed to its effective resonance forms and its ability to donate a proton to suppress the formation of lipid peroxy radicals in the propagation step of oxidation (Labuza, 1971; Roozen, 1987).

Although the STPP/TBHQ treatment appeared to have lower TBARS values and sensory scores than the STPP/OR treatments, results of contrasts 3 and 4 showed no significant differences between STPP/TBHQ and STPP/OR (both 0.05% and 0.1%) treatments (Table 13). This indicated that STPP/OR is equally as effective as STPP/TBHQ in retarding lipid oxidation. The antioxidant properties of oleoresin rosemary have been attributed to a variety of isoprenoid quinones (Wu et al., 1982; Houlihan et al., 1984; 1985) which are capable of terminating free radical reactions and quenching reactive oxygen species (Richardson and Dahl, 1983).

Comparable antioxidant effects of OR to synthetic phenolic antioxidants have been demonstrated by other researchers. Barbut et al. (1985) studied the antioxidant

effect of OR and a commercial blend of BHA/BHT/citric acid in turkey breakfast sausages prepared with 75% hand deboned turkey meat and 25% mechanically deboned turkey meat during 16 days refrigerated storage. They reported no differences in TBARS values and sensory scores between the two antioxidant treatments.

Sheu (1988) evaluated the effects of OR and STPP in prerigor, precooked, vacuum-packaged pork patties during refrigerated storage. He reported that the combination of STPP and OR (0.05%) provided a better protective effect than Tenox^{TM} II (BHA/PG/citric acid) in cooked pork patties during 48 days storage. Resurreccion and Reynolds (1989) also found that OR was as effective as BHA/BHT in retarding lipid oxidation in chicken/pork fankfurters which were vacuum packaged and refrigerated for 35 days.

In the current study, a strong linear effect between OR treatments from their TBARS values was found at each test day ($P < 0.01$), which indicated the effectiveness of OR increased as its concentration increased. Furthermore, no significant interaction between STPP and OR was obtained for either TBARS values or sensory scores. These findings demonstrated that there was an additive effect between STPP and OR , although no synergism was found in the two antioxidants.

In the study of Sheu (1988), however, a pronounced synergism was obtained in the combination of STPP and OR (0.025% and 0.05%) in cooked pork patties. Different

results from the present study could be due to the different meat systems used. In Sheu's study, pork patties were processed with prerigor pork loins (within one hour after slaughtering) and were vacuum packaged, thus minimizing the factors influencing oxidation of meats.

The chicken meats used for the manufacture of the chicken nuggets were purchased approximately 12 hours after slaughtering. Although the TBARS values of the meats before processing were low (Table 5), it is belived that some oxidation may have already taken place in the meat. Poultry meat, especially mechanically deboned chicken meat is very susceptible to oxidation.

OR contains phenolic compounds and thus acts as an antioxidant by interrupting the free radical chain in the propagation step of the oxidative process. Phosphates function by chelating metal ions which have been suggested to be the major catalyst in nonenzymatic oxidation (Love, 1983), thus retarding the lipid oxidation in the initiation step. The metal ions could be naturally present or be released from heme compounds by cooking. The use of simplified meat models and well-controlled processing conditions may result in the clarification of the interactions between OR and STPP.

The results of constrast 9 and 10 (Table 13) indicated no beneficial effects of adding OR to the oil on the oxidative stability of the nuggets. These results imply that either the antioxidant principles in OR were stripped

from the oil during the cooking of the nuggets or that the OR did not penetrate into the chicken meat during storage. TBA analyses were carried out with the whole nugget, i.e., breading and meat were blended together before samples were taken for analysis. It is most likely that the meat lipids would deteriorate more rapidly than the vegetable oil absorbed by the breading of the nuggets because of their closer contact with the non-heme iron in the meat.

Lipid Stability During Frozen Storage

The effectiveness of OR, STPP and TBHQ in stabilizing lipids in chicken nuggets during frozen storage was evaluated. TBA test, sensory evaluation, fatty acid analysis and quantitation of hexanal were performed to monitor the influence of antioxidants over 6 months of frozen storage.

(1) TBA Test and Sensory Evaluation

Analyses of variances (ANOVAs) of TBARS values and sensory scores were performed based on a three factor model as previously described for the refrigerated study. Strong treatment and time effects ($P < 0.01$) as well as significant interactions between treatments and times were found.

Generally, the results of the TBARS values and sensory scores of the frozen nuggets were similar to those from the refrigerated study. As shown in Table 14 and Figures 5 and 6, all antioxidant treatments yielded lower TBARS values

Table 14. TBARS values[1,2,3] of chicken nuggets during frozen storge.

Treatment	Month 0	Month 2	Month 4	Month 6
Salt	1.67^{b}	3.29^{a}	3.65^{a}	3·61^{a}
Salt/OR (0.05%)	1.28^{b}	2.73^{a}	3.24^{a}	3.57^{a}
Salt/OR (0.1%)	0.94^{b}	2.44^{a}	2.85^{a}	2.78^{a}
Salt/STPP	0.64^{c}	1.15^{bc}	1.99^{a}	1.58^{ab}
Salt/STPP/OR (0.05%)	0.50^{a}	0.98^{a}	1.35^{a}	1.35^{a}
Salt/STPP/OR (0.1%)	0.46^{b}	0.99^{ab}	0.97^{ab}	1.19^{a}
Salt/STPP/TBHQ	0.38^{a}	0.52^{a}	0.48^{a}	0.54^{a}
Oil + OR[4] (0.025%)	0.58^{b}	1.27^{a}	1.51^{a}	1.59^{a}
Oil + OR (0.05%)	0.64^{b}	1.30^{ab}	2.11^{a}	1.82^{a}

[1] All values represent the average of three replicated experiments.
[2] mg malonaldehyde/kg sample.
[3] All numbers in the same row bearing the same superscript do not differ significantly at $p < 0.05$.
[4] Samples fried in oil containing OR contained salt and STPP.

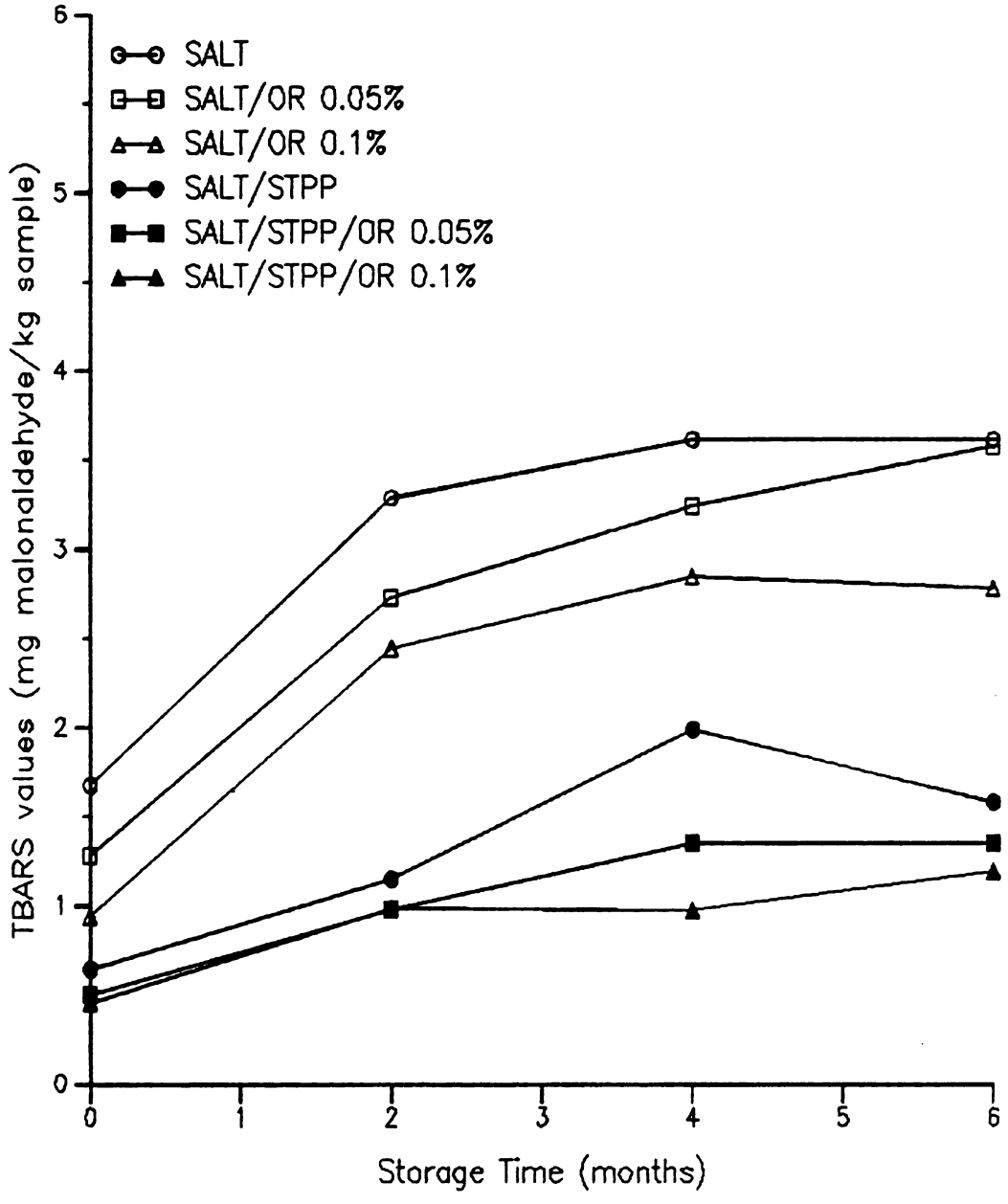


Figure 5. Effects of oleoresin rosemary (OR) and sodium tripolyphosphate (STPP) on TBARS values of chicken nuggets during frozen storage.

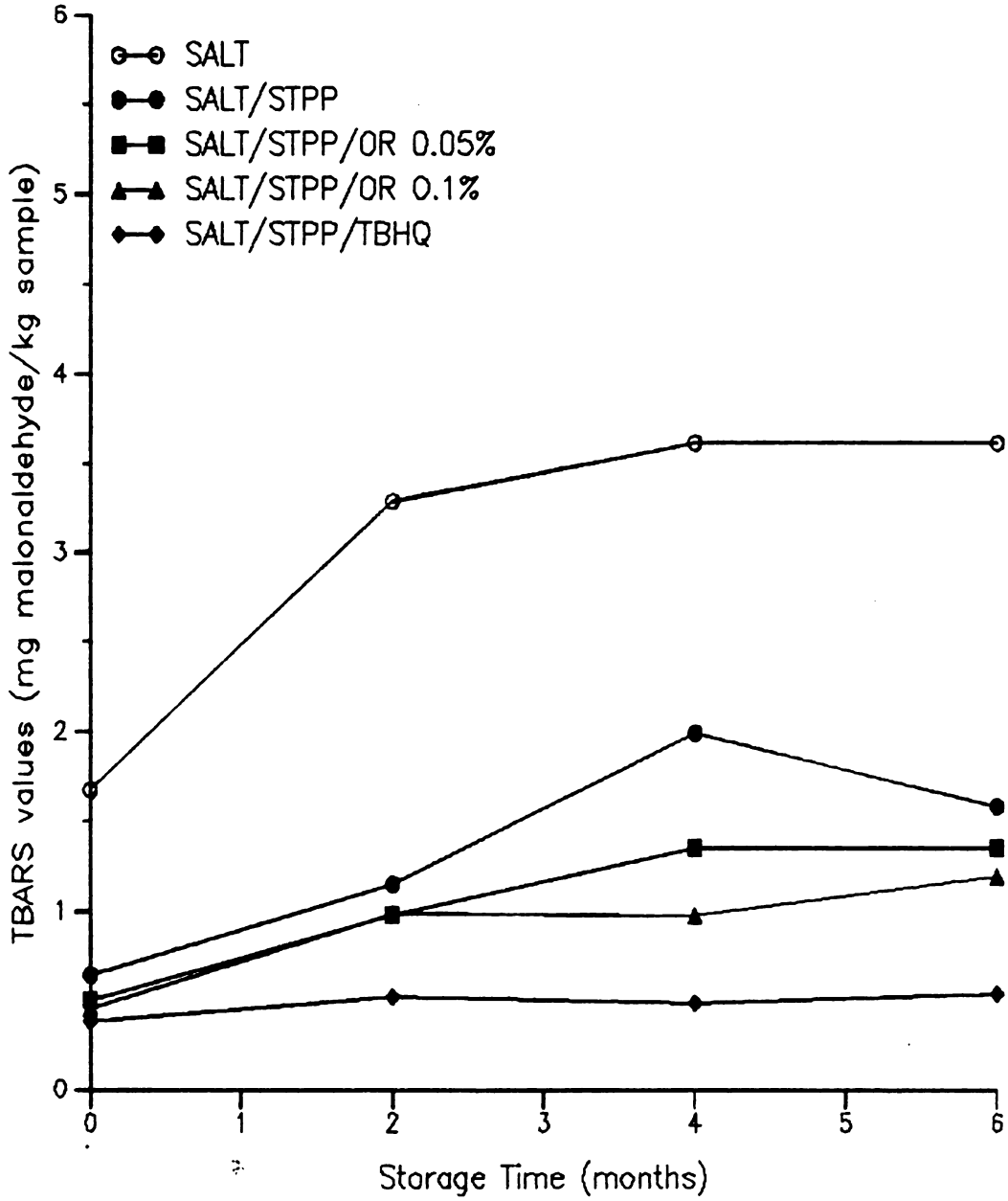


Figure 6. Effects of oleoresin rosemary (OR) and tertiary butylhydroquinone (TBHQ) on TBARS values of chicken nuggets during frozen storage.

than did the control samples (salt only). The STPP/TBHQ treatment again offered the most effective protection against salt-catalyzed oxidation.

TBARS values in samples processed without STPP, including the salt control and samples containing OR (0.05% and 0.1%), significantly increased ($P < 0.05$) after 2 months of frozen storage and then did not change significantly for the rest of the storage period. All chicken nuggets processed with STPP exhibited a slower increase in TBARS values compared to those without STPP. Unlike the results of the refrigerated storage in which no significant differences between TBARS values among storage periods were found in all STPP-treated samples, there were some variations in TBARS values of STPP- and STPP/OR 0.1%-treated samples during frozen storage.

The different effects of storage time on TBARS values between refrigerated and frozen storage may be due to the slower rate of oxidation and the decline in TBARS values after 4 months of frozen storage (Figure 5). The apparent loss of malonaldehyde or other TBA-reactive substances after several months of frozen storage is not unusual and has been reported previously (Tarladgis and Watts, 1960; Igene et al, 1979b; Gokalp et al., 1983). The decrease in TBARS values during frozen storage could be due to the interaction of malonaldehyde and amino groups (Buttkus, 1967; Gokalp et al., 1983; Smith, 1987). This implies that the TBA test may not be totally accurate in measuring oxidative products

during long-term frozen storage and should be compared with sensory evaluation and other chemical methods.

Results of the sensory evaluation of chicken nuggets during frozen storage are presented in Table 15. Generally, the changes of sensory scores over storage times were in agreement with those of TBARS values for all treatments. This is in contrast to the refrigerated study and is probably due to the smaller TBARS values observed during frozen storage. The greatest TBARS values in frozen storage was only 3.6, in comparison to 5.1 in refrigerated storage. As previously discussed, the sensory panel was more sensitive to varying degrees of WOF in the less oxidized samples and was unable to distinguish differences in the intensity of oxidized flavor in samples having TBARS values greater than 3.5.

The designed contrasts for investigating the treatment effect from the results of the TBA test and sensory evaluation are summarized in Table 16. The protective effect of STPP again was demonstrated ($P < 0.01$) by the results of the TBA test and sensory evaluation obtained at each analytical period. This result was similar to that found in the refrigerated study, and implied that STPP functions in the same manner under both storage conditions.

Contrast 2 (Table 16) for TBARS values and sensory scores showed a significant difference ($P < 0.05$) between STPP-treated and STPP/TBHQ-treated samples after 4 months of frozen storage. The difference between these two

Table 15. Sensory scores[1,2,3] of chicken nuggets during frozen storge.

Treatment	Month 0	Month 2	Month 4	Month 6
Salt	1.17^{b}	4.38^{a}	3.67^{a}	4.33^{a}
Salt/OR (0.05%)	1.38^{b}	3.82^{a}	4.00^{a}	4.17^{a}
Salt/OR (0.1%)	1.50^{b}	3.38^{a}	3.90^{a}	4.00^{a}
Salt/STPP	0.93^{b}	2.15^{ab}	2.93^{a}	2.40^{ab}
Salt/STPP/OR (0.05%)	0.32^{b}	1.82^{ab}	2.03^{ab}	2.40^{a}
Salt/STPP/OR (0.1%)	0.63^{a}	1.90^{a}	1.90^{a}	2.33^{a}
Salt/STPP/TBHQ	0.30^{b}	1.35^{a}	1.37^{a}	1.57^{a}
Oil + OR[4] (0.025%)	0.93^{a}	2.03^{a}	2.87^{a}	2.77^{a}
Oil + OR (0.05%)	0.73^{b}	2.48^{ab}	2.40^{ab}	2.93^{a}

[1] All values represent the average of three replicated experiments.
[2] Based on a scale of 0 to 5, 0 corresponds to no WOF and 5 to a very strong WOF.
[3] All numbers in the same row bearing the same superscript do not differ significantly at $p < 0.05$.
[4] Samples fried in oil containing OR contained salt and STPP.

Table 16. Significance levels of designed contrasts for treatment effects on TBARS values and sensory scores of chicken nuggets in the frozen study.

Contrast	Test	Test Month 0	2	4	6
(1) STPP vs No STPP	TBA	0.01	0.01	0.01	0.01
	Sensory	0.01	0.01	0.01	0.01
(2) STPP vs STPP/TBHQ	TBA	ns	ns	0.05	0.05
	Sensory	ns	ns	0.05	ns
(3) STPP/OR 0.05% vs STPP/TBHQ	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(4) STPP/OR 0.01% vs STPP/TBHQ	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(5) OR Linear Effect (OR_L)	TBA	0.01	0.05	0.05	0.05
	Sensory	ns	ns	ns	ns
(6) OR Quadratic Effect (OR_Q)	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(7) Interaction STPP & OR_L	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(8) Interaction STPP & OR_Q	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(9) Oil with OR Linear Effect	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns
(10) Oil with OR Quadratic Effect	TBA	ns	ns	ns	ns
	Sensory	ns	ns	ns	ns

ns: Not significant

antioxidants was detected after only 2 days of refrigerated storage (Table 13). This result may be due to a better antioxidant effect of STPP at lower temperatures, or because of the slow rate of oxidation during frozen storage.

Although visual observation of the data indicated that STPP/TBHQ was the most effective antioxidant combination, no significant differences in TBARS values and sensory scores were found between the STPP/TBHQ and STPP/OR (0.05% and 0.1%) treatments after six months of storage (contrasts 3 and 4). These results indicated STPP/OR was comparable to STPP/TBHQ in suppressing lipid oxidation during frozen storage as well as refrigerated storage.

The linear concentration effect of OR ($P < 0.05$) was again observed, although it was not as pronounced as that obtained for the refrigerated study. No significant interaction between STPP and OR was found during frozen storage. However, an additive effect between STPP and OR was exhibited in frozen storage as in refrigerated storage.

Nuggets cooked in soybean oil containing OR were compared to those cooked in oil without OR. Results of the TBA test and sensory evaluation indicated no significant effect of incorporating OR in the oil. These results agree with those of the refrigerated study and may be due to the more rapid deterioration of meat lipids than the vegetable oil adsorbed by the breading on the nuggets. However, the addition of OR to frying oil would probably be beneficial in stabilizing the oil in other food products such as potato

chips and frozen french-fried potatoes.

(2) Fatty Acid Profiles

Lipids extracted from the meat of the chicken nuggets were separated into triacylglycerols and phospholipids prior to methylation. Fatty acid methyl esters from the triacylglycerol and phospholipid fractions were analyzed by gas chromatography and reported as a weight percent of the total methyl esters. No differences in individual fatty acids were apparent among treatments during frozen storage. Thus, only the changes in the fatty acid composition of the control (salt only), STPP/OR (0.05%), and STPP/TBHQ-treated samples during storage will be discussed .

The fatty acid composition of the triacylglycerol and phospholipid fractions of chicken nuggets are presented in Tables 17 and 18. The initial fatty acid profiles of these three treatments (0 month) were very similar and agreed with previously reported values (Moerck and Ball, 1974; Jantawat and Dawson, 1980; Igene et al., 1980; Igene et al., 1981; Cokalp et al., 1983, Pikul et al., 1985). The slight variability found between literature values and the data of the current study was mainly due to the muscle type analyzed. The lipids of the chicken nuggets were a composite of the lipids from white meat, dark meat and mechanical deboned meat, however, the proportions of the major fatty acids in each fraction remained the same.

Table 17. Fatty acid composition of the triacylglycerols in chicken nuggets stored at -20°C for six months[1,2,3].

Fatty Acid	Control		STPP/OR		STPP/TBHQ	
	0 mo	6 mo	0 mo	6 mo	0 mo	6 mo
14:0	0.6	0.7	0.7	0.7	0.6	0.7
16:0	18.5	18.7	18.6	19.0	18.7	19.2
16:1	3.3	3.6	3.4	3.7	3.4	3.6
17:0	0.2	0.2	0.2	0.2	0.2	0.2
18:0	4.5	4.5	4.5	4.4	4.7	4.5
18:1	40.0	43.5	40.6	42.8	40.4	41.4
18:2	29.9	26.1	29.1	26.6	28.8	27.6
18:3	2.0	1.7	1.9	1.9	2.0	1.9
20:0	0.2	0.2	0.1	0.1	0.2	0.2
20:1	0.4	0.4	0.4	0.3	0.4	0.3
20:2	0.1	0.1	0.1	0.0	0.1	0.0
20:3	0.2	0.1	0.2	0.1	0.2	0.1
20:4	0.1	0.1	0.2	0.2	0.2	0.2
Saturated	24.0	24.4	24.1	24.4	24.4	24.8
Unsaturated						
Total	76.0	75.6	75.9	75.6	75.6	75.1
Mono-	43.7	47.5	44.4	46.8	44.3	45.3
Di-	30.0	26.2	29.2	26.6	28.9	27.6
Poly[3]-	2.3	1.9	2.3	2.2	2.4	2.1

[1] All values represent the average of three replicated experiments.
[2] As percent fatty acid.
[3] Polyunsaturated fatty acids in this study are defined as fatty acids containing three or more double bonds.

Table 18. Fatty acid composition of the phospholipids in chicken nuggets stored at -20°C for six momths[1,2,3].

Fatty Acid	Control		STPP/OR		STPP/TBHQ	
	0 mo	6 mo	0 mo	6 mo	0 mo	6 mo
14:0	0.2	0.6	0.2	0.4	0.4	0.4
16:0	18.6	22.2	18.5	20.5	19.5	20.1
16:1	1.4	2.9	1.3	2.9	2.5	2.6
17:0	0.2	0.2	0.2	0.2	0.2	0.2
18:0	9.0	8.7	9.2	9.7	9.2	9.8
18:1	25.7	37.9	25.8	28.2	23.6	26.4
18:2	27.1	19.9	26.1	24.2	26.1	24.5
18:3	0.7	0.6	0.9	0.8	0.9	0.9
20:0	0.1	0.1	0.1	0.1	0.1	0.1
20:1	0.4	0.3	0.4	0.4	0.4	0.4
20:2	0.2	0.1	0.2	0.1	0.2	0.1
20:3	1.6	0.5	1.5	0.7	1.6	1.2
20:4	9.1	4.5	9.9	7.8	9.2	8.6
20:5	0.2	0.0	0.2	0.1	0.2	0.1
22:4	2.3	0.7	2.2	1.5	2.4	2.0
22:5	1.8	0.4	1.8	1.3	2.1	1.6
22:6	1.3	0.6	1.3	0.9	1.3	1.1
Saturated	28.1	31.8	28.2	31.0	29.4	30.6
Unsaturated						
Total	71.9	68.2	71.7	69.0	70.6	69.4
Mono-	27.5	41.1	27.5	31.6	26.5	29.5
Di-	27.3	20.0	26.3	24.3	26.3	24.6
Poly[3]-	17.1	7.1	17.9	13.1	17.8	15.4

[1] All values represent the average of three replicated experiments.
[2] As percent fatty acid.
[3] Polyunsaturated fatty acids in this study are defined as fatty acids containing three or more double bonds.

Approximately 95 to 97% of the fatty acids found in the triacylglycerol fraction were comprised of $C_{16:0}$, $C_{16:1}$, $C_{18:0}$, $C_{18:1}$, and $C_{18:2}$ acids. Very low levels of polyunsaturated fatty acids ($C_{18:3}$ - $C_{20:4}$) were found in the triacylglycerol fraction. On the other hand, the phospholipid fraction contained higher levels of $C_{18:0}$ and $C_{20:3}$ to $C_{22:6}$ polyunsaturated fatty acids than did the triacylglycerol fraction. The polyunsaturated fatty acid content in the phospholipid fraction was about 9-fold higher than that in the triacylglycerol fraction. Igene et al. (1981) reported that the phospholipids of chicken meat contained about 15 times more polyunsaturated fatty acids than the triacylglycerols. The lower amount of polyunsaturated fatty acids in the phospholipids of the chicken nuggets could be due to the presence of mechanical deboned chicken meat. Phospholipids extracted from this meat have been reported to contain less polyunsaturated fatty acids than the phospholipids from hand deboned chicken meat (Jantawat and Dawson, 1980).

Although the difference in the amount of saturated fatty acids in the triacylglycerols and phospholipids was small, the latter actually contained higher amounts of the saturated fatty acids. This observation agrees with the results of Igene et al. (1981), who reported that the percentage of saturated fatty acids in the triacylglycerols of chicken meat was lower than that in phospholipids of chicken meat and lower than that of beef triacylglycerols.

In general, the more unsaturated fatty acids in a lipid system, the greater the rate of its autoxidation (Badings, 1970). Thus, the higher susceptibility of chicken meat to oxidation relative to red meat is due not only to the higher amount of polyunsaturated fatty acids in the phospholipids but also to the greater unsaturation in the triacylglycerols in chicken meat.

Changes in the fatty acid composition of the triacylglycerols in chicken nuggets during frozen storage are shown in Table 17. Although total unsaturation of the control and antioxidant treatments decreased slightly during frozen storage, the difference in unsaturation among treatments was not significant. In addition, there were no apparent changes in the proportions of mono-, di- and polyunsaturated fatty acids in the antioxidant-treated samples after 6 months of storage. In the control samples, $C_{18:1}$ increased from its original values by 9% after 6 months, while $C_{18:2}$ and $C_{18:3}$ decreased from their original values by 13% and 17%, respectively. These results reflected the effect of the antioxidants on the stability of the unsaturated fatty acids, although the changes in unsaturation in the triacylglycerols during frozen storage were not so obvious.

Changes in the fatty acid profiles of phospholipids of chicken nuggets during frozen storage are shown in Table 18. A slight decrease in the percentage of unsaturated fatty acids during frozen storage was evident in all samples, with

the control samples exhibiting the greatest loss. El-Gharbawi and Dugan (1965) reported that the loss of unsaturated fatty acids in dried beef tissue lipids was more pronounced in the phospholipids than in the triacylglycerols during storage. Most of the variations in unsaturation during frozen storage or among treatments in chicken neuggets were due to alterations in the levels of $C_{18:1}$, $C_{18:2}$, $C_{20:4}$, and $C_{22:4}$ fatty acids. Whereas the relative proportion of mono-unsaturated fatty acids increased significantly in all samples during frozen storage, the level of polyunsaturated fatty acids declined markedly over the same period. The amounts of diunsaturated fatty acids remained relative constant from the 2nd month to the end of the storage period. These changes were in close agreement with results from other studies (Moerck and Ball, 1974; Igene et al., 1980; Igene et al., 1981).

Although polyunsaturated fatty acids, especially those with four or more double bonds, including $C_{20:4}$, $C_{20:5}$, $C_{22:4}$, $C_{22:5}$, and $C_{22:6}$, comprise only a small portion of the total fatty acids, these acids underwent the greatest oxidative deterioration during storage. Figure 7 graphically shows the loss of polyunsaturated fatty acids in the phospholipid fractions of chicken nuggets during frozen storage. A rapid decline in the polyunsaturated fatty acid content of the control samples was observed during frozen storage. The polyunsaturated fatty acids in the phospholipids decreased from their original value by 20%,

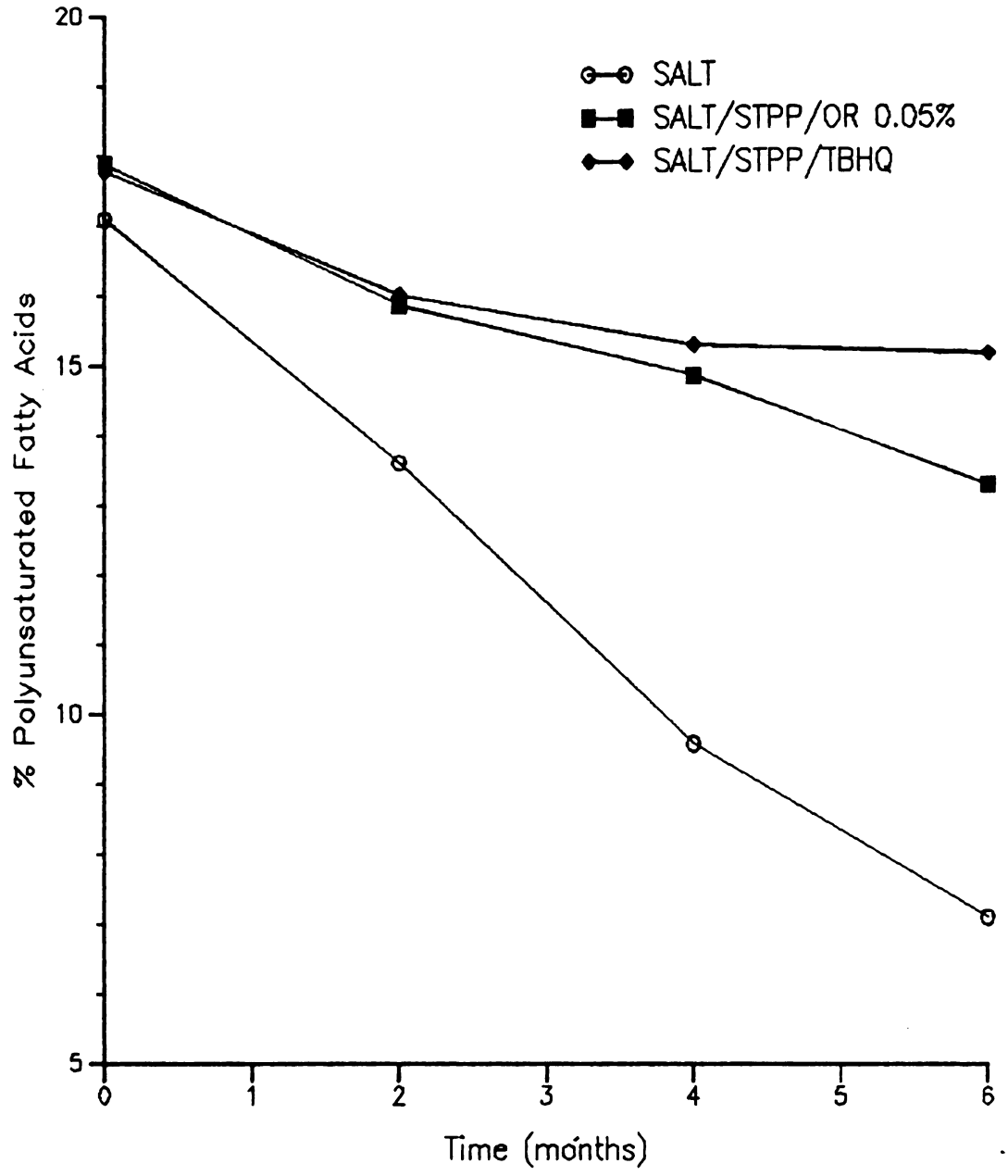


Figure 7. Changes in the percent polyunsaturated fatty acids in the phospholipid fraction of chicken nuggets after 2,4,6 months of frozen storage.

44%, and 58% after 2, 4, 6 months, respectively. The changes in the polyunsatured fatty acid content in the antioxidant-treated samples were more gradual than in the control samples. At the end of storage period, the loss of the polyunsaturated fatty acids in the phospholipids was only 27% and 14% in STPP/OR (0.05%) and STPP/TBHQ samples, respectively.

Pearson et al. (1977), Pearson and Gray (1983), and Pikul et al. (1985) have suggested that the abundance of polyunsaturated fatty acids in animal tissues makes these tissues highly susceptible to oxidation. Results from the fatty acid analyses indicated that the antioxidants (both STPP/OR 0.05% and STPP/TBHQ) effectively prevented the oxidative degradation of polyunsaturated fatty acids, thus retarding the development of WOF.

(3) GC Quantitation of Hexanal

Oxidative deterioration of polyunsaturated fatty acids, particularly $C_{18:2}$, $C_{18:3}$, $C_{20:4}$, was found in the chicken nuggets after 6 months of frozen storage. Quantitation of the volatile products formed by the degradation of unsaturated fatty acids during the storage of cooked meats has been used as a means of monitoring lipid oxidation in muscle foods (Vercellotti et al., 1987; Shadidi et al., 1987; St. Angelo et al., 1987, 1988). Consequently, quantitation of hexanal, a secondary product of linoleic

acid oxidation, has been utilized to monitor lipid oxidation in meat systems (St. Angelo et al., 1988).

The hexanal levels in the chicken nuggets after 6 months at -20°C ranged from 0.80 ppm to 18.55 ppm. As shown in Figure 8, the highest hexanal content was found in the control samples (salt only), followed by samples treated with OR 0.05%, OR 0.1% and STPP. Hexanal production in the nuggets was reduced 13%, 47% and 80% by the addition of OR 0.05%, OR 0.1% and STPP, respectively. Although STPP was more effective than OR in preventing hexanal formation, OR also showed a significant antioxidant effect ($p < 0.05$).

Love and Pearson (1976) reported that the addition of STPP caused a 50% decrease in hexanal concentration in a model meat system. Shahidi et al. (1987) suggested that the hexanal content was a good indicator of oxidative stability in cooked ground pork and found 62% less hexanal in pork samples containing 3000 ppm STPP. St. Angelo et al. (1988) also evaluated the effects of antioxidants on WOF development by measuring the hexanal content. They found that STPP at levels of 200 ppm and 5000 ppm inhibited hexanal formation by 32% and 90%, respectively.

Of all the antioxidant treatments tested, the STPP/TBHQ combination showed the greatest effect by decreasing hexanal production by 95%. However, no significant difference between STPP/TBHQ and STPP/OR (0.05% and 0.1%) in preventing hexanal formation was found. Combinations of STPP/OR (0.05% and 0.1%) decreased the hexanal contents by

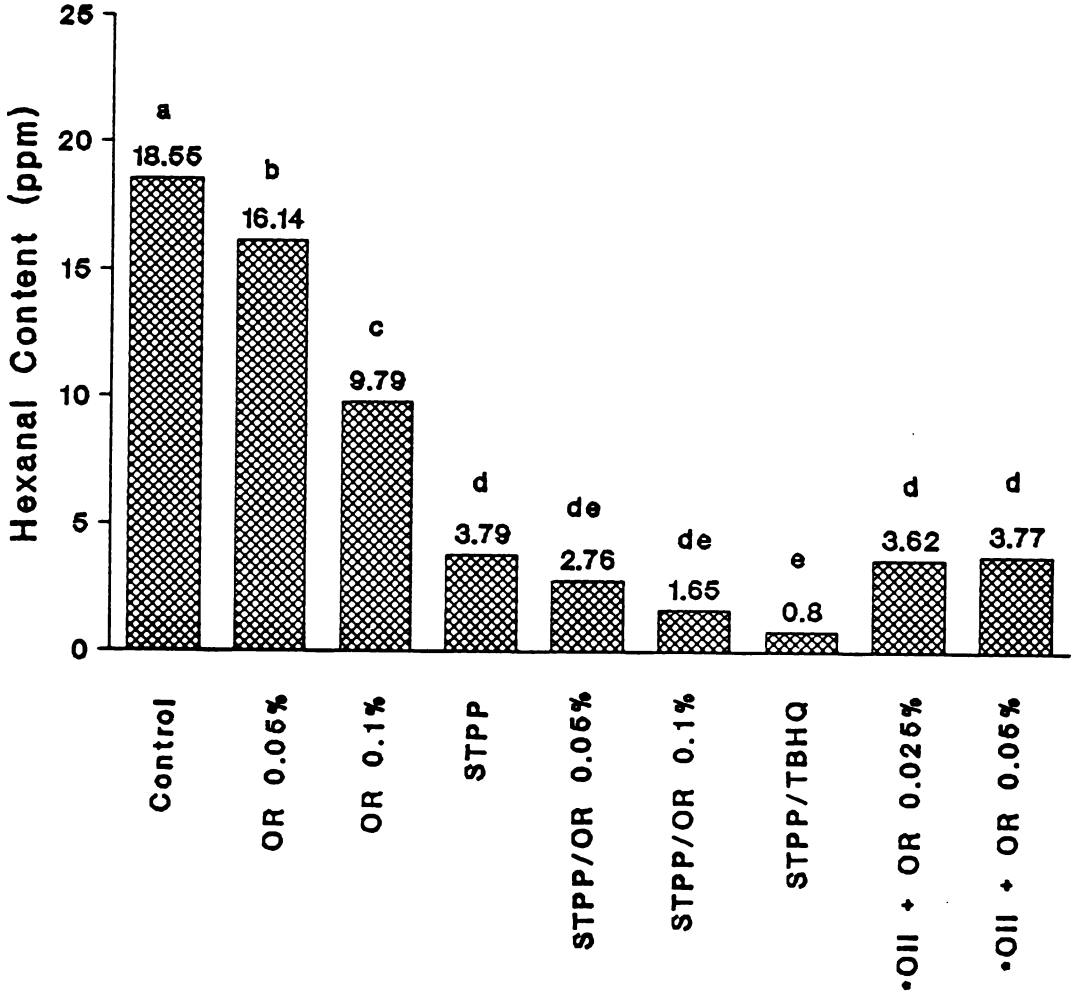


Figure 8. Hexanal content of chicken nuggets after 6 months of frozen storage (Means of three replicated experiments with different letters are significantly different at $p < 0.05$; * Samples fried in oil containing OR contained salt and STPP).

85% and 91%, respectively. These results agree with those from the TBA test and sensory evaluation and again confirmed the effectiveness of the STPP/OR combinations.

Correlation of TBARS Values, Sensory Scores and Hexanal Content

One of the objectives of this study was to correlate TBARS values and sensory scores of chicken nuggets during storage. The TBA test has been widely used to assess the extent of oxidative deterioration in muscle foods (Gray, 1978). This method is based on the quantitation of the complex formed between malonaldehyde, an end product of lipid oxidation, and thiobarbituric acid (Sinnhuber and Yu, 1958). However, the interaction of malonaldehyde with amino acids (Buttkus, 1967; Gokalp et al., 1983) and the presence of other non-lipid TBA reactive-substances in biological samples (Igene et al., 1985; Janero and Burghardt, 1989) may limit the quantitative use of the TBA test. Thus, establishing a relationship between TBARS values and sensory scores for warmed-over flavor was attempted in this study.

Statistically significant ($P < 0.05$) but moderate correlation coefficients (pooled, within treatments and storage times) of $r = 0.45$ and $r = 0.56$ were found between TBARS values and sensory scores of chicken nuggets in the refrigerated and frozen studies, respectively. These results confirmed the relationship between TBARS values and sensory scores, although the degrees of association

indicated that changes in TBARS values accounted for only 20% and 30% of the variation (r^2) in sensory scores for the refrigerated and frozen studies, respectively. These data are in agreement with the results of Zipser et al. (1964) and Igene and Pearson (1979), who reported a good relationship between TBARS values and the development of oxidized flavor in cooked meats.

However, the correlation coefficients obtained for this study are relatively low compared to those cited in the literature (Igene and Pearson, 1979; Igene et al., 1985; Smith et al., 1987; St. Angelo et al., 1987). The variability among literature values may be due to the different methods used to calculate the r value (pooled within groups, across groups, means values only) and the number of factors involved in the experiments.

Because strong effects of treatments and storage times were involved, the correlations between TBARS values and sensory scores were calculated by pooling information within treatments and storage times (Appendix B). Many "significant" correlations of TBARS values and sensory scores which have been previously reported were calculated on the individual observations over groups differing in mean values of TBARS values and sensory scores and without any comment.

The correlation computed from samples from heterogenous populations may be almost worthless as information about the relationship in a specific subpopulation, because the

measure is highly dependent on the range of the variables sampled, i.e., correlation over groups differing in mean values of x and y is necessarily higher than the correlation within a group (Gill, 1978). For example, the intra-treatment, intra-storage time correlations for the refrigerated and frozen studies, calculated by simply pairing the individual observations of TBARS values and sensory scores over treatments and storage times were 0.85 and 0.87, respectively. However the statistical significance and strong correlations do not necessarily mean the relationships are sufficiently close to be of any practical use.

Compared with other researchers who studied the relationship between TBARS values and sensory scores and obtained high correlation coefficients, more experimental factors and more treatments within a factor were involved in the present study and may affect the overall correlation. To investigate the effect of individual factors on the correlation of TBARS values and sensory scores, correlation within each factor (treatments and storage times) were calculated separately.

The correlation coefficients obtained by pooling information within storage times for each treatment are presented in Table 19. The within-time correlations of the control and the OR (0.05%)-treated samples were very low in refrigerated and frozen storage. As previously discussed, panelists were unable to detect the variation over storage

Table 19. Within-time correlation coefficients between TBARS values and sensory scores of chicken nuggets for each treatment.

Treatment	Refrigerated Storage	Frozen Storage
Salt	0.00	0.07
Salt/OR (0.05%)	0.04	0.16
Salt/OR (0.1%)	0.71*	0.38
Salt/STPP	0.41	0.40
Salt/STPP/OR (0.05%)	0.60*	0.83*
Salt/STPP/OR (0.1%)	0.75*	0.95*
Salt/STPP/TBHQ	-0.51	0.00
Oil + OR (0.025%)[1]	0.35	0.77*
Oil + OR (0.05%)	0.78*	0.72*

[1] Samples fried in oil containing OR contained salt and STPP.
* Correlation coefficient is significant at $p < 0.05$.

Table 20. Within-treatment correlation coefficients between TBARS values and sensory scores of chicken nuggets at each sampling period during storage.

Sampling period	Refrigerated Storage	Frozen Storage
0 (Day/Month)	0.20	0.20
2 (Day/Month)	0.20	0.70*
4 (Day/Month)	0.47*	0.74*
6 (Day/Month)	0.65*	0.55*

* Correlation coefficient is significant at $p < 0.05$.

times for these two treatments which developed very strong WOF even during the early stages of storage. Poste et al. (1986) reported that the scores of WOF for cooked meat increased up to 3 days of storage, then either increased or decreased, depending on the panelist's threshold for rancidity. Thus, the different sensitivities of the TBA test and sensory evaluation of the samples which oxidized most rapidly and to the greatest extent may be responsible for the low correlations obtained.

An interesting difference between refrigerated and frozen storage is the correlations for the STPP/TBHQ-treated samples. The STPP/TBHQ treatment resulted in almost total inhibition of lipid oxidation in chicken nuggets during refrigerated and frozen storage, as indicated by the results of the TBA test. The negative and zero correlations obtained for refrigerated and frozen storage may be attributed to the inability of the sensory panel to detect very low levels of WOF.

The limited threshold of the panelists also resulted in insignificant within-treatment correlations at the beginning of storage (Table 20). The within-treatment correlations between TBARS values and sensory scores were not significant at days 0 and 2 because of little variance in the degree of oxidation among samples, but became significant and increased at the days 4 and 6 of storage. A similar trend was observed for within-treatment correlations during frozen storage. The exception was a decreased correlation after 6

months of frozen storage which may be attributed to the decline in TBARS values of some treatments after 4 months of frozen storage.

As previously discussed, the TBA test may be less valid in following WOF development in meats because of the instability of malonaldehyde in long-term frozen storage. Thus, correlations of hexanal contents, TBARS values and sensory scores in the chicken nuggets after 6 months of frozen storage were made to assess the efficacy of these measurements in monitoring lipid oxidation. A higher correlation coefficient (pooled, within treatments) between hexanal contents and sensory scores (0.68) was obtained than that between hexanal content and TBARS values(0.14) and between sensory scores and TBARS values(0.55). This indicated a better consistency between hexanal content and sensory scores. The use of instrumental and sensory analyses may be more accurate for evaluating the flavor quality, i.e.,warmed-over flavor, of the chicken nuggets after 6 months of frozen storge.

Although sensory scores correlated significantly ($p < 0.05$) with hexanal contents and with TBARS values, the changes in sensory scores accounted for only 46% and 30% of the variation (r^2) in hexanal contents and TBARS values, respectively. These results were different from the findings of Shahidi et al. (1987), who reported that the hexanal content, sensory scores, and TBARS values of treated meats were linearly interrelated. They reported the correlation

coefficient for the linear relationship between the hexanal content and the TBARS values to be 0.995. Since an untrained panel was used in their study, linear correlations of sensory scores with other measurements seemed impossible to reach. Although the procedures for the statistical analyses were not provided by these investigators, the results may have been computed on the means of replicates for each treatment, leading to the higher correlation coefficients. In addition, the measurements were not performed at the same time, i.e., hexanal content and sensory evaluation were determined after 2 days of storage while the TBA test was assayed after 35 days of refrigerated storage. These differences made their results less valuable as a source of comparison.

The data from this study indicated that sensory evaluation is a useful means to assess the development of WOF in chicken nuggets during storage. However, there was a threshold for detection of oxidized flavors which limited the ability of the panel to discriminate between varying degrees of oxidation among the samples. The judges involved in this experiment did not receive extensive training, though most had experience in evaluating warmed-over flavor in meat. Proper selection and training of the sensory panel may improve the accuracy in differentiating the intensity of WOF in chicken nuggets.

Love (1988) reported that the descriptors and scoring scales used in sensory evaluation also affect the ability of

panelists to properly respond to the degrees of WOF perceived. When panelists were asked to use a scale to evaluate the intensity of a single attribute, descriptors such as "warmed-over" (Igene et al., 1985), "rancid" (Younathan and Watts, 1958) and "oxidized" (Greene, 1969; Greene et al., 1971) often have been employed. Since lipid oxidation has been considered to be the major contributor to WOF, the terms rancidity and WOF seem to be interchangeable (Younathan et al., 1980; Love, 1988). It has not been established whether a sensory panel can differentiate WOF from other types of autoxidative changes (Poste et al., 1986).

However, the off-flavors of the chicken nuggets developed during storage were due to not only the WOF developed in the meat but also to the rancid odor attributed to the deterioration of the vegetable oil absorbed by the breading of the nuggets. Thus, the panel evaluating oxidized flavor in chicken nuggets may be only partially characterizing WOF. In addition, the incorporation of salt and oleoresin rosemary into chicken nuggets may mask the WOF or oxidized flavor, or influence the panelists' perception of WOF in some cases, though it has not been established by any statistical information.

It was observed that a few panelists consistently employed a rather restricted range (i.e., a range of 3 or less units) of numbers for attributes that the majority of

panelists judged to cover over a range of 6 scale units in the samples. Increasing the range of the scale units by these panelists may improve the scoring patterns that differed from those of the majority of the panel. However, the different range of scale employed by panelists in this study may be due to the different threshold of panelists in their perception of oxidized flavor.

Love (1988) suggested the use of defined standards of all meat flavor characteristics and the replacing of terms such as warmed-over flavor, stale or off-flavor with more specific terms such as the flavor of cooked lean beef, cardboard, and painty to obtain additional information for effective panel training. To train sensory panels for more accurate evaluation of the intensity of WOF in samples, proper designs of scoring scales and descriptors for specific flavors are necessary in future sensory studies.

Effects of Oleoresin Rosemary on the Resistance of Oil to Oxidation

Although the results from the storage studies of chicken nuggets indicated no apparent beneficial effects of adding OR to the oil on the oxidative stability of the nuggets, the frying oil containing OR had higher induction times after 24 hours of continuous heating (Figure 9). The induction times of the oil samples containing OR (0.025% and

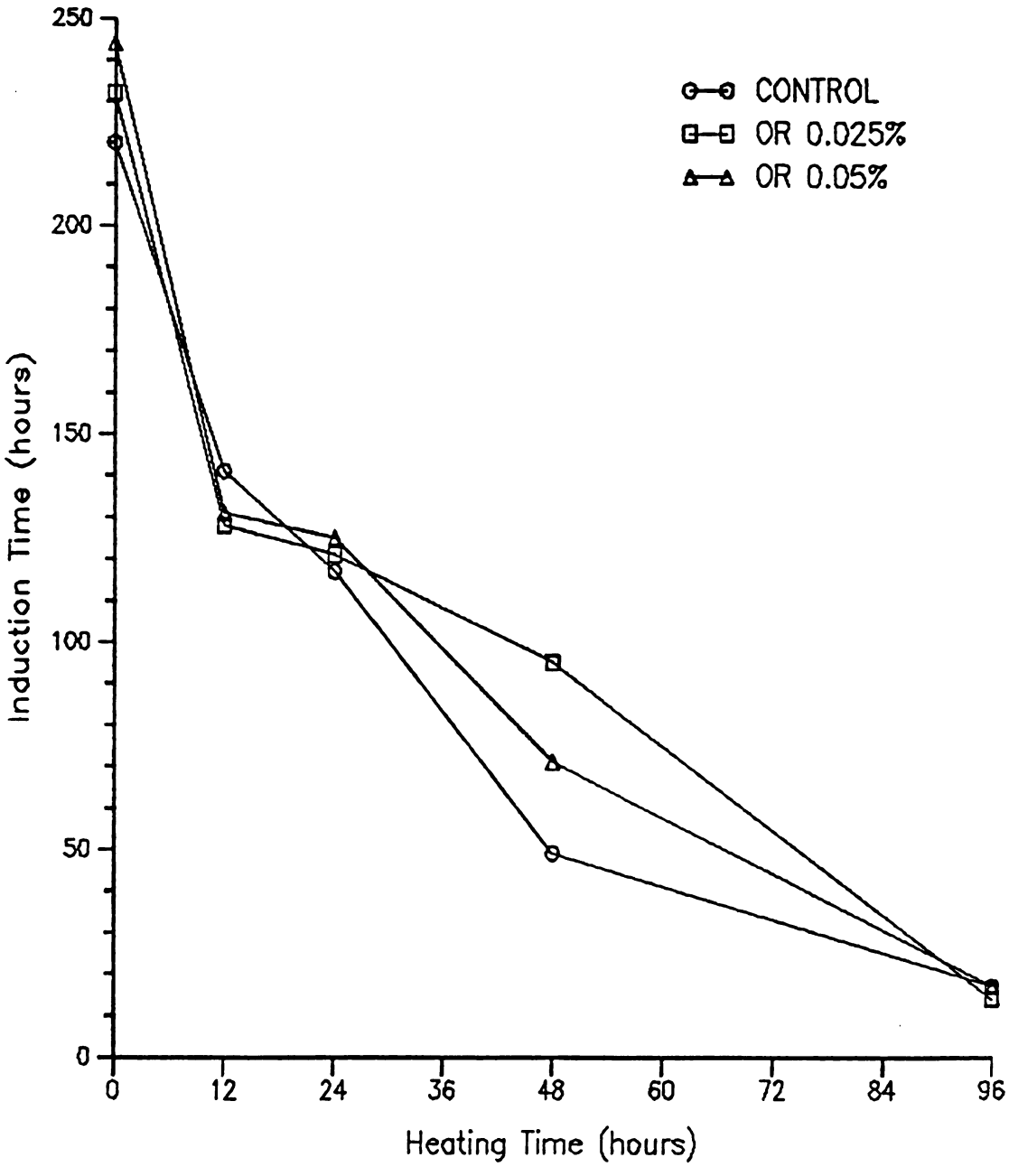


Figure 9. Antioxidant activity of oleoresin rosemary in soybean oil during continuous heating at 176°C for 96 hours.

0.05%) were consistently higher than those of the control, then declined after 48 hours of continuous heating.

Löliger (1983) studied the antioxidant activity of rosemary extracts in chicken fat during an accelerated oxidation test at 100°C using Rancimat methodology. The induction time of the rosemary-treated sample was five times longer than that of the control. Several active compounds isolated from rosemary extracts, such as carnosol, rosmanol, rosmaridiphenol and rosmariquinone, have been tested in animal fats and found comparable to synthetic phenolic antioxidants (Wu et al., 1982; Inatani et al., 1982, 1983; Houlihan et al., 1984, 1985). However, no studies have involved the evaluation of OR in frying media.

Gray et al. (1988) evaluated the thermal stability of OR in a refined soybean oil heated in an open atmosphere at 204°C for 4 hours. They observed a consistent but moderate loss of antioxidant activity with time. The loss of antioxidant activity of OR may explain the decline in induction time in the oils containing OR during long term continuous heating at high temperature.

The changes in the fatty acid composition of soybean oil after continuously heating for 96 hours are shown in Table 21. A decrease in $C_{18:2}$ and $C_{18:3}$ was found in the oil after heating, while an increase in $C_{16:0}$, $C_{18:0}$ and $C_{18:1}$ was observed. $C_{14:0}$, $C_{20;0}$, and $C_{20;1}$ which existed in the original oil in low amounts disappeared from the frying oil after 96 hours of heating. However, the control

oil sample contained higher $C_{18:2}$ and $C_{18:3}$ than the oil containing OR after 96 hours of heating at 176°C. The protective effect of OR on the polyunsaturated fatty acids in the chicken nuggets which was exhibited during storage was not apparent in the frying oil during continous heating. This may be due to the different mechanisms involved in the degradation of the oil during continuous heating.

Table 21. Changes in the fatty acid composition of soybean oil during continuous heating at 176°C for 96 hours[1].

Fatty Acid	Control		OR 0.025%		OR 0.05%	
	0 hr	96 hr	0 hr	96 hr	0 hr	96 hr
14:0	0.1	--	0.1	--	0.1	--
16:0	10.0	11.4	10.2	13.0	10.4	14.0
16:1	0.1	--	0.1	--	0.1	--
18:0	4.4	5.4	4.4	6.0	4.4	6.2
18:1	25.0	29.4	24.8	31.1	24.7	32.2
18:2	52.6	49.1	52.6	46.2	52.6	44.8
18:3	7.0	4.8	7.0	3.6	6.8	3.2
20:0	0.4	--	0.4	--	0.3	--
20:1	0.2	--	0.2	--	0.2	--

[1] As percent of fatty acids.

Loss of unsaturation during continuous heating of oil occurs only via thermal oxidation and polymerization reactions at elevated temperatures, as the autoxidation reaction is unable to occur under high constant temperature conditions (Perkins and Van Akkeren, 1965; Perkins, 1967). Thus OR, a free radical terminator, was less effective in retarding thermal degradation of the frying oil than it was

in protecting chicken nuggets during storage where a free radical chain reaction was involved in the deteriorative process.

SUMMARY AND CONCLUSIONS

The antioxidant effects of oleoresin rosemary (OR), sodium tripolyphosphate (STPP) and TBHQ during the refrigerated and frozen storage of restructured chicken nuggets were evaluated. The effect of OR on the resistance of oil to oxidation was also investigated.

TBARS values and sensory scores for chicken nuggets demonstrated that STPP/TBHQ exhibited the best protective effect during refrigerated storage. Both OR (0.1%) and STPP, when used alone, reduced the extent of salt-catalyzed lipid oxidation, although STPP was more effective than the OR over the entire storage period. However, the combination of OR (0.05% and 0.1%) and STPP was very effective and demonstrated an additive antioxidant effect. No significant difference was found between the STPP/OR (both levels) and STPP/TBHQ treatments, indicating that OR/STPP was equally as effective as STPP/TBHQ in protecting refrigerated chicken nuggets.

Similar trends were observed for the chicken nuggets during frozen storage. Results indicated that STPP/OR was comparable to STPP/TBHQ in supressing lipid oxidation during frozen storage. The additive effect between OR and STPP was again observed, although it was not as pronounced as that obtained for the refrigerated study. Fatty acid and hexanal

data were in agreement with these results. Both STPP/OR and STPP/TBHQ effectively protected the oxidative degradation of the polyunsaturated fatty acids and inhibited the formation of hexanal in the chicken nuggets during frozen storage.

Significant ($p < 0.05$) but moderate correlation coefficients between TBARS values and sensory scores were found in the refrigerated and frozen studies. Correlation of hexanal contents and sensory scores was higher than that of hexanal contents and TBARS values, which indicated a better consistency between hexanal contents and sensory scores.

Studies involving the cooking of nuggets in soybean oil containing OR and subsequent storage of the cooked nuggets under refrigerated and frozen conditions indicated no apparent beneficial effects of adding OR to the oil on the oxidative stability of the chicken nuggets.

In conclusion, results from this study demonstrated that STPP/OR was as effective as STPP/TBHQ in protecting restructured chicken nuggets during storage. There was a good relationship between TBARS values and sensory scores, although the hexanal content was a better index of oxidative rancidity than TBARS values in frozen samples during long-term storage.

Appendix A

Tast panel form for sensory evaluation of WOF in chicken nuggets.

NAME_____ DATE_____

(1) You are receiving a series of coded samples and a reference sample, marked **R.**

(2) Smell and taste each sample; consider the reference sample to have no warmed over flavor and evaluate the degree of **warmed over flavor, i.e., oxidative off-flavor**

(3) Then mark the intensity of the flavors that you detect from the samples.

(4) Also, indicate with **yes** or **no** the presence of other non-meat flavors.

THE INTENSITY OF WARMED OVER FLAVOR

Absent : 0
Very Slight : 1
Slight : 2
Moderate : 3
Strong : 4
Very Strong : 5

CODE	INTENSITY OF WOF	NON-MEAT FLAVOR	COMMENTS
Set #			
------	-------	-------	------------------
------	-------	-------	------------------
------	-------	-------	------------------

Appendix B

Calculation of correlation coefficients (pooled, within treatments and storage times) between TBARS values and sensory scores of chicken nuggets in frozen study.

x = sensory scores
y = TBARS values

i = 1,2,····9 (treatments)
j = 1,2,3,4 (months)
k = 1,2,3 (replications)

Variances (s_x^2) of sensory scores (x) within three replications

$$s^2_x = (\sum_{k=1}^{3} x_k^2 - (\sum_{k=1}^{3} x_k)^2/3)/(3-1)$$

Treatments (i)	Months (j) 0	2	4	6	Σs^2_x
1	0.26	0.23	0.02	0.00	0.51
2	0.93	0.07	0.12	0.16	1.28
3	0.25	0.06	0.07	0.12	0.50
4	0.59	0.08	0.37	0.19	1.23
5	0.08	0.93	0.56	0.84	2.41
6	0.10	0.97	0.76	1.36	3.19
7	0.07	0.02	0.14	0.17	0.40
8	0.20	0.11	0.30	0.57	1.18
9	0.25	0.65	0.63	0.32	1.85
Σs^2_x	2.73	3.12	2.97	3.73	

Variances (s_y^2) of TBARS values (y) within three replications

$$s^2_y = (\sum_{k=1}^{3} y_k^2 - (\sum_{k=1}^{3} y_k)^2/3)/(3-1)$$

Treatments (i)	Months (j) 0	2	4	6	Σs^2_y
1	0.19	0.30	0.43	0.07	0.99
2	0.19	0.02	0.71	0.43	1.35
3	0.31	0.10	0.02	0.07	0.50
4	0.12	0.07	0.13	0.44	0.76
5	0.08	0.11	0.58	0.27	1.04
6	0.06	0.23	0.16	0.41	0.86
7	0.06	0.00	0.01	0.01	0.08
8	0.10	0.37	0.21	0.26	0.94
9	0.08	0.18	0.70	0.20	1.16
Σs^2_y	1.19	1.38	2.95	2.16	

Covariances (s_{xy}) of sensory scores (x) and TBARS values (y) within three replications

$$s_{xy} = (\sum_{k=1}^{3} x_k y_k) - (\sum_{k=1}^{3} x_k)(\sum_{k=1}^{3} y_k)/3)/(3-1)$$

Treatments (i)	Months (j) 0	2	4	6	Σs_{xy}
1	0.11	-0.13	0.08	-0.01	0.05
2	0.34	-0.01	0.07	-0.19	0.21
3	0.05	0.02	0.03	0.09	0.19
4	0.03	0.06	0.21	0.09	0.39
5	-0.03	0.32	0.55	0.47	1.31
6	0.03	0.45	0.34	0.75	1.57
7	-0.05	0.00	0.03	0.02	0.00
8	-0.06	0.43	0.25	0.19	0.81
9	-0.06	0.32	0.64	0.15	1.05
Σs_{xy}	0.36	1.46	2.20	1.56	

$$\sum_{i}^{9}\sum_{j}^{4} s^2_{x_{ij}} = 12.55$$

$$\sum_{i}^{9}\sum_{j}^{4} s^2_{y_{ij}} = 8.09$$

$$\sum_{i}^{9}\sum_{j}^{4} s_{x_{ij}y_{ij}} = 5.65$$

$$r_{xy} = s_{xy}/(s^2_x \cdot s^2_y)^{0.5}$$

$$= \sum_{i}^{9}\sum_{j}^{4} s_{x_{ij}y_{ij}} / \left(\sum_{i}^{9}\sum_{j}^{4} s^2_{x_{ij}} \cdot \sum_{i}^{9}\sum_{j}^{4} s^2_{y_{ij}}\right)^{0.5}$$

$$= 5.65/(12.55 \cdot 8.09)^{0.5}$$

BIBLIOGRAPHY

Acosta, S.O., Marion, W.W. and Forsythe, R.H. 1966. Total lipids and phospholipids in turkey tissues. Poultry Sci. 45: 169.

Addis, P.B. and Pearson, A.M. 1986. Sensory evaluation techniques to assess oxidative rancidity. In " Proceedings of 38th Annual Reciprocal Meat Conference," p. 99. Natl. Live Stock and Meat Board, Chicago, Ill.

Allen, C.E. and Foegeding, E.A. 1981. Some lipid characteristics and interactions in muscle foods. Food Technol. 35: 253.

Ang, C.Y.W. 1988. Comparison of broiler tissues for oxidative changes after cooking and refrigerated storage. J. Food Sci. 53: 1072.

Ang, C.Y.W. and Hamm, 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.

A.O.A.C. 1975. Official Methods of Analysis, 12th ed. Association of Official Analytical Chemists, Washington, DC.

Apte, S. and Morrissey, P.A. 1987. Effect of hemoglobin and ferritin on lipid oxidation in raw and cooked muscle systems. Food Chem. 25: 127.

Asghar, A., Gray, J.I. Buckley, D.J., Pearson, A.M. and Booren, A.M. 1988. Perspectives on warmed-over flavor. Food Technol. 42(6): 102.

Badings, H.T. 1970. Cold-storage defects in butter and their relation to the oxidation of unsaturated fatty acids. Neth. Milk Dairy J. 24: 147.

Bailey, M.E., Dupuy, H.P. and Legendre, M.G. 1980. Undesirable meat flavor and its control. In "The Analysis and Control of Less Desirable Flavors in Food and Beverages," G. Charalambous (Ed.), Academic Press, N.Y.

Barbut, S., Josephson, D.B. and Maurer, A.J. 1985. Antioxidant properties of rosemary oleoresin in turkey sausage. J. Food Sci. 50: 1356.

Benedict, R.C., Strange, E.D. and Swift, C.E. 1975. Effect of lipid antioxidants on the stability of meat during storage. J. Agric. Food Chem. 23: 167.

Blumer, T.N. 1963. Relationship of marbling to the palatability of beef. J. Anim. Sci. 22: 771.

Booren, A.M., Mandigo, R.W., Olson, D.G. and Jones, K.W. 1981. Effect of muscle type and mixing time on sectioned and formed beef steaks. J. Food Sci. 46: 1665.

Bracco, U., Loliger, J. and Viret, J.L. 1981. Production and use of natural antioxidants. J. Am. Oil. Chem. Soc. 58: 686.

Brieskorn, C.H., Fuch, A., Bredenberg, T.B., Machesney, T. D. and Wenkert, E. 1964. The structure of carnosol. J. Org. Chem. 29: 2293.

Buttkus, H. 1967. The reaction of myosin with malonaldehyde. J. Food Sci. 32: 432.

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

Chang, S.S., Ostric-Matijasevic, B., Hsieh, O.A.L. and Huang, C. 1977. Natural antioxidants from rosemary and sage. J. Food Sci. 42: 1103.

Chastain, M.F., Huffman, D.L., Hsieh, W.H. and Cordray, J.C. 1982. Antioxidants in restructured beef/pork steaks. J. Food Sci. 47: 1779.

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

Chen, C.C., Pearson, A.M., Gray, J.I. and Merkel, R.A. 1984b. Effects of salt and some antioxidants upon the TBA numbers of meat. Food Chem. 14: 167.

Chen, T.C. and Waimaleongora-ek, C. 1981. Effect of pH on TBA values of ground raw poultry meat. J. Food Sci. 46: 1946.

Chipault, J.R., Mizuno, G.R. and Lundberg, W.O. 1956, The antioxidant properties of species in food. Food Technol. 10(5): 209.

Chipault, J.R., Mizuno, G.R., Hawkins, J.M. and Lundberg, W. O. 1952. The antioxidant properties of natural species. Food Res. 17: 46.

Chipault, J.R., Mizuno, G.R., and Lundberg, W.O. 1955. Antioxidant properties of species in oil-in-water emulsion. Food Res. 20: 443.

Christie, W.W. 1978. The composition, structure and function of lipids in the tissues of ruminant animals. Prog. Lipid Res. 17: 111.

Corliss, G.A. and Dugan, L.R., Jr. 1970. Phospholipid oxidation in emulsions. Lipids 5: 486.

Crackel, R.L., Gray, J.I., Booren, A.M., Pearson, A.M. and Buckley, D.J. 1988a. Effect of antioxidants on lipid stability in restructured beef steaks. J. Food Sci. 53: 656.

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

Crawford, D.L., Sinnhuber, R.A., and Aft, H. 1961. Effect of methylation on the antioxidant and chelation capacity of quercetin and dehydroquercetin in a lard substrate. J. Food Sci. 26: 139.

Cross, C.K. and Ziegler, P. 1965. A comparison of the volatile fractions from cured and uncured meat. J. Food Sci. 30: 610.

Csallany, A.S., Guan, M.D., Manwaring, J.D. and Addis, P.B. 1984. Free malonaldehyde determination in tissues by high performance lipid chromatography, Anal, Biochem. 142: 227.

Dawson, L.E. and Gartner, R. 1983. Lipid oxidation in mechanically deboned poultry. Food Techno. 37(7): 112.

Decker, E.A. and Schanus, E.G. 1986a. Catalysis of linoleate oxidation by soluble chicken muscle proteins. J. Am. Oil. Chem. Soc. 63: 101.

Decker, E.A. and Schanus, E.G. 1986b. Catalysis of linoleate oxidation by nonheme and heme-soluble chicken muscle protein. J. Agric. Food Chem. 34: 991.

Dougherty, M.E., Jr. 1988. Tocopherols as food antioxidants. Cereal Foods World. 33: 222.

Dugan, L.R., Jr. 1987. Fats. In "The science of Meat and Meat Products," J.F. Price and B.S.Schweigert (Ed.), 3nd ed. p. 103. Food and Nutrition Press, Inc., Westport, CT.

El-Gharbawi, M.I. and Dugan, L.R., Jr. 1965. Steam volatile components from freeze-dried beef stored at room temperature. J. Food Sci. 30: 814.

Farr, A.J., and May, K.N. 1970. The effect of polyphosphates and sodium chloride on cooking yields and oxidative stability of chicken. Poultry Sci. 49: 268.

Frank, J., Geil, J.V., and Freaso, R. 1982. Automatic determination of oxidation stability of oil and fatty products. Food Technol. 36(6): 71.

Frankel, E.N., Heff W.E. and Selke, E. 1981. Analysis of autooxidized 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.

Fritsch, C.W. and Gale, J.A. 1977. Hexanal as a measure of rancidity in low fat foods. J. Am. Oil Chem. Soc. 54: 225.

Froning, G.W. 1970. Poultry meat sources and their emulsifying characteristics as related to processing variables. Poultry Sci. 49: 1625.

Froning, G.W. 1973. Effect of chilling in the presence of polyphosphates on the characteristics of mechanically deboned fowl meat. Poultry Sci. 52: 920.

Gill, J.L. 1978. "Design and Analysis of Experiments in the Animal and Medical Sciences." Iowa State Univ. Press, Ames, IO.

Gerhardt, U. and Blat, P. 1984. Dynamische messmethode zur ermittlung der fettstabilitat. Einfluss von gewurzen and zusatzstoffen. Fleichwirtschaft. 64: 484.

Gokalp, H.Y., Ockerman, H.W., Plimpton, R.F. and Harper, W. 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, G.M. and Macfarlane, M.G. 1961. Composition of phospholipids of rabbit, pigeon and trout muscles and various pig tissues. Biochem. J. 81: 480.

Gray, J.I. 1978. Measurement of lipid oxidation. J. Am. Oil Chem. Soc. 55: 39.

Gray, J.I., Crackel, R.L., Cook, R.J., Gastel, A.L., Evans, R.L. and Buckley, D.J. 1988. Observations on the antioxidant properties and thermal stability of an oleoresin rosemary. Presented at 3rd International Conference on Ingredients Europe, London, Nov 15-17.

Gray, J.I. and Pearson, A.M. 1987. Rancidity and warmed-over flaver. In "Advances in Meat Research," A.M. Pearson and T.R. Dutson (Ed.), Vol III, p. 221. AVI Publ. Co., Westport, CT.

Greene, B.E. 1969. Lipid oxidation and pigment changes in raw beef. J. Food Sci. 34: 110.

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

Greene, B.E., Hsin, I.N. and Zipser, M.W. 1971. Retardation of oxidative color changes in raw ground beef. J. Food Sci. 36: 940.

Grunden, L.P., MacNeil, J.H. and Dimick, P.S. 1972. Poultry product quality: chemical and physical characteristics of mechanically deboned poultry meat. 37: 247.

Hardorn, J. and Zürcher, K. 1974. Zur Bestimmung der Oxidations-stabilität von Oelen und Fetten. Deutsch. Lebensm.-Rudsch 70: 57.

Herz, K.O. and Chang, S.S. 1970. Meat flavor. Adv. Food Res. 18: 1.

Houlihan, C.M. and Ho, C.T. 1985. Natural antioxidants. In "Flavor Chemistry of Fats and Oils," D.B. Min and T.H. Smouse, (Ed.), P. 140. Am. Oil Chem. Soc., Champaign, Ill.

Houlihan, C.M., Ho, C.T. and Chang, S.S. 1984. Elucidation of the chemical structure of a novel antioxidant, rosari diphenol, isolated from rosemary. J. Am. Oil. Chem. Soc. 61: 1036.

Houlihan, C.M., Ho, C.T. and Chang, S.S. 1985. The structure of rosmariquinone - a new antioxidant isolated from Rosmarinus officinalis L. J. Am. Oil Chem. Soc. 62: 96.

Huang, W.H. and Greene, B.E. 1978. Effect of cooking method on TBA numbers of stored beef. J. Food Sci. 43: 1201.

Huffman, D.L., Cross, H.R., Campbell, K.J. and Cordray, J.C. 1981a. Effect of salt and tripolyphosphate on acceptability of flaked and formed hamburger patties. J. Food Sci. 46: 34.

Huffman, D.L., Ly, A.M. and Cordray, J.C. 1981b. Effect of salt concentration on quality of restructured pork chops. J. Food Sci. 46: 1563.

Inatani, R., Nakatani, N., Fuwa, H., and Seto, H. 1982. Structure of a new antioxidative phenolic diterpene isolated from rosemary (Rosmarinus officinalis L.) Agric. Bid. Chem. 46: 1661.

Inatani, R., Nakatani, N., and Fuwa, H. 1983. Antioxidative effect of the constituents of rosemary (Rosmarinus officinalis L.) and their derivatives. Agric. Boil. Chem. 47: 521.

Igene, J.O., King, J.A., Pearson, A.M. and Gray, J.I. 1979a. 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., Dugan, L.R., Jr. and Price, J.F. 1980. Role of triglycerides and phospholipids on development of rancidity in model meat systems during frozen storage. Food Chem. 5: 263.

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

Igene, J.O., Pearson, A.M., Merkel, R.A. and Coleman, T.H. 1979b. Effect of frozen storage time, cooking and holding temperature upon extractable lipids and TBA values of beef and chicken. J. Anim. Sci. 49: 701.

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.

Ingold, K.V. 1962. Metal catalysis. In "Symposium on Foods: Lipids and their Oxidation," H.W. Schultz, E.A. Day and R.O. Sinnhuber (Ed.) AVI Publ. Co., Westport, CT.

Janatawat, P. and Dawson, L.E. 1980. Composition of lipid from mechanically deboned poultry meat and their composite tissues. Poultry Sci. 59: 1043.

Janero, D.R. and Burghardt, B. 1989. Thiobarbituric acid-reactive malondialdehyde formation during superoxide-dependent, iron-catalyzed lipid oxidation: Influence of peroxidation conditions. Lipids 24(2): 125.

Janky, D.M. and Froning, G.W. 1975. Factors affecting chemical properties of heme and lipid components in mechanically deboned turkey meat. Poultry Sci. 54: 1378.

Josephson, D.B., Lindsay, R.C. and Stuiber, D.A. 1983. Identification of compounds characterizing the aroma of fresh whitefish (Coregonus clupeaformis). J. Agric. Food Chem. 31: 326.

Jurdi-Haldeman, D., MacNeil, J.H. and Yared, D.M. 1987. Antioxidant activity of onion and garlic juices in stored cooked ground lamb. J. food protect. 50: 411.

Kanner, J. and Harel, S. 1985. Initiation of membranal lipid peroxidation by activated metmyoglobin and methemoglobin. Arch. Biochem. Biophys. 237: 314.

Katz, M.A., Dugan, L.R., Jr. and Dawson, L.E. 1966. Fatty acids in neutral lipids and phospholipids from chicken tissues. J. Food Sci. 31: 717.

Khayat, A. and Schwall, D. 1983. Lipid oxidation in seafood. Food Technol. 37(7): 130.

King, A.J. and Earl, L.A. 1988. Effect of selected sodium and potassium salts on TBA values of dark meat turkey patties. J. Food Sci. 53: 723.

King, A.J., Patel, S.B., and Earl, L.A. 1986. Effect of water, egg white, sodium chloride and polyphosphates on several quality attributes of unfrozen and frozen, dark, ground turkey muscle. Poultry Sci. 65: 1103.

Korczak, J., Flaczyk, E. and Pazola, Z. 1988. Effects of species on stability of minced meat products kept in cold storage. Fleischwirtschaft 68: 64.

Labuza, T.P. 1971, Kinetics of lipid oxidation in foods. CRC Crit. Rev. Food Technol. 2: 355.

Laubli, M.W. and Bruttel, P.A. 1986. Determination of the oxidative stability of fats and oils: Comparison between the active oxygen method (AOCS cd 12-57) and the rancimat method. J. Am. Oil Chem. Soc. 63: 792.

Legendre, M.G., Fisher, G.S., Schuller, W.H., Dupuy, H.P. and Rayner, E. T. 1979. Novel technique for the analysis of volatiles in aqueous and nonaqueous systems. J. Am. Oil Chem. Soc. 56: 552.

Lillard, D.A. and Ayres, J.C. 1969. Flavor compounds in country used hams. Food Technol. 23: 251.

Lin, C.F. 1988. Lipid peroxidation in poultry meat and its subcellular membranes as influenced by dietary oil and antioxidant supplementation. Ph.D. dissertation, Michigan State Univ., East Lansing.

Löliger, J. 1983. Natural antioxidants. In "Rancidity in Foods". J.C. Allen and R.I. Hamilton (Ed.), p. 89. Applied Science Publishers, London.

Love, J.D. 1983. The role of heme iron in the oxidation of lipids in red meats. Food Technol. 37(7): 116.

Love, J. 1988. Sensory analysis of warmed-over flavor in meat. Food Technol. 42(6): 140.

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 non-heme iron as prooxidants in cooked meat. J. Agric. Food Chem. 22: 1032.

Love, J.D. and Pearson, A.M. 1976. Metmyoglobin and non-heme iron as prooxidants in egg yolk phospholipids dispersions and cooked meat. J. Agric. Food Chem. 24: 494.

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. Am. Oil Chem. Soc. 37: 486.

MacDonald, B., Gray, J.I., Kakuda, Y. and Lee, M.L. 1980. The role of nitrite in cured meat flavor. II. Chemical analysis. J. Food Sci. 45: 889.

MacNeil, J.H., and Dimick, P.S., Mast, M.G. 1973. List of chemical compounds and a rosemary spices extract in quality maintenance of deboned meat. J. Food Sci. 38: 1080.

Marion, W.W., and Forsythe, R.H. 1962. Protection afforded lipids of turkey meat by butylated hydroxyanisole, egg white solids, and gelatin and tripolyphosphate. Poultry Sci. 41: 1663.

Marion, T.E. and Woodroof, J.G. 1966. Composition and stability of broiler carcasses as affected by dietry protein and fat. Poultry Sci. 45: 241.

Marmer, W.N. and Maxwell, R.J. 1981. Dry column method for the quantitative extraction and simultaneous class separation of lipids from muscle tissue. Lipids 16: 365.

Maxwell, R.J. and Marmer, W.N. 1983. Systematic protocol for the accumulation of fatty acid data from multiple tissue samples: Tissue handling, lipid extraction and class separation, and capillary gas chromatographic anaiysis. Lipids 18: 453.

Maxwell, R.J. and Marmer, W.N., Zubllaga, M.P. and Dalickas, G.A. 1980. Determination of total fat in meat and meat products by a rapid, dry column method. J. Assoc. Off. Anal. Chem. 63: 600.

McNeill, J., Kakuda, Y. and Findlay, C. 1988. Influence of carcass parts and food additives on the oxidative stability of frozen mechanically separated and hand-deboned chicken meat. Poultry Sci. 67: 270.

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

Melton, S.L., Davidson, P.M., and Mount, J.R. 1987. Sensory analysis of undesirable flavors in meat. In "Warmed-over Flavor of Meat", A. J. St. Angelo and M. E. Bailey (Ed.), p. 141 Academic Press, Inc., Orlando, FL.

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

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 Prot. 49: 222.

Moody, W.G. and Cassens, R.G. 1968. A quantitative and morphological study of bovine longissimus fat cells. J. Food Sci. 33: 47.

Morrison, W.R. and Smith, L.M. 1964. Preparation of fatty acid methyl esters and dimethyl acetals from lipids with boron fluoride-methanol. J. Lipid Res. 5: 600.

Nakatani, N., and Inatani, R. 1984. Two antioxidative diterpenes from rosemary (Rosmarirus officinalis L.) Agric. Biol. Chem. 47: 353.

Nawar, W.W. 1985. Lipids. In "Food Chemistry," O.R. Fennema (Ed.) p. 139, Marcel Dekker, Inc., N.Y.

Neer, K.L. and Mandigo, R.M. 1977. Effect of salt, sodium tripolyphosphate and frozen storage temperature on properties of a flaked, cured pork product. J. Food Sci. 42: 738.

Olson, D.G. and Rust, R.E. 1973. Oxidative rancidity in dry cured hams: Effect of low pro-oxidant and antioxidant salt formulations. J. Food Sci. 38: 251.

Pearson, A.M. 1966. Desirability of beef - Its characteristics and their measurement. J. Anim. Sci. 25: 843.

Pearson, A.M. and Gray, J.I. 1983. Mechanism responsible for warmed-over flavor in cooked meat. In, "The Maillard Reaction in Foods and Nutrition," G.R. Waller and M.S. Feather (Ed.) ACS Symposium Series 215, American Chemical Society, Washington DC.

Pearson, A.M., Love, J.D. and Shorland, F.B. 1977. Warmed-over flavor in meat, poultry and fish. Adv. Food Res. 23: 1.

Pearson, A.M. and Tauber, F.W. 1984. "Processed Meats," 2nd ed. AVI Publ. Co., Westport, CT.

Peng, C.Y. and Dugan, L.R., Jr. 1965. Composition and structure of phospholipids in chicken muscle tissues. J. Am. Oil Chem. Soc. 42: 533.

Perkin, E.G. 1967. Formation of non-volatile decomposition products in heated fats and oils. Food Technol. 21: 611.

Perkins, E.G. and Van Akkeren, L.A. 1965. Heated fats. IV. Chemical changes in fat subjected to deep fat frying processes: Cottonseed oil. J. Am. Oil Chem. Soc. 42: 782.

Pikul, J., Leszczynski, D.E., Bechtel, P.J. and Kummerow, F. A. 1984a. Effects of frozen storage and cooking on lipid oxidation in chicken meat. J. Food Sci. 49: 838.

Pikul, J., Leszczynski, D.E. and Kummerow, F.A. 1983. The elimination of sample autoxidation by butylated hydroxytoluene additions before thiobarbituric acid

assay for malonaldehyde in fat from chicken meat. J. Agric. Food Chem. 31: 1338.

Pikul, J. Leszczynski, D.E. and Kummerow, F.A. 1984b. Relative role of phospholipids, triacylglycerols and cholesterol ester on malonaldehyde formation in fat extracted from chicken meat. J. Food Sci. 49: 704.

Pikul, J. Leszczynski, D.E., and Kummerow, F.A. 1985. Influence of fat content and composition on malonaldehyde concentration in chicken meat and skin. Poultry Sci. 64: 311.

Pikul, J., Leszczynski, D.E., Niewiarowicz, A., and Kummerow, F.A. 1984c. Lipid oxidation in chicken breast and leg meat after sequential treatments of frozen storage, cooking, refrigerated storage and reheating. J. Food Technol. 19: 575.

Pokorny, J. 1971. Stabilization of fats by phenolic antioxidants. Can. Inst. Food Technol. J. 4: 68.

Pokorny, J. and Janick, G. 1971. The effect of heam derivatives on the oxidation of lipid mixed with protein. Nahrung 15: 389.

Poste, L.M., Willemot, C., Butler, G. and Patterson, C. 1986. Sensory aroma scores and TBA values as indicates of warmed-over flavor in pork. J. Food Sci. 51: 886.

Pratt, D.E. and Watts, B.M. 1964. Antioxidant activity of vegetable extracts. I. Flavone aglycones. J. Food Sci. 29: 27.

Resurreccion, A.V.A. and Reynold, A.E. Jr. 1989. Evaluation of natural antioxidants in frankfurter containing chicken and pork. Paper No. 278. Presented at 50th Annual Meeting of Inst. of Food Technologists, Chicago, Ill, June 25-29.

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

Rhee, K.S. 1987. Natural antioxidants for meat products. In "Warmed-over flavor of meats," A.J. St. Angelo and M.E. Bailey (ED.), p. 267. Academic Press. Orlando, Fl.

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

Richardson, T. and Dahl, M.K. 1982. Lipid oxidation. In "Development in Dairy Chemistry," F. Fox. (Ed.), p. 309. Applied Sci. Publ., England.

Roozen, J.P. 1987 Effect of type I, II and III antioxidants on phospholipid oxidation in a meat model for warmed-over flavor. Food Chem. 24: 167.

Rossell, J.B. 1983. Measurement of rancidity. In "Rancidity in Foods," J.C. Allen and R.J. Hamilton (Ed.) p. 21. Applied Sci. Publish Ltd. London.

Ruenger, E.L., Reineccius, G.A. and Thompson, D.R. 1978. Flavor compounds related to warmed-over flavor of turkey. J. Food Sci. 43: 1198.

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

Sato, K., Hegarty, G.R. and Herring, H.K. 1973. The inhibition of warmed-over flavor in cooked meats. J. Food Sci. 38: 398.

Satterlee, L.D., Froning, G.W. and Janky, D.M. 1971. Influence of skin content on composition of mechanically deboned poultry meat. J. Food Sci. 36: 979.

Schmedes, A. and Holmer, G. 1989. A new thiobarbituric acid (TBA) method for determining free malondialdehyde (MDA) and hydroperoxides selsctively as a measure of lipid oxidation. J. Am. oil Chem. Soc. 66: 813.

Schricker, B.R., and Miller, D.D. 1983. Effects of cooking and chemical treatment on heme and nonheme iron in meat. J. Food Sci. 48: 1340.

Schricker, B.R., Miller, D.D., and Stouffer, J.R. 1982. Measurement and content of nonheme and total iron in muscle. J. Food Sci. 47: 740.

Schwartz, W.C. and Mandigo, R.W. 1976. Effect of salt, sodium tripolyphosphate and storage on restructured pork. J Food Sci. 41: 1266.

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: 104.

Sherwin, E.R. 1978. Oxidation and antioxidants in fat and oil processing. J. Am. Oil Chem. Soc. 55: 809.

Sherwin, E.R. 1985. Synthetic antioxidants for fats and oils. In "Flavor chemical of fats and oils," D,B. Min and T.H. Smouse (Ed.), p. 155, Am. Oil Chem. Soc., Champaign, Ill.

Sheu, C.-C. 1988. Effects of rosemary olerresin and other antioxidants on the sensory and chemical properties of precooked pork patties. M.S. thesis, Iowa State Univ., Ames.

Shults, G.W., Russell, D.R. and Wierbicki, E. 1972. Effect of condensed phosphates on pH, swelling and water-holding capacity, of beef. J. Food Sci. 37: 860.

Shults, G.W. and Wierbicki, E. 1973. Effect of sodium chloride and condensed phosphates on the water-holding capacity, pH and swelling of chicken muscle. J. Food Sci. 38: 991.

Simic, M.G. and Taylor, K.A. 1987. Free radical mechanisms of oxidation reactions. In "Warmed-over flavor in meats", A.J. St. Angelo. and M. E. Bailey (Ed.), p. 63. Academic Press, Inc. Orlando, Fl.

Sinnhuber, R.O., Yu, T.C. and Yu, T.C. 1958. Characterization of the red pigment formed in the 2-thiobarbituric acid determination of oxidative rancidity. Food Res. 23: 626.

Smith, D.M. 1987. Functional and biochemical changes in deboned turkey due to frozen storage and lipid oxidation. J. Food Sci. 52: 22.

Smith, D.M., Salih, A.M. and Morgan, R.G. 1987. Heat treatment effects on warmed-over flavor in chicken breast meat. J. Food Sci. 52: 842.

St. Angelo, A.J., Vercellotti, J.R., Dupuy, H.P. and Spanier, A.M. 1988. Assessment of beef flavor quality: A multidisciplinary approach. Food Technol. 42(7): 133.

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

Tappel, A.L. 1962. Heme compounds and lipoxidase as biocatalysts. In "Symposium on Foods: Lipids and their Oxidation," H.W. Schultz, E.A. Day and R.O. Sinnhuber (Ed.) AVI Publ. Co., Westport, CT.

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.

Tarladgis, B.G. and Watts, B.M. 1960. Malonaldehyde production during controlled oxidation of pure, unsaturated fatty acids. J. Am. Oil Chem. Soc. 37: 403.

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

Theno, D.M., Siegel, D.G. and Schmidt, G.R. 1978. Meat massaging: effects of salt and phosphate on the microstructural composition of the muscle exudate. J.Food Sci. 43: 483.

Thomson, J.E. 1964. Effect of polyphosphates of oxidative deterioration of commercially cooked fryer chicken. Food Technol. 18: 1805.

Tichivangana, J.Z. and Morrissey, P.A. 1984. Factors influencing lipid oxidation in heated fish muscle systems. Ir. J. Food Sci. Technol. 8: 47.

Tichivangana, J.Z. and Morrissey, P.A. 1985. Metmyoglobin and inorganic metals as prooxidants in raw and cooked muscle systems. Meat Sci. 15: 107.

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

Trout, G.R. and Schmidt, G.R. 1984. Effect of phosphate type and concentration, salt level and method of preparation on binding in restructured beef rolls. J. Food Sci. 49: 687.

Tsai, L.S. and Smith, L.M. 1971. Role of the bases and phosphoryl bases of phospholipids in the autoxidation of methyl linoleate emulsions. Lipids 6: 196.

Uebersax, K.L., Dawson, L.E. and Uebersax, M.A. 1978. Storage stability (TBA and color) of MDCM and MDTM processed with CO_2 cooling. Poultry Sci. 57: 670.

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

Vladimirov, Y.A., Olenew, V.I., Suslova, T.B., and Cheremisina, I. P. 1980. Lipid peroxidation in mitochondrial membrane. Adv. Lipid Res. 17: 173.

Wangen, R.M., Marion, W.W., and Hotchkiss, D.K. 1971. Influence of age on total lipids and phospholipids of turkey muscle. J. Food Sci. 36: 560.

Watt, B.M. 1961. The role of lipid oxidation in lean tissues in flavor deterioration of meat and fish. In "Processings Flavor Chemistry Symposium," p. 83. Compbell Soup Co., Camden, N.J.

Watt, B.M. 1962. Meat products. In "Symposium on Foods: Lipids and Their oxidation," H.W. Schultz, E.A. Day and R.O. Sinnhuber (Ed.), p. 202. AVI Publ. 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 muscles from several species as measured by TBA analysis. J. Agric. Food Chem. 24: 7.

Wu, T.W., Lee, M.H., Ho, C.T. and Chang, S.S. 1982. Elucidation of the chemical structures of natural antioxidants isolated from rosemary. J. Am. Oil Chem. Soc. 59: 339.

Wu, T.C. and Sheldon, B.W. 1988. Influence of phospholipids on the development of oxidized off flavors in cooked turkey rolls. 53: 55.

Yamauchi, K. 1972. Effect of heat treatment on the development of oxidative rancidity in meats and its isolated tissue fraction. Bull. Fac. Agric., Miyazaki Univ. 19: 147.

Yamauchi, K., Nagai, Y., and Ohashi, T. 1982. quantitative relationship between alpha-tocopherol and polyunsaturated fatty acids and its connection to development of oxidative rancidity in chicken skeletal muscle. Agric. Biol. Chem. 46: 2719.

Younathan, M.T., Marjan, Z.M. and Arshad, F.B. 1980. Oxidative rancidity in stored ground turkey and beef. J. Food Sci. 45: 274.

Zipser, M.W. and Watts, B.M. 1961. Lipid oxidation in heat-sterilized beef. Food Technol. 15: 445.

Zipser, M.W., Kwon, T. and Watts, B.M. 1964. Oxidative changes in cured and uncured frozen cooked pork. J. Agr. Food Chem. 12: 105.