THESIS

MICHIGAN STATE UNIVERSITY LIBRARIES
3 1293 00793 5806

This is to certify that the

thesis entitled

STABILIZATION OF TURKEY LIPIDS THROUGH DIETARY SUPPLEMENTATION WITH VITAMIN E

presented by

Kuang-Hua Hsieh

has been accepted towards fulfillment
of the requirements for

Master of Science degree in Food Science

J. J. Gray
Major professor

Date 8-4-92

O-7639

MSU is an Affirmative Action/Equal Opportunity Institution

LIBRARY
Michigan State
University

PLACE IN RETURN BOX to remove this checkout from your record.
TO AVOID FINES return on or before date due.

DATE DUE	DATE DUE	DATE DUE
JUN		
JUN 11 1996		
920		
12110		

MSU Is An Affirmative Action/Equal Opportunity Institution
c:\circ\datedue.pm3-p.1

STABILIZATION OF TURKEY LIPIDS THROUGH DIETARY SUPPLEMENTATION WITH VITAMIN E

BY

KUANG-HUA HSIEH

A THESIS

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

MASTER OF SCIENCE

Department of Food Science and Human Nutrition

1992

ABSTRACT

STABILIZATION OF TURKEY LIPIDS THROUGH DIETARY SUPPLEMENTATION WITH VITAMIN E

BY

KUANG-HUA HSIEH

The study was designed to examine the influence of three levels of dietary vitamin E (12.5, 50 and 100 IU/kg feed) on the deposition of α-tocopherol in turkey muscle, and on selected quality characteristics of raw turkey meat (oxidative stability of lipids, color, drip loss) during refrigerated and frozen storage. Muscle and membranal lipids from turkeys fed the higher levels of vitamin E (50 and 100 IU/kg feed) had significantly ($p < 0.05$) higher α-tocopherol concentrations and greater oxidative stability than those from turkeys fed the control diet. The beneficial effect of dietary vitamin E on color stability and drip loss was also observed during storage. The stability of cholesterol in cooked turkey meat during refrigerated storage was improved by dietary vitamin E. Cholesterol oxidation was significantly ($p < 0.05$) influenced by supplementation at the 100 IU vitamin E/kg feed level. Lipid oxidation and cholesterol oxide product formation were positively correlated in cooked dark and light turkey meat ($r=0.79$ and $r=0.55$).

ACKNOWLEDGMENTS

The author expresses sincere gratitude to her academic advisor, Dr. J. Ian Gray, for his guidance and consistent encouragement throughout my graduate studies.

Appreciation is extended to my committee, Dr. Alden Booren, Dr. James F. Price and Dr. Cal J. Flegal, for their technical assistance and critical review of this manuscript. Further acknowledgements are also due to Dr. A. Asghar and Dr. E. Gomma for their technical assistance.

Special thanks are to Mr. Napolitano Angelo, Poultry Science Research and Teaching Center, Michigan State University, for his assistance during turkey feeding periods.

The author recognizes laboratory associates Dr. Frank J. Monahan, Shu-Hui Chan and Cheng-Hsiung Chen for their assistance throughout this research program.

Finally, I would like to thank to express my heartfelt appreciation to my parents and family for all of the love, encouragement and support during this graduate study.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

INTRODUCTION

Oxidative deterioration of meat lipids can directly affect many quality characteristics of meat products including color, flavor, texture, nutritive value and even safety (Pearson et al., 1983). The lipids associated with the subcellular organelles are especially susceptible to oxidation by virtue of their high content of phospholipids containing relatively large amounts of polyunsaturated fatty acids (Pearson et al., 1977). Recent studies have shown the advantages of vitamin E supplementation at supranutritional levels in the diets of broilers (Asghar et al., 1989b, 1990; Lin et al., 1989b) and pigs (Buckley et al., 1989; Monahan et al., 1992b) in terms of increasing the oxidative stability of lipids in the subcellular membranes in muscle meat systems. In addition, dietary supplementation of vitamin E for the subsequent benefit of increased lipid stability in animal food products has been extensively reported. Furthermore, vitamin E might help to exploit the full growth potential of meat animals and yield better quality meat (Ashgar et al., 1991a).

More recently, interest in the possible toxicological effects of lipid oxidation products, particularly

cholesterol oxidation products (COPS), has increased (Addis and Park, 1989; Kubow, 1990). Animal studies provide some compelling evidence implicating dietary cholesterol oxides as initiators of atherosclerotic lesions in blood vessels (Taylor et al., 1979; Imai, 1980; Peng et al., 1985, 1987).

In a recent study, Engeseth (1990) reported that the environment in which cholesterol exists in muscle tissues may play an important role in determining its susceptibility to oxidation, and that the oxidation of nearby unsaturated lipids can result in the formation of free-radicals which may attack cholesterol. Therefore, the stabilization of membrane lipids by dietary vitamin E might enhance the stability of cholesterol in meat products.

The overall objective of this study was to evaluate the effects of three levels of dietary vitamin E on the growth performance of male turkeys, and on the quality of turkey meat during refrigerated and frozen storage. Specific objectives of the study included the evaluation of the effects of dietary vitamin E on lipid oxidation, including cholesterol, in cooked ground turkey meat, and on the peroxidative stability of membranal lipids.

LITERATURE REVIEW

Lipid Oxidation in Meat Systems

Lipid oxidation is one of the major deteriorative processes responsible for the loss in quality of frozen meat or precooked meat products during storage (Pearson et al., 1977, 1983; Tichivangana and Morrissey, 1985; Gray and Pearson, 1987). Oxidative deterioration of meat lipids causes undesirable changes in the color, flavor, texture, nutritive value, and safety of meat products (Pearson et al., 1983). Lipid oxidation occurs in all stored foods to some extent and there is some concern about the toxicity of oxidation products (Pearson, 1981). With increasing emphasis on the role of diet in human diseases such as coronary heart disease, cancer, and stroke (Senate Select Committee, 1977a,b), questions have been raised relative to the role of lipids and their breakdown products in the etiology of these diseases.

Mechanism of lipid oxidation

The overall mechanism of lipid oxidation consists of three distinct phases: initiation, propagation and termination (Gunstone and Norris, 1983).

Initiation:	RH	------> R. + H.
Propagation:	R. + O_2	------> ROO.
	ROO.+ RH	------> ROOH + R.
Termination:	R. + R.	------> RR
	R. + ROO.	------> ROOR
	ROO.+ ROO.	------> ROOR

The reaction is initiated when a labile hydrogen is abstracted from a site on the unsaturated lipid (RH), with the production of a lipid alkyl radical (R.). In the propagation step, the reaction of the alkyl radical with oxygen which yields a peroxyl radical (ROO.) is followed by the abstraction of another hydrogen. A hydroperoxide (ROOH) and another free alkyl radical (R.) capable of perpetuating the chain reaction, are formed. In the termination step, the chain reaction can be stopped by the interaction of free radicals which produces non-initiating and non-propagating products.

The important lipids involved in oxidation are the unsaturated fatty acid moieties, oleic, linoleic, and linolenic. The rate of oxidation of these fatty acids increases somewhat geometrically with the degree of unsaturation (Labuza, 1971).

Initiation and catalysis of lipid oxidation

Lipid oxidation in meat systems may be initiated and/or catalyzed by nonenzymic and/or enzymic reactions (Asghar et al., 1988; Rhee, 1988).

In nonenzymic catalysis, the meat pigment myoglobin has

been implicated by many studies as playing an important role in the catalysis of lipid oxidation in uncooked red meat (Greene, 1969; Govindarajan et al., 1977; Verma et al., 1984; Rhee et al., 1986, 1987). Prior to 1970, myoglobin was viewed as the major catalyst of lipid oxidation in meat (Tappel, 1952, 1953; Younathan and Watts, 1959). However, Liu (1970a,b) determined the effects of pH and additives on linoleate oxidation catalyzed by metmyoglobin (MetMb), Fe^{2+}-EDTA (1:1) chelate, and raw beef homogenates, and concluded that the catalytic activity of raw beef homogenates was due to both heme iron and nonheme iron. Later, Sato and Hegarty (1971), Love and Pearson (1974), and Igene et al.(1979) proposed that nonheme iron plays a major role in accelerating lipid oxidation in cooked meat and that heme iron does not catalyze the oxidative process. Tichivangana and Morrissey (1985) reported that purified metmyoglobin (MetMb), when added to water-extracted beef or lamb muscle fibers containing the indigenous lipids, exerted virtually no catalytic effect on lipid oxidation. Rhee et al. (1987) determined the effect of MetMb plus hydrogen peroxide (MetMb-H_2O_2) on lipid oxidation in a model meat system where MetMb-H_2O_2 was added to water-extracted beef muscle residue, rather than to a subcellular membrane fraction isolated from beef muscle. They demonstrated that the lipid-oxidizing activity of MetMb-H_2O_2 in the raw system may be due primarily to the activated MetMb and secondarily to the nonheme iron released from MetMb by H_2O_2.

Recently, Johns et al. (1989) reported that ferric heme pigments are more powerful catalysts of lipid oxidation than inorganic iron compounds in raw muscle residue systems. They suggested that the apparently contradictory results obtained in previous studies with muscle model systems may be due to differences in sample storage time over which oxidation was monitored, and to the levels of prooxidants incorporated into the model systems. Monahan et al. (1992a) also determined that myoglobin and hemoglobin were more effective prooxidants than $FeSO_4$ in raw and cooked muscle systems.

It is now generally accepted that these catalytic species decompose preformed hydroperoxides in cooked meat and are thus catalysts of the propagation stage of the oxidative process (Asghar et al., 1988). However, they do not account for the initiation of the oxidation process. A number of initiators of lipid oxidation in raw meat and other biological systems have been suggested and are summarized in Table 1. The extremely reactive hydroxyl ($HO^{\cdot}$) radical has received much attention as one such initiator and is formed in biological tissues either by the Haber-Weiss reaction or by the superoxide-driven Fenton reaction. The hydroxyl radical has the ability to abstract hydrogen from unsaturated lipids. These reactions occur by the following mechanisms:

Haber-Weiss reaction:

$$O_2^{-\cdot} + Fe^{3+} \text{------>} O_2 + Fe^{2+}$$

$$2O_2^{-\cdot} + 2H^{+} \text{------>} O_2 + H_2O_2$$

Fenton reaction:

$$Fe^{2+} + H_2O_2 \text{------>} Fe^{3+} + OH^{-} + HO\cdot$$

Many of the proponents of the hydroxyl radical theory hold the Fenton-type reaction to be responsible for hydroxyl radical formation in biological systems (Fong et al., 1976; Gutteridge, 1984; Wilson, 1984).

Table 1. Proposed initiators of lipid oxidation

Proposed initiators
Singlet oxygen
Superoxide radical
Hydroperoxyl radical
Hydroxyl radical
Crypto-hydroxyl radical
Perferryl radical
Ferryl radical
Oxygen-bridged di-iron
Porphyrin cation radical

From Asghar et al. (1988)

The initiation of lipid oxidation in biological membranes by perferryl and ferryl radicals has been extensively studied by Aust and his associates (Svingen et al., 1979; Tien and Aust, 1982). They reported that lipid oxidation is initiated by NADPH-cytochrome P450 reductase and by xanthine oxidase in liver microsomes (Figure 1). The

cytochrome P-450 reductase catalyzes the transfer of an electron from NADPH to ferric ion to generate ferrous ion, which can form a complex with dioxygen to produce perferryl or ferryl radicals. They assumed that these radicals have the ability to abstract hydrogen from polyunsaturated lipids to initiate the oxidation process. In these systems, chelation of iron by adenosine diphosphate (ADP) or a similar nucleotide or pyrophosphate was needed to enhance the solubility of iron in aqueous solution (Aust and Svingen, 1982).

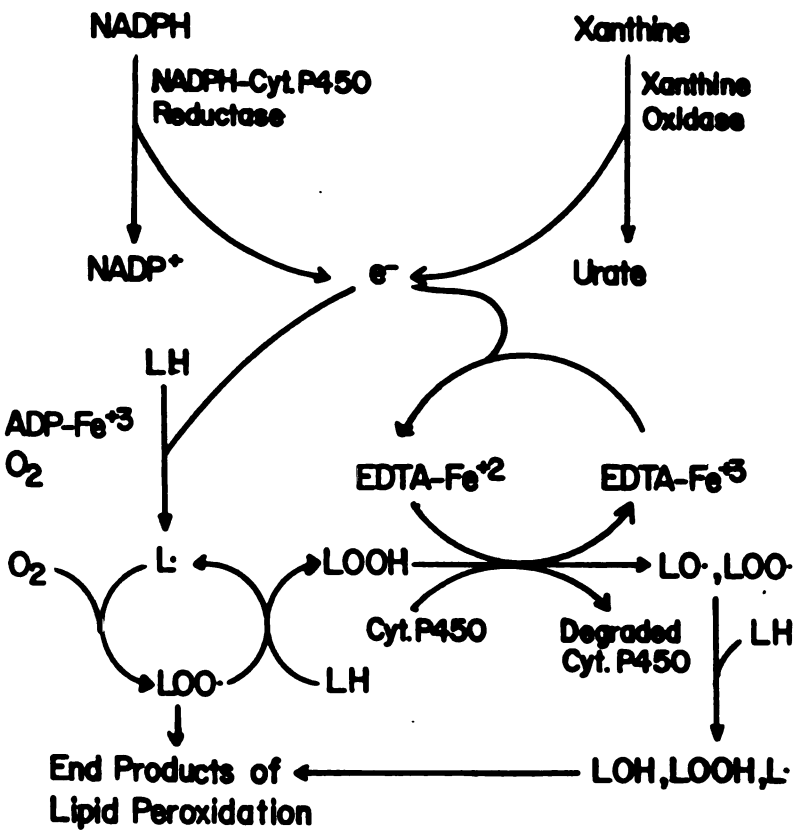


Figure 1. Proposed scheme of lipid peroxidation (Tien and Aust, 1982)

A direct involvement of heme protein (MetMb and methemoglobin) in lipid oxidation in muscle tissues has been suggested by Kanner and Harel (1985) and Harel and Kanner (1985). They reported that the interaction of H_2O_2 with MetMb generated activated MetMb capable of initiating lipid oxidation. The active MetMb complex is called the porphyrin cation radical ($P.^{+}- Fe^{4+} = O$). Initiation of lipid oxidation by the porphyrin cation radical proceeds via the two electron-reduction of the catalyst (Kanner and Harel, 1985):

$$P.^{+}- Fe^{4+} = O + RH \text{ ---------->} P - Fe^{4+} = O + R^{\cdot} + H^{+}$$

$$R^{\cdot} + O_2 \text{ ---------->} ROO^{\cdot}$$

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

$$P - Fe^{4+}= O + ROOH \text{ ---------->} P - Fe^{3+} + ROO^{\cdot} + OH^{-}$$

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

Later, Kanner et al. (1988a,b,c) reported that lipid peroxidation in muscle foods is initiated by "free" iron ions and hydrogen peroxide. This reaction is cycled by superoxide anion (Haber-Weiss) or by reducing compounds, e.g., ascorbic acid (redox cycling)(Kanner et al., 1987). Furthermore, Kanner et al. (1991) found the cytosolic extract of beef muscle can inhibit the activity of H_2O_2-activated myoglobin. Membrane lipid peroxidation initiated by an iron-redox cycle (ascorbic acid-ferric ions and NADPH enzymatic reaction) was inhibited partially by the high molecular weight fraction of the cytosolic extract, but

accelerated by the low molecular weight fraction.

Enzymatic oxidation of muscle microsomes has been reported in numerous studies (Hochstein and Ernster, 1963; Lin and Hultin, 1976; Rhee et al., 1984; German and Kinsella, 1985; McDonald and Hultin, 1987). Lipoxygenase catalyzes the hydroperoxidation of polyunsaturated fatty acids interrupted by a cis-methylene group (Tappel, 1963). Lipoxygenase has also been reported to initiate microsomal lipid oxidation in fish skin (German and Kinsella, 1985). Grossman et al. (1988) reported that lipoxygenase may be responsible for some of the oxidative changes occurring in fatty acids on frozen storage of chicken meat. Hultin and his colleagues (Lin and Hultin, 1976; Player and Hultin, 1977; McDonald et al, 1979; Hultin, 1980; Slabyj and Hultin, 1982) demonstrated the presence of enzymic systems in chicken and fish skeletal muscles that were capable of catalyzing the oxidation of microsomal lipids in the presence of cofactors, NADPH, ADP and ferric ions. Recent studies also revealed the presence of similar oxidation systems associated with beef and pork muscle microsomes (Rhee et al., 1984, 1985, 1987). The enzymic lipid peroxidation in skeletal muscle microsomes is dependent on NADH or NADPH - the relative effectiveness of the two differing among species - and requires ADP and Fe^{2+} or Fe^{3+} for maximum activity.

Current research data indicate that enzymes or enzyme systems and possibly activated metmyoglobin are responsible

for initiation of lipid oxidation in raw muscle tissues and that metal catalysts (e.g., nonheme iron in red meat) also promote lipid oxidation in raw tissues, apparently through their role in the propagation of free radicals.

Composition of poultry lipids

Lipids in meat are commonly classified as depot or adipose lipids and as intramuscular or tissue lipids (Watts, 1962; Love and Pearson, 1971). The depot fats are largely present as subcutaneous deposits and are appreciably present in the thoracic and abdominal cavities and between the muscles as intermuscular deposits. Depot fat tissue consists mainly of triacylglycerols which are composed of straight-chain, even-numbered carbon fatty acids, typically containing 16 or 18 carbon atoms (Dugan, 1971). The predominant fatty acids of depot fat of poultry are saturated or contain only one or two double bonds (Pearson et al., 1977).

Tissue lipids are composed of deposits of triacylglycerols in fat cells, commonly called marbling in red meats, and membrane-bound lipids consisting chiefly of the phospholipids and lipoproteins (Love and Pearson, 1971). The phospholipids are susceptible to oxidation (Buege and Aust, 1978) and are believed to be the site of initiation of the oxidative process (Asghar et al., 1988) because of their high levels of polyunsaturated fatty acids (Marion and Woodroof,1966; Love and Pearson, 1971; Salmon and O'Neil,

1973; Igene et al., 1980; Melton, 1983). Although the phospholipid content of meat is relatively small, it still is very important in determining meat quality.

The differences in fatty acid composition of the triacylglycerol and phospholipid fractions increase ongoing successively from poultry to pigs through ruminants, in which the polyunsaturated fatty acids are confined largely to the phospholipid fraction (Shorland, 1962). The main components of phospholipids are phosphatidylcholine (PC), phosphatidylethanolamine (PE), phosphatidylinositol (PI), phosphatidylserine (PS), sphingomyelin (SP), and other minor components. Dawson et al. (1990) reported the fatty acids from the neutral lipid fraction contributed 94% of the total lipid fatty acids of chicken meat and the fatty acids from phosphatidylcholine and lysophosphatidylethanolamine fractions contributed 38% of the total phospholipid fatty acids. Within the phospholipid fraction, the lysophosphatidylethanolamine fraction has the greatest concentration of polyunsaturated fatty acids (70%) and is believed to be the most important phospholipid component contributing to development of warmed-over flavor (WOF). Pikul and Kummerow (1991) also indicated that the PC, PE, PI, and PS fractions accounted for 86-97% of the total PL, and that PC and PE were present in greater amounts than the other phospholipid fractions in chicken liver, heart, plasma, and egg yolk.

The structural and metabolic functions of phospholipids

indicate that they are actively involved in oxidation during processing and storage. Igene et al. (1980) demonstrated that during frozen storage of model meat systems, the polyenoic fatty acids of PC and PE are not stable and undergo oxidation. The role of phospholipids in the formation of thiobarbituric acid-reactive substances (TBARS) has been extensively studied by Pikul et al. (1984), while Pikul and Kummerow (1991) reported that the concentration of TBARS and the percentage of polyenoic fatty acid, especially arachidonic acid, were highest in PI followed by PE, PS, PC and SP.

The composition of tissue lipids is relatively constant (Koch et al., 1968). There is still considerable variation in the phospholipid content among species (Kaucher et al., 1944; Waku and Uda, 1971; Tahin et al., 1981), from location to location within the carcass (Dugan, 1971), and as a consequence of the activity of muscle (Bloor, 1943; Acosta et al., 1966). The lipid content and composition of different poultry tissues vary considerably and influence the oxidative potential (Dawson and Gartner, 1983). The lipid content of turkey breast muscle is approximately half of that in thigh muscle (Wilson, 1974). 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 that in dark meat (Wangen et al., 1971; Pikul et al.,1984). On a

weight-percentage basis, the white and dark meat of chicken contain about the same amount of phospholipid (Katz et al., 1966).

Lipid oxidation products

Lipid oxidation in muscle foods leads to the formation of short-chain aldehydes, ketones, and fatty acids, as well as to the development of some polymers (Frankel, 1962; Keeney, 1962; Lundberg, 1962), all of which are believed to contribute to the undesirable flavor in meat, poultry and fish (Olcott, 1962; Watts, 1962; Love and Pearson, 1974). Hydroperoxides which do not contribute to off-flavor are the primary products of the reaction of oxygen with unsaturated lipids. The hydroperoxides are unstable and can undergo carbon-carbon cleavage on either side of the alkoxy radical. The alkoxy radicals also react with other radicals. These reactions account for many of the aliphatic aldehydes, alcohols, ketones, and hydrocarbons (Frankel, 1984) which are secondary products of lipid oxidation and which are responsible for the oxidized flavor in meat (Lillard, 1987; Love, 1987).

Aldehydes, such as hexanal and heptanal, are major contributors to the loss of desirable flavor in meats because of their high rate of formation during lipid autoxidation and their low flavor thresholds (Ullrich and Grosch, 1987). Hexanal is the major autoxidation product of linoleic acid (Badings, 1970; Ullrich and Grosch, 1987).

Alcohols contribute measurably less to the undesirable flavor quality of meat than the aldehydes due to the relatively higher flavor thresholds (Drumm and Spanier, 1991). Pentanol and 1-octen-3-ol are the major alcohols formed during the storage of beef patties (Ullrich and Grosch, 1987). Drumm and Spanier (1991) found that the major ketones developed during the refrigerated storage of cooked beef were 2-heptanone and 2,3-octanedione. Hydrocarbons, on the other hand, have high flavor thresholds and make minimal contribution to desirable or undesirable flavors (Min et al., 1979; Frankel, 1985). Further oxidation may occur in the original peroxides or in the unsaturated aldehydes, which then undergo further degradation to form epoxides, cyclic peroxides and bicyclic endoperoxides (Enser, 1987).

The interaction between lipids and protein/amino acids is contributed to by both lipid hydroperoxides and their secondary products of decomposition. Hydroperoxide radicals are very reactive with sulfur and amino functional groups of amino acids, whereas aldehydes and epoxides react with thiols from cysteine (Gardner, 1979). Cysteine and methionine side chains are especially susceptible to these oxidative, free radical reactions (Gardner, 1983; Lillard, 1987; Ladikos and Lougovois, 1990). Non-enzymic browning (Maillard) reactions also give rise to loss in nutritional value as well as to organoleptic changes. Carbonyls, such as aldehydes and dialdehydes, form Schiff bases with amines

and contribute less to the rancid odor; however, high molecular weight brown materials known as melanoidins, resulting from polymerization of Schiff bases, are unstable and produce new volatiles which affect flavor characteristics, especially during cooking and processing (Frankel, 1984). The Maillard reaction produces acyclic sulfur compounds, aromatic and non-aromatic heterocyclic compounds, and lactones which contribute to off-flavors (Chang and Peterson, 1977; Golovnja and Rothe, 1980; Shibamoto, 1980; Bailey, 1983; Shahidi et al.,1986). Protein solubility, emulsification and water-binding capacity associated with texture and rheological properties, also appear to be affected by the interaction between lipid oxidation products and proteins (Hall, 1987).

Cholesterol Oxidation in Meat Systems

Cholesterol, a member of the family of animal sterols, is the most abundant sterol in animal tissue, and occurs predominantly either in the free form or esterified to any one of a number of long chain fatty acids (Kritchevsky and Tapper, 1961; Smith et al., 1983).

Cholesterol distribution in meat

In an earlier report, Stromer et al. (1966) reported that the *Longissimus* beef muscle contained 36-46 mg cholesterol /100g raw tissue, while the subcutaneous adipose tissue contained 113-150 mg cholesterol/100g lipid. Rhee et

al. (1982a) determined the cholesterol concentration in raw beef intermuscular fat (108 mg/100g raw tissue) and subcutaneous fat (114 mg/100g raw tissue) to be much higher than that of raw beef *Longissimus* muscle (62 mg/100g raw tissue).

Most researchers agree that adipose tissue has a much higher concentration of cholesterol (mg per 100g tissue, wet weight) than muscle tissue. Furthermore, researchers have sought to clarify the distribution of cholesterol between the muscle and the intramuscular fat by studying cholesterol distribution at the subcellular level. Hoelscher et al. (1988) found that the membrane contained 60-80% of the total cholesterol in muscle, while the cytosolic storage component contained 20-40%. In the adipose tissue, the membrane contained 8-12%, while the cytosolic storage component contained 88-92% of the total cholesterol. These investigations also showed that most of cholesterol in muscle tissue was contributed by the membrane fraction, while cholesterol in the adipose tissue, was contained primarily in the cytosolic storage fraction. They also demonstrated that increasing adipose tissue did not increase the total cholesterol content but changed the cholesterol distribution in muscle. Sweeten et al. (1990) determined the cholesterol content and subcellular distribution within intramuscular adipose tissue (marbling) and reported an average cholesterol concentration of 118 mg per 100 g intramuscular adipose tissue, with 54% in the cytoplasm and

46% in the membrane.

Mechanism of cholesterol oxidation

Cholesterol, being an unsaturated lipid, is susceptible to oxidation (Smith, 1981, 1987; Maerker, 1987), and cholesterol oxidation products (COPS) have been detected in food products of animal origin (Missler et al., 1985; Park and Addis, 1985a, 1987; Nourooz-Zadeh and Appelqvist, 1989; Pie et al., 1991).

Cholesterol may be oxidized *in vivo* chemically, enzymically by specific enzymes, and perhaps by lipid peroxidation products (Smith et al., 1982). Initial stable cholesterol autoxidation products are the epimeric cholesterol 7-hydroperoxides, with the quasiequatorial 7β-hydroperoxide predominating (Teng et al., 1973a; Smith et al., 1973b), formed by reaction of ground-state molecular oxygen ($^3O_2$) with a C-7 cholesteryl radical (Figure 2). Formal reduction of the 7-hydroperoxides yields the corresponding 7-alcohols, cholest-5-ene-3β,7α-diol (7-α-hydroxycholesterol) and cholest-5-ene-3β,7β-diol (7-β-hydroxycholesterol). Formal dehydration yields the 7-ketone, 3α-hydroxycholest-5-en-7-one (7-ketocholesterol) (Van Lier and Smith, 1970a; Smith et al., 1973a; Teng et al., 1973b). Dehydration of the 7-ketone gives cholesta-3,5-dien-7-one. The formation of epimeric cholesterol 5,6-epoxides during the autoxidation of cholesterol is facilitated by direct attack of an unoxidized cholesterol

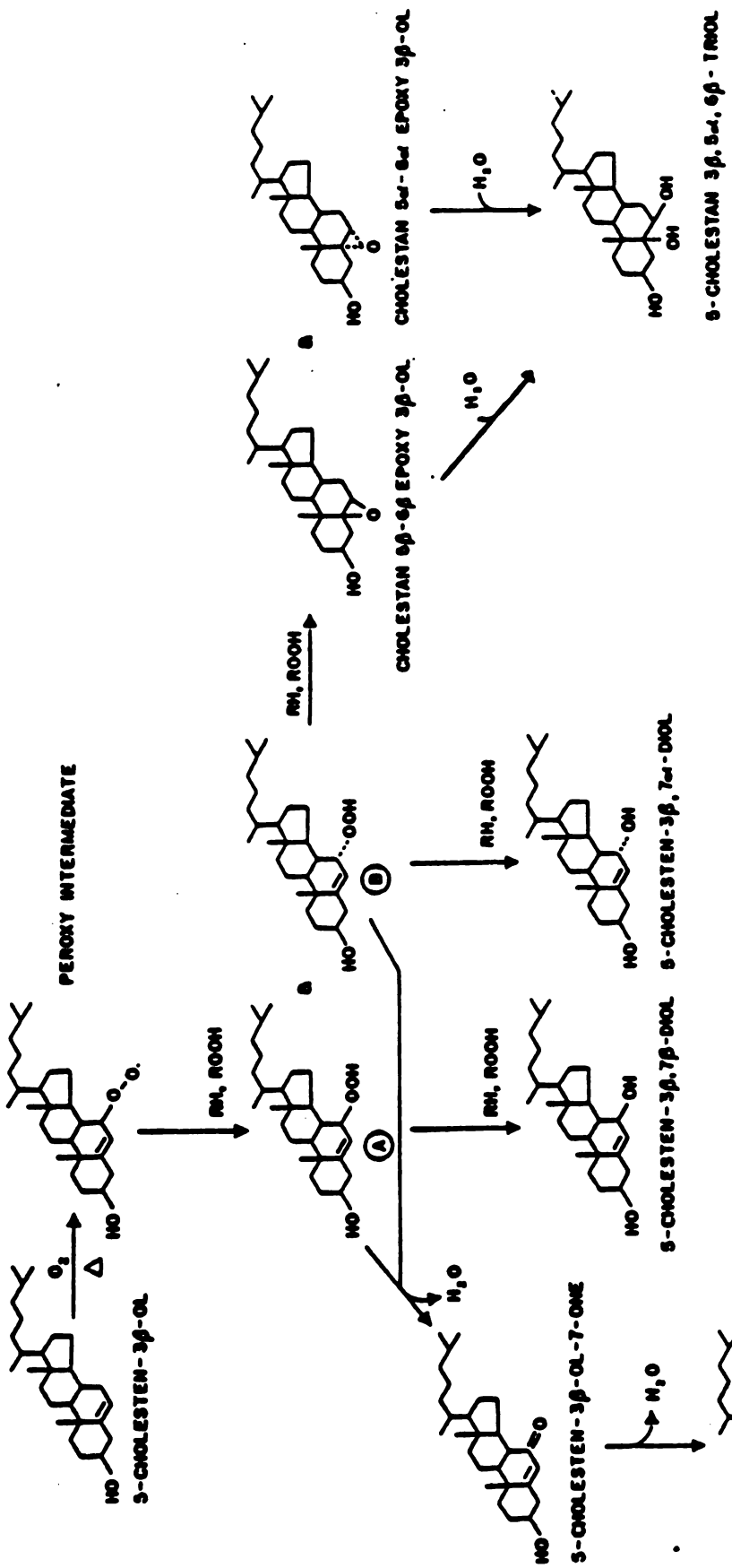


Figure 2. Scheme for cholesterol oxidation (Luby, 1982)

molecule by either 7α- or β-hydroperoxy cholesterol to form cholestan-5,6α-epoxy-3β-ol and cholestan-5,6β-epoxy-3β-ol, respectively. Hydration of either 5,6-epoxides gives 5α-cholestane-3β,5,6β-triol (Smith and Kulig, 1975). Other autoxidation reactions occur, including attack on the side chain to form hydroperoxides, chief among which is cholesterol 25-hydroperoxide from which cholest-5-ene-3β, 25-diol (25-hydroxycholesterol) is derived (Van Lier and Smith, 1970a,b; Smith et al., 1973a; Teng et al., 1973b; Smith and Kulig, 1975).

Cholesterol oxidation products in foods

Recently, the possible oxidation of cholesterol in foods has caused some concern due to undesirable biological implications of COPS. These products may interrupt sterol metabolism, and also produce cytotoxic, angiotoxic, mutagenic and even carcinogenic responses (Kandutsch et al., 1978; Taylor et al., 1979; Smith, 1981; Addis et al., 1983; Addis, 1986). Many studies have demonstrated the presence of COPS in animal foodstuffs such as egg products (Missler et al., 1985; Morgan and Armstrong, 1987; Nourooz-Zadeh and Appelqvist, 1987), heated tallow (Bascoul et al.,1986; Park and Addis, 1986), dairy products (Nourooz-Zadeh and Appelqvist, 1988; Sander et al., 1989), and meat products (Higley et al., 1986; Park and Addis, 1987; De Vore, 1988; Engeseth, 1990; Pie et al., 1991; Zubillaga and Maerker, 1991).

Eight common COPS have been identified: 25-hydroxycholesterol, cholestane-triol, α- and ß-epoxide, 7α- and 7ß-hydroxycholesterol, 7-ketocholesterol and cholesta-3,5-dien-7-one, which is actually an artifact derived from 7-ketocholesterol during isolation (Peng et al., 1978, 1979; Smith, 1980, 1981; Tsai, 1984; Missler et al., 1985; Park and Addis, 1985a,b, 1987; Nourooz-Zadeh and Appelqvist, 1987).

Egg products have been the most extensively studied as they have high cholesterol contents. Missler et al. (1985) analyzed a scrambled egg mix which had been stored 5 years and identified 7 COPS. Nourooz-Zadeh and Appelqvist (1987) did not detect COPS in fresh egg yolk, but a dehydrated egg mix stored for 12 months contained α- and ß-epoxide, 7α- and 7ß-hydroxycholesterol and 7-ketocholesterol. After 8 years of storage, a dehydrated egg yolk sample contained the above COPS plus 25-hydroxycholesterol and cholestane-triol. Finocchiaro et al. (1984) reported COPS in 4 of 8 brands of grated cheeses but not in fresh ungrated cheeses. No COPS were found in freshly bleached butter oils but after storage for 90 days at 15°C, the 7-hydroxycholesterols were identified.

Park and Addis (1987) reported finding similar cholesterol oxides in various meats, with 7-ketocholesterol being most predominant. Cholestane-triol, a very atherogenic cholesterol oxide, was also detected for the first time in a meat sample. Sander et al. (1989) reported

7-ketocholesterol and the two epoxides in dehydrated chicken, beef and turkey, but the method which involved solvent extraction (Folch et al., 1957) and overnight cold saponification was somewhat cumbersome. Zubillaga and Maerker (1991) demonstrated that the concentrations of 7-keto-cholesterol and α- and ß-epoxide in raw veal, beef, pork and chicken muscle tissues are less than 1 mg/kg, while 7-ketocholesterol constituted more than 50% of the cholesterol oxidation products. Engeseth (1990) reported 7-ketocholesterol, 7-ß-hydroxycholesterol and the ß-epoxide as the predominant cholesterol oxides in veal and beef samples.

The study of Sander et al. (1989) indicated that the concentrations of COPS in fresh products were generally lower than those in processed products. Foods containing the greatest concentrations of COPS were dehydrated egg and meat products. Generally, the greater the cholesterol content and the more severe the conditions of processing, the greater the resultant COP content. Park and Addis (1987) and Monahan et al. (1992b) noted greater cholesterol oxide development with increasing degrees of rancidity (as determined by TBARS value) in meat. Pie et al. (1991) also reported that cooking and frozen storage lead to cholesterol oxidation in meat products. The percentage of oxysterols to cholesterol varied on average from 0.5 to 0.8%. Engeseth (1990) also showed that dietary vitamin E supplementation decreased cholesterol oxidation in veal because the membranal lipids would be stabilized by α-tocopherol, thus

protecting cholesterol from oxidation. Monahan et al. (1992b) demonstrated that after two days of refrigerated storage, cholesterol oxides represented 2.7% of the initial cholesterol content of cooked pork from pigs fed a basal level of vitamin E (10 mg/kg diet) and 1.6% of the initial cholesterol content of cooked pork from pigs fed a supplemented diet (200 mg/kg diet).

Stabilisation of Lipids with Antioxidants

Phenolic antioxidants are compounds capable of slowing the rate of oxidation in fat or fat-containing foods. Their action is due to donation of hydrogen or an electron which reacts with free radicals to form inert products, thus terminating the chain reaction mechanisms of lipid oxidation (Dziezak, 1986). The stability of lipids can be improved by antioxidants, such as synthetic phenolic antioxidants (Chastain et al., 1982) and natural antioxidants (Pratt, 1972; Rhee et al., 1983; Rhee, 1987).

Synthetic phenolic antioxidants

Synthetic phenolic antioxidants include butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), propyl gallate (PG) and tertiarybutyl hydroquinone (TBHQ). A number of studies have indicated that these antioxidants can effectively inhibit lipid oxidation in meat (Greene, 1969, Greene et al., 1971; Chastain et al., 1982). However, there are two major limitations to the use of synthetic

antioxidants in meats. First, they are approved for use in only a limited number of meat products and each new synthetic antioxidant must be subjected to a lengthy and costly process of evaluation for proof of safety. Second, consumers are increasingly concerned about the use of chemical additives in foods (Rhee, 1987).

Naturally occurring antioxidants

A promising approach to retarding lipid oxidation in meat products is through the use of naturally occurring antioxidants. Compounds possessing antioxidative activity have been found in vegetable extracts (Pratt, 1965), citrus products (Park et al., 1983), oilseed products (Whittern et al., 1984), oat flour (Daniels and Martin, 1968), rosemary extracts (Houlihan and Ho, 1985) and retorted meat extracts (Einerson and Reineccius, 1978). The predominant antioxidant compounds include flavonoids, phenolic acids, tocopherols, and phenolic terpenes (Rhee, 1987). Flavones and flavonoids have molecular structures which make them useful as antioxidants. Their antioxidant activity, which is related to the ketone in the 4 position of the pyrone ring and the 2,3-olefin group, has been investigated by several workers (Simpson and Uri, 1959; Metha and Seshadri, 1959; Crawford et al., 1961). However, the flavones (e.g., quercetin) have structural characteristics associated with mutagenic and perhaps carcinogenic activity (McGregor and Lard, 1978). Oleoresin rosemary (OR) is one of the

important spices which has antioxidant properties and contains a number of compounds such as rosmanol, carnosol, rosmaridiphenol, and rosmariquinone which impart antioxidant activity similar to or greater than that of BHT (Houlihan and Ho, 1985). Lai (1989) found that sodium tripolyphosphate (STPP)/OR and STPP/TBHQ both effectively retarded the oxidative degradation of polyunsaturated fatty acids in chicken nuggets during frozen storage. Stoick et al. (1991) also demonstrated that OR was an effective antioxidant which, when combined with sodium tripolyphosphate, produced an additive protective effect in restructured beef steak during frozen storage. Development of such products as antioxidants is desirable because they are derived from naturally occurring substances. Recently, St. Angelo et al. (1990) reported that lipid oxidation in ground beef patties was retarded by the addition of metal chelators, free radical scavengers, rosemary and sodium alginate.

Alpha-Tocopherol in Meat Systems

Alpha-tocopherol is the most prominent natural antioxidant in meat (Dugan, 1976; Watts, 1962). It is a powerful antioxidant and functions as a lipid-soluble free radical scavenger (Linder, 1985). It has been extensively studied, with its antioxidant properties being widely used in meat products.

Alpha-tocopherol distribution

At least eight compounds with vitamin E activity ((α-, ß-, γ-, δ-tocopherol and α-, ß-, γ-, δ-tocotrienol) have been isolated from plant sources. All have a 6-chromonal ring structure and a side chain. The tocols have a side chain, and the trienols have a similar structure, with double bonds at the 3', 7', and 11' positions of the side chain. Both tocols and trienols occur as a variety of isomers that differ by the number and location of methyl groups on the chromanol ring. Alpha(α)-tocopherol is the most active form of vitamin E, and in foods of animal origin it accounts for almost all the vitamin E activity (Bauernfeind, 1980).

Many studies on the distribution of tocopherols in animal tissue have been made for several species. The uptake of tocopherol by tissues varies directly with the logarithm of the tocopherol intake (Bieri, 1972; Gallo-Torres, 1980). This is illustrated by data on the tocopherol content of lung in rats and in the muscle of chickens (Machlin, 1991). This relationship represents a departure from what occurs with most other vitamins, which usually have distinct deposition thresholds in tissues other than the liver and may provide an explanation for the pharmacological effects of vitamin E. Tissues vary considerably in their vitamin E levels with no consistent relationship to lipid parameters. Asghar et al. (1989b) reported tissue α-tocopherol concentration in swine in the

following order: liver >heart >lung >kidney. Monahan et al. (1989) found the same trend in tissue samples of pigs supplemented with 200 international units (IU) vitamin E/kg feed. However, Engeseth (1990) reported the concentrations of α-tocopherol for the tissues of veal calves decreased in the following order: liver >adipose tissue >kidney >lung >heart. With supplementation, the same order of tocopherol concentration was maintained, but adipose tissue had the highest concentration of α-tocopherol because it is one of the major storage deposits of vitamin E. In addition, the rate of depletion of tocopherol in adipose tissue is slower than in other tissues (Machlin, 1980). Tissue distribution studies demonstrated significant increases in α-tocopherol of various tissues; however, the percentage increase of each tissue varied from study to study.

The overall deposition of α-tocopherol in the muscle tissues was studied by Lin et al. (1989a). They reported that approximately 0.88% of the total α-tocopherol consumed by the broilers was deposited in the tissues, with 0.56% in the dark meat and 0.32% in the light meat. The different rates of deposition apparently result from physiological variations in the vascular network of dark and white muscle tissues (Sheldon, 1984). Dark meat from broilers has a more highly developed vascular system and a higher lipid content than white meat. Therefore, α-tocopherol is deposited to a greater degree in dark meat than in white meat.

In membranes, the earliest reliable estimate of the

subcellular distribution of α-tocopherol was made by Krishnamurthy and Bieri (1963) who found that 52-62% of the total tocopherol in the homogenate of rat liver was in the mitochondria, 21-23% was in the microsomal fraction, 6-9% was in the nuclei and 15-19% was in the soluble fraction. Mellors and Barnes (1966), on the other hand, reported values of 14%, 12%, 44%, 31% for the nuclei, mitochondria, microsome and supernatant fraction, respectively. These results are clearly different from those of previous workers. Recent studies reported higher α-tocopherol concentrations in microsomes than in mitochondria. (Asghar et al., 1989b; Monahan et al., 1989; Engeseth, 1990).

The biochemical function of alpha-tocopherol

It has been proposed that vitamin E plays a role in the regulation of heme biosynthesis (Nair, 1972), acts as a modulator in immune response (Bendich et al., 1986) and in gene regulation (Patnaik and Nair, 1977), and protects the selenium-containing proteins (Caygill et al., 1971). The primary function of the tocopherols is as antioxidants in hydrophobic environments. All of the four tocopherol vitamers (α-, β-, γ-, δ-tocopherol) are effective chain-breaking antioxidants in bulk unsaturated lipids. However, since α-tocopherol is the predominant tocopherol found in human and animal biomembranes, most studies have focused on this vitamer.

The commonly accepted theory for initiation and

propagation of free radical-mediated oxidation and their inhibition by antioxidants is outlined in Table 2.

Table 2. Autoxidation and antioxidant reactions involving vitamin E

1. Initiation (formation of a free radical)
   $LH \xrightarrow{\text{Initiators}} L\cdot$
2. Reaction of radical with oxygen
   $L\cdot + O_2 \longrightarrow LO_2\cdot$
3. Propagation
   $LO_2\cdot + LH \longrightarrow L\cdot + ROOH$
4. Antioxidant reaction
   $LO_2\cdot + E \longrightarrow E + LOOH$
5. Regeneration
   $E\cdot + C \longrightarrow E + C\cdot$
   $C\cdot + NADPH \xrightarrow{\text{Semidehydro ascorbate reductase}} C + NADP$
   $E\cdot + 2GSH \xrightarrow{\text{Enzyme?}} E\cdot + GSSG$
   $GSSG + NADPH \xrightarrow{\text{Glutathione reductase}} 2GSH + NADP$
6. Termination
   $E\cdot + E\cdot \longrightarrow E\text{-}E$ (dimer)
   $E\cdot + LO_2\cdot \longrightarrow EOOL$ (?)

Abbreviation: LH, fatty acid; $L\cdot$, fatty acid radical; $LO_2\cdot$, peroxy radical; E, tocopherol; $E\cdot$, tocopheroxy radical; LOOH, hydroperoxide; C, ascorbic acid; $C\cdot$, ascorbyl radical; GSH, reduced glutathione; GSSG, oxidized glutathione. (Machlin, 1991)

Alpha-tocopherol is currently believed to act as a quencher of oxygen- or carbon-centered fatty acyl radicals or active oxygen via donation of a hydrogen atom from the phenyl hydroxyl of the chromanol ring. The rate of reaction of α-tocopherol and other phenols with peroxyl radicals of styrene under conditions of controlled peroxyl radical formation has been reported by Burton and Ingold (1981, 1986) and Burton et al. (1985). Their results demonstrate

that the radical quenching (hydrogen donating) capability of α-tocopherol exceeds that of the other tocopherols and synthetic phenolic antioxidants, and thus may explain in part the superior vitamin E activity of the α-vitamer relative to the β-, γ-, and δ- forms. Apparently, α-tocopherol is a more effective hydrogen donor than the other tocopherols since electron-releasing methyl groups ortho to the phenolic hydroxyl, lacking in the other vitamers, stabilize the phenoxyl radical and increases the rate of the reaction:

$$ROO\cdot + ArOH \longrightarrow ROOH + ArO\cdot$$

The number of alkyl radicals with which each phenoxyl group reacts is approximately two (Burton and Ingold, 1981).

The efficient antioxidant activity of α-tocopherol, despite its low concentration in biological membranes relative to that of polyunsaturated fatty acids, suggests that the effective half-life of the vitamin in membranes is rather long. Several studies have indicated that the chromanoxyl radical of α-tocopherol may be reduced to regenerate α-tocopherol by ascorbic acid (Packer et al., 1979) or glutathione (Pryor, 1976).

Vitamin E is a special antioxidant compared with synthetic antioxidants for the following reasons: (1) α-tocopherol is located primarily in the membrane where free radicals are generated by lipid oxidation (Molenaar et al., 1972); (2) α-tocopherol acts as a free radical scavenger and thus protects against lipid oxidation (Linder, 1985); (3) α-

tocopherol could be regenerated by ascorbic acid or glutathione to maintain a longer half life in the membranes.

Molenaar et al. (1980) proposed that α-tocopherol is situated in the membranes such that the chromanol ring is at the polar surface and the phytol side chain is in contact with the unsaturated fatty acids of the phospholipids in the nonpolar interior. This positioning of α-tocopherol allows effective radical scavenging as it would be adjacent to the membrane-bound enzymes such as NADPH oxidase, which generates free radicals. McCay and King (1980) reported that the superoxide anion ($O_2^{\cdot -}$) interacts with hydrogen ions to produce hydrogen peroxide, which is distributed in both the aqueous and membrane phases of the cell. Glutathione peroxidase, a selenium-containing enzyme, destroys the hydrogen peroxide in the aqueous phase, thus shifting most of the hydrogen peroxide into the membrane. Any hydrogen peroxide remaining in the membrane may react with superoxide anion to form hydroxy radicals ($^{\cdot}OH$), which can react with tocopherol localized in the membrane. If insufficient tocopherol is available to trap $^{\cdot}OH$, this extremely reactive radical may initiate peroxidation of the polyunsaturated fatty acid in the membrane. This hypothesis accounts for many experimental observations, including the role of selenium (as part of glutathione peroxidase), in alleviating some of the symptoms of vitamin E deficiency. Some of these concepts are summarized in Figures 3. and 4.

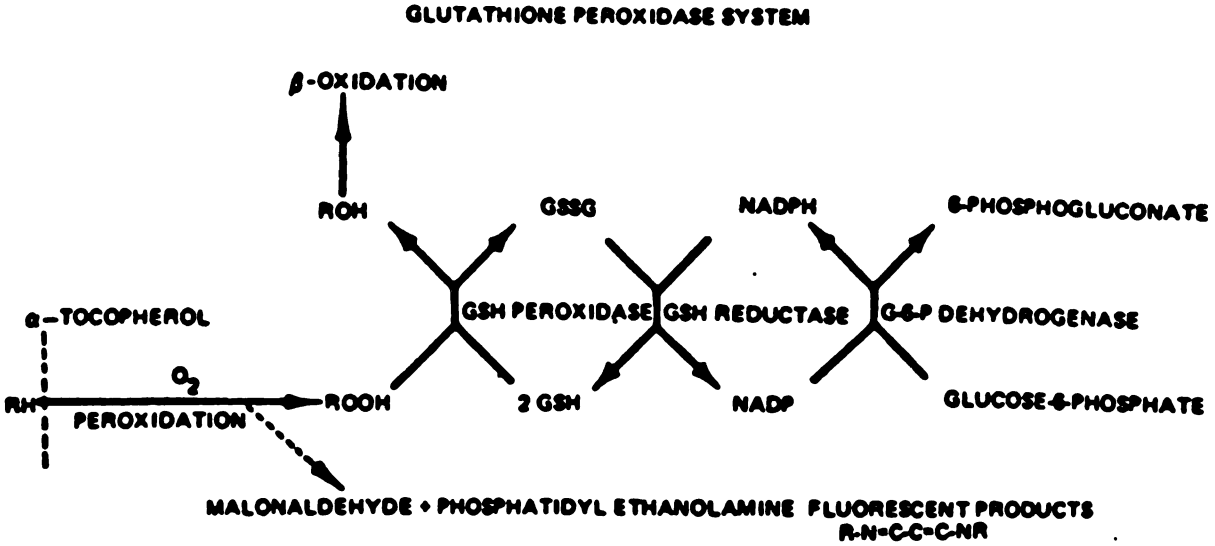


Figure 3. Glutathione peroxidase system. RH: Polyunsaturated fatty acid; ROOH: fatty acid hydroperoxide; ROH: hydroxy fatty acid; GSH: reduced glutathione; GSSG: oxidized glutathione (From Machlin, 1984)

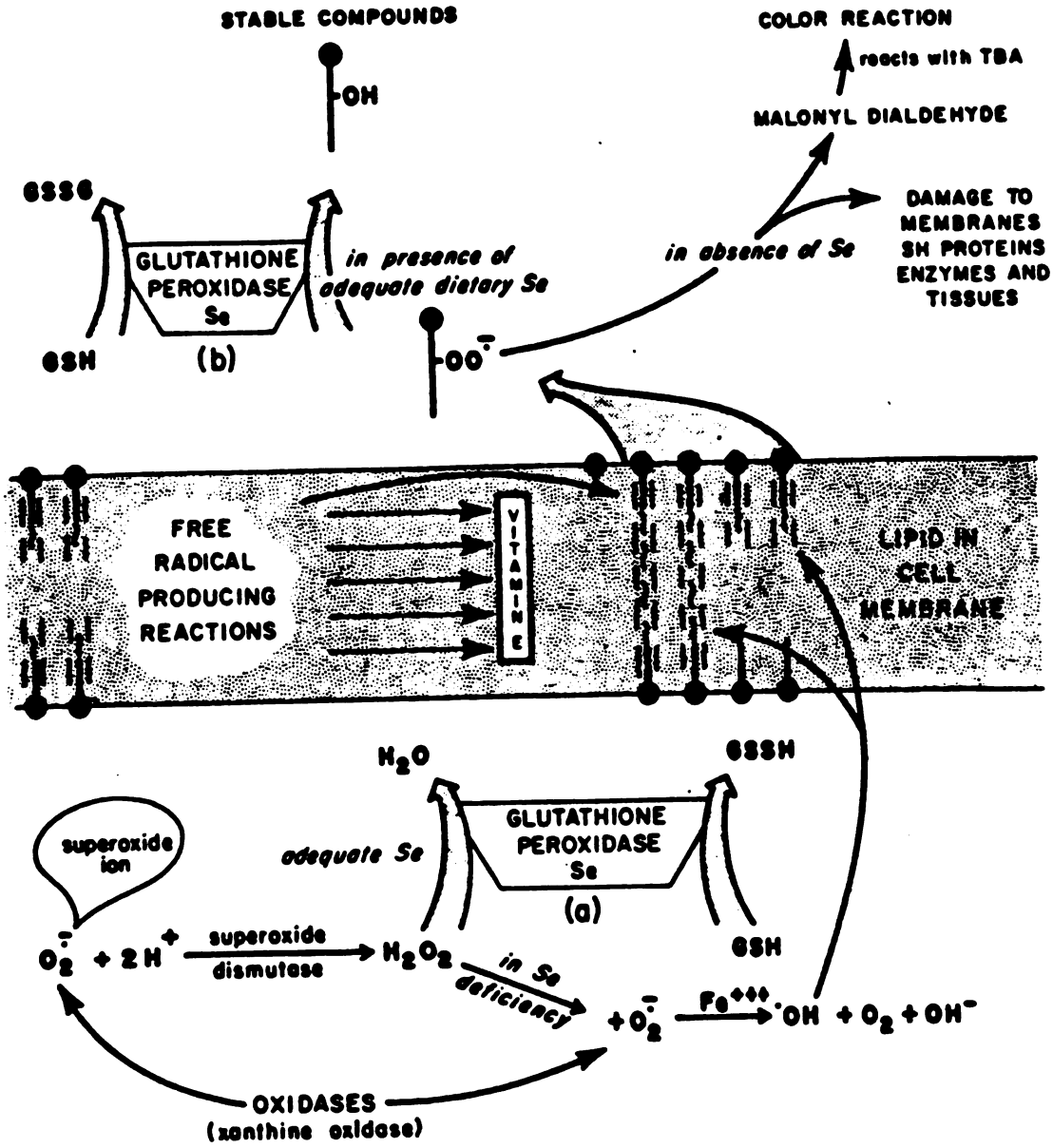


Figure 4. Protection of membranal lipids by α-tocopherol (From Machlin, 1984)

Effect of Dietary Vitamin E Supplementation on the Quality Characteristics of Meat

Lipid oxidation plays a major role in the loss of quality of muscle foods during storage (Pearson et al., 1983). Off-flavor development in meat has been attributed to the formation of a complex mixture of carbonyls during the autoxidation of unsaturated fatty acids (Reindl and Stan, 1982; Morrissey and Apte, 1988). Oxidation products also adversely affect color, texture, nutritive value (Pearson et al., 1983) and even the safety of meat (Addis, 1986).

Vitamin E is known to be the major lipid antioxidant of nature because of the benefits of its biochemical function which were mentioned in the previous section. Tocopherols are absorbed by the animal, transported, and deposited in body lipids. Deposition is usually relatively uniform throughout the animal body on a lipid basis. Incorporation of α-tocopherol into the muscle prior to slaughter by dietary supplementation with vitamin E has a more beneficial antioxidative effect than addition of α-tocopherol to meats just prior to processing (Faustman et al., 1989b). This is expected, as it is difficult to distribute lipid-soluble antioxidants to the sites of lipid oxidation in intact meats (Watts, 1961; Faustman et al., 1989b). Free radical scavengers are most effective during the initial stages of oxidation so that a build-up of hydroperoxides can be prevented. It is well established that vitamin E

supplementation of diets results in increased α-tocopherol concentrations in the tissue and a concomitant increase in the stability of chicken (Sklan et al., 1983; Sheldon, 1984; Lin et al., 1989b), pork (Buckley and Connolly, 1980) and veal (Shorland et al., 1981; Engeseth, 1990).

Buckley and Connolly (1980) reported lower thiobarbituric acid reactive substances (TBARS) during the storage of fresh and frozen raw pork from swine which were fed a vitamin E-supplemented diet. Buckley et al. (1989) evaluated the effects of dietary vitamin E (200 IU/kg feed) on the oxidative stability of pork chops and ground pork during refrigerated and frozen storage. They found that the stability of ground pork patties from the pigs fed the short-term (4 weeks) and long-term (10 weeks) vitamin E supplements was not significantly different. However, they did report that the vitamin E-supplemented pork was significantly ($p < 0.05$) more stable than the control pork. In a further study, Asghar et al. (1991b) evaluated the influence of three levels of dietary vitamin E (10, 100 and 200 IU/kg feed) on the subcellar deposition of α-tocopherol in the muscle and on certain quality characteristics of pork (oxidative stability of lipids, color and drip loss). The concentrations of α-tocopherol in the adipose and muscle tissues increased with increasing levels of dietary vitamin E, with concomitant decreases in TBARS, color loss and drip loss. Erin et al. (1986) proposed that α-tocopherol protects the membrane from the action of phospholipases

which remove the fatty acid moiety from phospholipid molecules. The loss of phospholipids from membranes causes a substantial reduction in their fluidity (Kameda et al., 1985) and hence may increase drip loss.

Poultry meat is very susceptible to oxidative rancidity during frozen storage (Pearson et al., 1977), although it can be stabilized by dietary vitamin E supplementation (Webb et al., 1972; Uebersax et al., 1978; Bartov et al., 1983). Marusich et al. (1975) found that turkeys required higher levels of vitamin E supplementation than broiler chickens to delay the onset of rancidity. Recently, Lin et al. (1989b) investigated the effects of dietary oil (with different degrees of unsaturation) and vitamin E supplementation on lipid composition and stability of broiler meat. They found the broilers which were fed the diets containing the more highly unsaturated oils had more unsaturated fatty acids in the muscle tissue, while dietary vitamin E significantly improved the oxidative stability of dark and white meat during refrigerated and frozen storage.

The effect of vitamin E supplementation on pigment and lipid stability of beef has been studied by Faustman et al. (1989a). They reported that steaks from Holstein steers fed 370 IU vitamin E/day maintained their red color longer during storage than those from the control animals. Faustman et al. (1989b) reported more than a two-fold increase in the concentrations of α-tocopherol in the steaks from the supplemented animals. Pigment and lipid stability

was improved by the absorption of α-tocopherol and its incorporation into the cellular membranes. Engeseth (1990) also evaluated the effect of vitamin E supplementation on membranal lipid peroxidation and meat lipid stability (including cholesterol). She proposed that the oxidative stability of raw and cooked veal muscle held at 4°C was improved by vitamin E supplementation and cholesterol oxide development was reduced in cooked meat samples from the supplemented animals.

Effect of Dietary Vitamin E Supplementation on the Growth Performance of Animals

Very few reports of growth performance, as affected by vitamin E supplementation, have been published. In an early study, the negative effect of oxidized dietary oil on the growth performance of rats was investigated by Burr and Barnes (1943). Oxidized dietary oil when fed to experimental animals (rats and pigs) causes various types and degrees of abnormalities. Severe oxidation causes a readily detected illness in broilers called encephalomalacia. Century and Horwitt (1959) found that vitamin E can prevent encephalomalacia which is a staxis symptom resulting from hemorrhages and edema within the molecular and granular layers of the cerebellum in chickens by functioning as a biological antioxidant and interrupting free radical-initiated lipid peroxidation. Shermer and Calabotta (1985) reported that broilers are routinely fed

diets containing slightly oxidized fat and this can adversely affect the growth performance of the broilers, thereby reducing the profits of the poultry grower. Waldroup et al. (1960) reported that turkeys receiving diets containing the synthetic antioxidant, ethoxyquin, were significantly heavier than birds receiving diets containing no antioxidant. This might be due to the prevention of oxidation of oil in the diet. Shermer and Calabotta (1985) also reported that alpha-tocopherol effectively delayed the symptoms associated with the presence of oxidized oils in the diet. Studies by Lin et al. (1989a) revealed that BHA, BHT and α-tocopherol can improve growth when added to broiler diets. These investigators also reported that short-term and long-term vitamin E supplementation significantly ($P < 0.05$) increased body weights of the broilers.

Asghar et al. (1991a) demonstrated the positive effects of supranutritional levels of vitamin E (100 and 200 IU/kg feed) on the performance of growing pigs, and feed conversion efficiency and carcass yield. Their studies confirmed the hypothesis that undesirable lipid peroxidation that possibly occurs in the subcellular membranes and other tissue components during normal growth and development of meat animals can be minimized by vitamin E supplementation of the diet at supranutritional levels. On the other hand, Asghar et al. (1991a) found high vitamin E supplementation may prevent growth enhancement because ingestion of large

amounts of vitamin E may inhibit absorption of vitamin A, which is an important vitamin for growth (Bieri et al., 1983).

EXPERIMENTAL

Cholesterol Oxide Standards

Cholesterol oxide standards used in this study were: cholest-5-en-3β-ol (cholesterol), cholest-5-ene-3β, 7α-diol (7α-hydroxycholesterol), cholest-5-ene-3β, 7β-diol (7β-hydroxycholesterol), cholest-5-ene-3β, 20α-diol (20α-hydroxycholesterol), cholest-5-ene-3β, 25-diol (25-hydroxycholesterol), 5-cholesten-3β-ol-7-one (7 ketocholesterol), cholestan-3β, 5α, 6β-triol (triol), cholestan-5β, 6β-epoxy-3β-ol (β-epoxide), cholestan-5α, 6α-epoxy-3β-ol (α-epoxide), 5α-cholestan-3β-ol-6-one (6-ketocholesterol) and 5α-cholestane. These chemicals were obtained from Steraloids Inc., Wilton, NH.

Reagents

Bis-(trimethylsilyl)-trifluoroacetamide (BSTFA) and pyridine were obtained from Pierce Chemical Co. (Rockford, IL). All other reagents used in the experiments were analytical grade.

Turkey Feeding Regimen

One hundred and eight, day-old male turkeys (Cooper Hatchery, Oakwood, OH) were randomly divided and placed into nine pens (12 turkeys/pen). The pens of birds were randomly assigned to receive one of three dietary treatments (3 pen/treatment). The three dietary treatments consisted of standard turkey feed formula (Table 3) containing different levels of vitamin E supplementation: (1) 12.5 IU vitamin E (as α-tocopherol acetate, BASF Corp, Wyandotte, MI)/kg feed, (2) 50 IU vitamin E/kg feed, and (3) 100 IU vitamin E/kg feed. The turkeys were grown to 18 weeks of age at the Poultry Science Research and Teaching Center, Michigan State University. The birds were weighed initially and at 6, 12 and 18 weeks, and body weights were individually recorded. The feed consumed by each pen of turkeys was recorded to determine feed efficiency. At the end of the feeding trial, four turkeys from each pen were slaughtered in the poultry processing lab of Animal Science, Michigan State University and immediately prepared for analysis as described below.

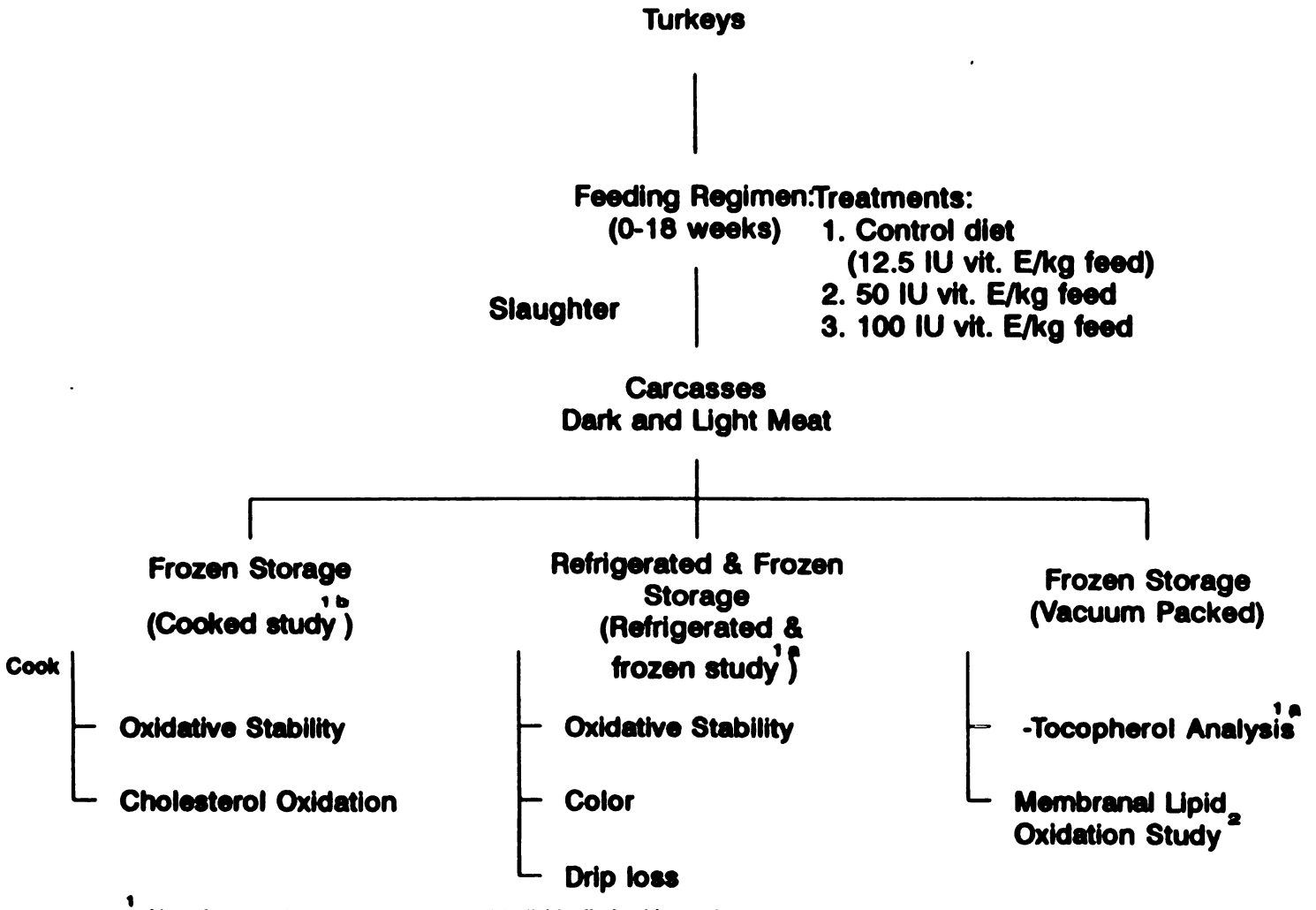
Sample Preparation

The carcasses were cut and separated by manual skinning and deboning. Dark (thigh) and light (breast) meat from the individual turkeys were separately wrapped and stored properly for analysis (Figure 5). For the refrigerated study, dark or light meat samples were placed on polystyrene trays, wrapped in oxygen-permeable PVC meat stretch-wrap

Table 3. Composition of turkey rations used in the feeding study

Ingredient	Pounds/ton of feed				
	Turkey Starter	Turkey Grower 1	Turkey Grower 2	Turkey Finisher 1	Turkey Finisher 2
Corn, yellow	737	743.5	990	1180.5	1421
Soybean meal (44%)	1082	1042	815.5	616	370
Meat and bone meal (50%)	50	50	50	50	50
Fat	49	86	80	89	92
Dicalcium phosphate	36	36	27	29	31
Limestone	26	26	22	21	21
Salt	8	8	8	8	8
Vitamin/trace minerals	8	7.5	6.5	6.5	6
Methionine	4	1	1	---	1
Total	2000	2000.0	2000.0	2000.0	2000
%Protein	28.5	27.5	23.6	20.0	15.7

Turkey Starter; 0-2 weeks; 1.6 lb of feed/bird.
Turkey Grower 1; 15-56 days; 16.8 lb of feed/bird.
Turkey Grower 2; 57-77 days; 15.5 lb of feed/bird.
Turkey Finisher 1; 77-112 days; 28.0 lb of feed/bird.
Turkey Finisher 2; 113-126 days; 15.0 lb of feed/bird.



[1] Meat from each turkey was analyzed individually in this study.a:4 birds/pen; b:2 birds/pen

[2] Meat from turkeys within the same group was pooled together in this study.

Figure 5. Flow diagram of the feeding, slaughtering, and analytical protocols.

(Reynolds Metal Company, Richmond, VA), and kept at 4°C under fluorescent light for 8 days to determine the oxidative stability, color change and drip loss of the raw meat individually during storage. For the frozen study, dark or light meat samples were wrapped in oxygen-permeable PVC meat stretch-wrap, stored at -20°C, and evaluated for oxidative stability after 3 and 6 months. Samples of the frozen meat were removed after 3 and 6 months (thawed at room temperature for one hour), and evaluated for changes in oxidative stability, color and drip loss during refrigerated storage. The oxidative stability of lipids in cooked meat from each turkey (previously frozen raw for 3 months) was determined by grinding the meat, cooking in plastic bags in a water bath at 70°C for 30 minutes, and then storing at 4°C. Dark or light meat from each turkey was vacuum-packed and kept at -20°C for α-tocopherol analysis. Dark and light meat samples from each treatment were pooled for the membranal lipid oxidation studies and stored at -20°C .

Measurement of Lipid Oxidation in Muscle Tissue

Lipid oxidation in the muscle tissues from four turkeys per pen was monitored using a modification of the 2-thiobarbituric acid (TBA) distillation procedure of Tarladgis et al. (1960). Ten grams of finely cut-up muscle tissue from individual turkeys were blended in a tissue homogenizer (Ultra-Turrax, Model #SDF 1810, Tekmar Co., Cincinnati, OH) with 10 ml of antioxidant solution (5%

propyl gallate and 5% ethylene-diaminetetraacetic acid (PG/EDTA) in distilled water). The homogenate was transferred to a distillation flask (500 ml), using 87.5 ml of deionized water. To the flask were added 2.5 ml HCl/H_2O (2:1) and 2 drops of antifoam agent (10% silicone emulsion in water, Thomas Scientific, Swedesboro, NJ). The sample was distilled until 50 ml of distillate were collected in a 50 ml screw-capped test tube. A 5 ml aliquot of the distillate was added to 5 ml of TBA reagent (0.2883 g TBA/100 ml distilled water) in a 150 x 18 mm screw-capped test tube. The capped tube was placed in a boiling water bath for 35 minutes. After the tube was cooled to room temperature, the color intensity was quantitated by measuring the absorbance at 532 nm with a Bausch and Lomb Spectronic 2000 spectrophotometer (Bausch and Lomb, Rochester, NY). Absorbances were converted to TBARS (TBA-reactive substances) values by multiplying by a factor of 6.2 (Crackel et al., 1988) and expressing as mg malonaldehyde (MDA)/kg tissue.

Isolation and Quantitation of Alpha-Tocopherol in Blood Plasma and Muscle Tissue

(a) Plasma:

Blood samples were taken from three turkeys per pen at specific times using 10 ml heparinized syringes. The blood was centrifuged (speed 4, IEC Clinical Centrifuge-International Equipment Co., Needham Height, MA) within 30

minutes of collection and the plasma fraction stored at -20°C until required. After thawing, 1 ml of the plasma was transferred to a screw-capped test tube containing 0.5 ml of a 5% aqueous ascorbic acid solution (prepared with distilled water) and the internal standard, 100 µl of an ethanolic solution (0.2 mg/ml) of α-tocopherol acetate (Sigma Chemical Co., St. Louis, MO). After flushing with nitrogen and capping, the tube was vortexed on a Big Vortexer (Glass-Col Apparatus Co., Terre Haute, IN) for 5 seconds. Ethanol (1.5 ml) was then added and the tube was flushed with nitrogen and vortexed for approximately 15 seconds. Hexane (5 ml) was added and the tube was vortexed for 4 minutes after flushing with nitrogen. The tube was centrifuged at 1000 x G until two layers were clearly identified. The hexane (top) layer was transferred by a disposable pipette to another clean test tube. Extraction with another 4 ml aliquot of hexane was repeated, and the hexane layers were combined and evaporated under a stream of nitrogen. When the hexane was completely evaporated, 200 µl of ethanol were added to the tube, which was then quickly flushed with nitrogen, vortexed and frozen until required for HPLC analysis.

(b) Muscle tissue:

Ground muscle tissue (5 g) from four turkeys per pen was homogenized with 5 ml of distilled water in a 50 ml beaker. One gram of the homogenate was placed in a screw-capped test tube, to which were added 2 ml 1% ethanolic

pyrogallol solution and 0.3 ml 50% KOH. The tube was flushed with nitrogen, capped and vortexed for 1 minute. The tube was placed in a water bath (70°C) for 30 minutes. After cooling to room temperature, 4 ml hexane were added to the tube which was recapped and vortexed for 1 minute. After centrifuging at 100 x G for 2 minutes, the upper layer was removed carefully to a clean screw-capped test tube. The hexane extraction step was repeated once more and the hexane extracts combined. After washing two times with distilled water and drying over anhydrous Na_2SO_4, the hexane extract was evaporated to dryness under a stream of nitrogen. The residue was redissolved in 200 µl ethanol and vortexed for 30 seconds. The tube was flushed with nitrogen, capped and stored at -20°C for HPLC (high performance liquid chromatographic) analysis.

(c)Microsomal fraction:

The microsomal fractions of control and vitamin E-supplemented turkey muscles were isolated by a centrifugation procedure similar to that reported by Asghar et al. (1990). Samples (100 g) of light or dark meat (pooled from 12 turkeys in each group) were homogenized with one liter of buffer solution (0.1M KCl, 0.2% glycerol, 0.025M Tris-HCl buffer) in a blender for 2 minutes and centrifuged at 1000 x G for 10 minutes in a Sorvall RC2-B centrifuge (Ivan Sorvall Inc. Norwack, Conn.) to isolate the sarcoplasmic components. The sarcoplasmic extracts were then subjected to the differential centrifugation procedure

of Schenkman and Cinti (1978) to separate the mitochondrial and microsomal fractions. The mitochondria extracts were centrifuged at 10,000 x G for 15 minutes. Calcium chloride was added to the supernatant to give a final concentration of 8 mM for aggregating the microsomes. The mixture was centrifuged at 13,200 x G for 30 minutes to separate the microsomal fraction. The subcellular fractions were not purified further to remove contaminating myofibrillar proteins and fragments of the plasma membrane.

The microsomes were assayed for α-tocopherol in a manner similar to that for muscle, using 0.5 g membrane to which were added 0.5 ml of deionized water, 2 ml 1% ethanolic pyrogallol and 0.3 ml 50% KOH.

(d) HPLC quantitation of alpha-tocopherol:

A Waters HPLC system (Water Associates, Milford, MA) with a reverse phase C_{18} column (ODS, Beckman Ultrasphere, 5 μ, 4.6 mm x 15 cm, Beckman Instruments Inc, Fullerton, CA) and a fixed wavelength detector (Waters, 440 absorbance detector) set at 280 nm was used. The mobile phase was 100% methanol at a flow rate of 1 ml/minutes. Peak areas were integrated using a HP 3380A integrator. Standard curves were prepared using dl-α-tocopherol (Sigma Chemical Co., St. Louis, MO) as described previously by Asghar et al. (1991b).

Color Analysis

Raw dark or light turkey meat from four turkeys per pen was individually placed in polystyrene petri dishes (100 x

15 mm) which were wrapped with oxygen-permeable PVC meat stretch-wrap film (Reynolds Metal company, Richmond, VA). Color changes in the raw dark and light turkey meat during refrigerated storage under fluorescent light were monitored by recording the L (luminance), a (redness) and b (yellowness) values using a Hunterlab, Model D25-2, tristimulus colorimeter (Fairfax, VA).

Determination of Drip Loss

Samples (80 g) of dark or light meat from four turkeys per pen was individually placed on polystyrene trays with absorbent pads. The meat was stored at 4°C under fluorescent light. Drip loss was determined by weighing individually the dark and light turkey meat on specific days and recording the decreasing weight (drip loss). The drip loss was expressed as a percentage of the original weight of the meat. After weighing, the absorbent pads in the trays were replaced before repackaging the turkey meat.

Determination of Cholesterol Oxides in Cooked Turkey Meat

Cholesterol oxides in cooked muscle (dark and light) samples from six turkeys per treatment were determined following the method of Monahan et al. (1992b). Total lipid extracts were prepared from 5 g cooked muscle samples by the method of Marmer and Maxwell (1981). The lipid extracts, in dichloromethane/methanol (90:10), were evaporated to dryness on a rotary evaporator (Brinkmann Instruments Inc.,

Westbury, NY) and redissolved in 4 ml 9:1 hexane/ethyl acetate. COPS were separated from cholesterol and other muscle lipids using the sample clean-up procedure of Park and Addis (1985b). Prior to sample clean-up, 6-keto-cholesterol was incorporated into the hexane/ethyl acetate extracts as an internal standard. Acetone extracts were evaporated to dryness by rotary evaporation and redissolved in 4 ml ethyl acetate. Prior to gas chromatographic (GC) analysis, 1 ml of the ethyl acetate solution was evaporated to dryness under nitrogen and trimethylsilyl (TMS) derivatives of cholesterol oxides were prepared by redissolving the COPS in 100 µl BSTFA (Pierce Chemical Co., Rockford, IL) and holding at room temperature for 30 minutes. The TMS ether derivatives of the sterols were then evaporated to dryness under nitrogen and redissolved in 100 µl ethyl acetate. A Hewlett Parkard 5890 gas chromatograph (Hewlett Parkard, Avondale, PA) equipped with a flame ionization detector, HP 5673A automatic injector, and a HP 3392A integrator, was used to quantify COPS. The COPS were separated on a fused silica capillary column DB-1 (15m x0.25mm id, film thickness 0.25 µm, J & W Scientific, Folsom CA). Carrier gas (helium) was delivered at 1.2 ml/min. Oven temperature programming was as follows: 170°C to 220°C at 10°C/min and held isothermally at 220°C for 5 min; 220°C to 234°C at 0.4°C/min; 234°C to 255°C at 1.5°C/min and held isothermally at 255°C for 40 min. Injector port and detector temperatures were 275°C and 330°C, respectively.

Four μl of the TMS derivatives of the COPS were injected onto the column with a split ratio of 11:1.

Cholesterol Determination

Cholesterol was extracted from cooked meat (dark and light) from six turkeys per group following the direct saponification procedure of Adams et al. (1986). The method involved the accurate weighing of the ground meat sample (3 g) into a flat bottom distillation flask to which 8 ml 50% KOH and 40 ml reagent alcohol (95:5 v/v ethanol-methanol) were added. The mixture was saponified while refluxing and stirring for 1 hour. Sixty ml of reagent alcohol were added and the mixture was cooled. Toluene (100 ml) was added and the mixture was vigorously stirred for 1 minute. The mixture was extracted with 110 ml 1N KOH in a 500 ml separatory funnel, shaking vigorously for 30 seconds, and then discarding the lower aqueous layer upon separation. To the toluene layer was added 40 ml 0.5N KOH and the separatory funnel was rotated gently. The lower layer was again discarded, and the extract was washed five times with water (100 ml) with increasing degrees of agitation. The toluene extract was dried over anhydrous Na_2SO_4. A 50 ml aliquot was removed and rotary evaporated to dryness, washed with 5 ml of acetone, dried, and redissolved in 3 ml dimethylformamide. The 3 ml extract was kept frozen in a glass vial (15 x 45 mm, 1 dram) for GC analysis. Cholesterol in the extract was quantified on a Hewlett

Packard 5840A gas chromatograph equipped with a flame ionization detector and a packed (1% SE-30 on 100/120 Gas Chrom Q) columns. The carrier gas was helium with a flow rate of 25 ml/minute and the GC was operated isothermally at 230°C. Injector port and detector temperatures were 225°C and 275°C, respectively.

Oxidative Stability of the Microsomal Lipids of Turkey Muscle

Membranal lipid oxidation studies were performed by the method reported by Kanner and Harel (1985). Microsomes were solubilized in a buffer solution which contained 0.1M KCl and 0.01M lactic acid and adjusted to pH 5.7 with NaOH. The protein content of the isolated fraction was determined by the method of Lowry et al. (1951). Membrane solutions of equal protein concentrations (0.4 mg/ml) were prepared at a final volume of 9.5 ml in a 150 ml erlenmeyer flask. The flasks were placed in a shaking water bath (GCA/Precision Scientific, Chicago, IL) at 37°C. Metmyoglobin (30 μM) and H_2O_2 (30 μM) were immediately added to initiate the oxidation reaction. Aliquots (1 ml) were taken initially (time 0) and after 15, 30, 60, 90, 120, 150 and 180 minutes. The aliquots were added to 2 ml TBA reagent (TBA:trichloroacetic acid:HCl, 1:25:6.25) in 100 x 15 mm screw-capped test tubes which were then capped and vortexed. At the end of the three hour assay, the tubes were placed in a boiling water bath for color development. The tubes were

centrifuged for 2 minutes at speed 7 (IEC Clinical centrifuge, Needham Heights, MA) prior to determination of the absorbance at 532 nm with a Bausch and Lomb Spectronic 2000 spectrophotometer. Using an extinction coefficient of 1.56×10^5 $M^{-1}cm^{-1}$ (Harel and Kanner, 1985), concentrations of malonaldehyde per gram of protein of the membrane solutions were calculated.

Statistical analysis

The analysis of data for dark and light turkey meat was performed using a three factor (treatment x time x replication) repeat-measure split-plot design for TBARS value, COP concentration, color change, and drip loss. The Improved Bonferronit t-test was used to determine the significance of means for multiple comparisons. The analyses of variance were performed using a SPSS-X statistical program (Computer Service Center, Michigan State University, East Lansing, MI). A two factor (treatment x replication) complete, randomized design was used for α-tocopherol concentration. Tukey's test was used to determine the significance of means for multiple comparisons. The analyses of variance were performed using MSTAT-C microcomputer statistical program (Michigan State University, East Lansing, MI, 1989).

RESULTS AND DISCUSSION

Effect of Dietary Vitamin E Supplementation on Growth Performance

Total feed consumption, body weight and feed conversion data are presented in Table 4. When the turkeys were fed the vitamin E-supplemented diets (50 and 100 IU/kg), their average body weights tended to be higher (3.7% and 2.2%) than the average body weight of the turkeys fed the control diet (12.5 IU vitamin E/kg feed). There was no significant ($p < 0.05$) differences, however, between the average body weights and feed conversion ratios for the different groups. These results confirmed the observations of Monahan et al. (1990b) for pigs fed vitamin E-supplemented diets. Similarly, Engeseth (1990) reported no advantage with respect to growth for calves fed vitamin E supplements.

Very few reports of growth performance, as affected by vitamin E supplementation, have been published. Popekhina and Tveritnev (1982) observed that α-tocopherol supplementation of swine diets produced pigs which were 10.6% heavier than pigs fed a basal diet. Ashgar et al. (1991b) reported 4 and 6% increases in daily body weight gain in the early growth stages (first 4 weeks) of pigs fed diets supplemented with 100 and 200 IU vitamin E /kg feed,

Table 4. Effects of dietary vitamin E level on feed consumption, feed conversion and body weights of turkeys

	Control	50 IU vitamin E /kg feed	100 IU vitamin E /kg feed
Total body weight (kg)[1]	460.6	505.4	498.1
Total feed consumption (kg)	1309.3	1388.5	1362.3
Feed conversion ratio[2]	2.9±0.1[a]	2.8±0.2[a]	2.7±0.1[a]
Average body weight (kg)[3]	13.5±1.2[a]	14.0±1.0[a]	13.8±1.2[a]

[1]Total body weight: 36 turkeys for the 50 and 100 IU vitamin E/kg treatments; 34 turkeys for the control (12.5 IU/kg) group

[2]Ratio = total feed consumption/total body weight gain
The values in the table are the group means of data from 36 or 34 turkeys (3 pens)

[3]The values in the table are the group means of data from 36 or 34 turkeys ± standard deviation.

[3]Mean values in a row with the same superscript are not significantly different ($p > 0.05$)

respectively. At later stages of the feeding period, the growth of the supplemented animals was parallel to that of the control animals; thus, the advantage persisted. Stuber et al. (1990), however, found slightly increased gain-to-feed ratios in supplemented pigs (100 and 200 IU/kg feed) as compared to controls (11 IU/kg feed).

Lin et al. (1989a) reported that short-term (10 days) and long term (6 weeks) vitamin E supplementation (200 mg/kg diet) significantly increased carcass weights of broilers ($p < 0.05$). Short-term supplementation produced increases of 1.8% and 2.5% in body and carcass weights, respectively, while long-term supplementation resulted in increases of 1.6% and 3.7%, respectively, when compared to the corresponding weights of the broilers fed a control diet.

Effect of Vitamin E Supplementation on Alpha-Tocopherol Concentration in Plasma and Muscle Tissue

Turkeys were bled every four weeks in order to determine the α-tocopherol concentrations in the blood plasma. The results are presented in Table 5. Although there were wide variations in the plasma α-tocopherol levels within a group, as is evident from the magnitude of the standard deviations, plasma concentrations of the supplemented animals were significantly higher ($p < 0.05$) than those from control animals during the entire period. At the 4th, 8th and 12th weeks of the feeding period, the α-

Table 5. Alpha-tocopherol content (μg/ml plasma) in plasma samples of turkeys fed three dietary levels of vitamin E

Age (weeks)	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
4	2.26 ± 0.40[c]	3.29 ± 0.35[b]	5.05 ± 0.66[a]
8	1.65 ± 0.03[c]	3.62 ± 0.20[b]	5.09 ± 0.22[a]
12	1.78 ± 0.15[c]	3.51 ± 0.05[b]	4.75 ± 0.16[a]
18	1.58 ± 0.15[b]	3.51 ± 0.32[a]	4.26 ± 0.23[a]

Values represent means of data from 9 turkeys ± standard deviation

Mean values having the same superscript in a row are not significantly different ($P < 0.05$)

tocopherol concentrations in the plasma from the different groups were significantly ($p < 0.05$) different. These results agree with those of Asghar et al. (1991a) and Engeseth (1990) who reported that supplementing the diets of swine and veal calves with vitamin E increased plasma α-tocopherol concentrations relative to those of the control animals. Monahan et al. (1989) also measured plasma α-tocopherol concentrations in pigs at the time of slaughter and observed significantly higher concentrations ($p< 0.01$) in plasma from supplemented animals. The deposition of α-tocopherol in plasma is different between animal species. In this study, the concentrations of α-tocopherol in the plasma from the control turkeys and those fed 100 IU vitamin E/kg feed were higher than the plasma concentration in pigs fed comparable levels of vitamin E (Asghar et al., 1991a).

The amounts of α-tocopherol deposited in the dark and light meat of turkeys fed the respective diets are shown in Table 6 and Appendix 1. There were significant ($p < 0.05$) differences in α-tocopherol concentrations between the control and supplemented groups for both light and dark meat. The α-tocopherol concentrations in dark meat from the turkeys fed supplemented diets (50 and 100 IU vitamin E/kg feed) were approximately 4 and 6 times greater than those in meat of the control animals. Similarly, the α-tocopherol concentrations in the light muscle from the turkeys fed the supplemented diets were approximately 4 and 5 times greater than those in the control animals. These results agree with

Table 6. Concentration of α-tocopherol (μg/g tissue) in the light and dark meat of turkeys fed three dietary levels of vitamin E

	Dietary level of vitamin E (IU/kg feed)		
Muscle	Control	50	100
Light	0.44 ± 0.07[c]	1.59 ± 0.19[b]	2.13 ± 0.07[a]
Dark	0.83 ± 0.20[b]	3.35 ± 0.77[a]	4.59 ± 0.45[a]

Values are the group means of data from 12 turkeys ± standard deviation

Mean values having the same superscript in a row are not significantly different from one another ($p < 0.05$)

those of previous studies (Sheldon, 1984; Lin et al., 1989b; Asghar et al., 1991a) in which it was demonstrated that dietary vitamin E supplementation could increase the α-tocopherol concentrations in muscle tissue. Sheldon (1984) reported that turkeys receiving 275 IU dl-α-tocopherol acetate/kg feed for three weeks before slaughter had mean α-tocopherol concentrations 3 to 3.3 times greater than tissues from turkeys fed control diets (1.63 IU/kg feed). Lin et al. (1989b) also reported that the concentration of α-tocopherol in the light and dark meat from broilers fed 100 IU vitamin E/kg feed were approximately 5 and 9 times greater than corresponding concentrations in the light and dark meat from broilers fed a control diet. Asghar et al. (1991a) reported that the concentrations of α-tocopherol in meat from pigs fed 10, 100, 200 IU vitamin E/kg feed showed sequentially increasing concentrations of tissue α-tocopherol (0.5, 2.6, 4.7 μg/g tissue).

The species of animal and muscle type (dark and light) might affect the absorption and deposition of vitamin E. Supplementing broiler diets with 100 IU vitamin E/kg feed resulted in a greater deposition of α-tocopherol (Lin et al., 1989b) into muscle tissue than was observed in the present study. This could be due to the fact the turkey has a less efficient absorption and deposition of vitamin E and/or greater physiological utilization of vitamin E compared to the chicken (Marusich, et al., 1975).

In the present study, the extent of deposition of α-

tocopherol in the dark and light meat was approximately 68% and 32% of the total deposition in the muscle tissue. Previous studies (Lin et al., 1989b; Sheldon, 1984) indicated similar ratios of α-tocopherol distribution in the dark and light muscle. Lin et al. (1989b) reported that dark meat accumulated approximately 50% more α-tocopherol than did the light meat. Sheldon (1984) also found the distribution of α-tocopherol in dark and light muscle to be approximately 72% and 27% of the total deposition in the turkey muscle. The different rate of deposition apparently results from physiological variations in the vascular network of dark and light muscle tissues (Sheldon, 1984). Dark meat from turkeys and broilers has a more highly developed vascular system and a higher lipid content than light meat. Therefore, α-tocopherol is deposited to a greater degree in dark meat than in light meat.

Effect of Vitamin E Supplementation on the Quality of Raw Meat During Refrigerated and Frozen Storage

Lipid stability

Raw dark and light meat from turkeys fed various levels of dietary vitamin E was held at 4°C under fluorescent light for 8 days. The degree of oxidation was monitored every two days by the TBA assay and expressed as TBA-reactive substances (TBARS) (Table 7, Appendix 2 and 3). The data indicated that both dark and light meat from the turkeys fed the diets with the higher (50 and 100 IU/kg feed) vitamin E

Table 7. The effect of vitamin E supplementation on TBARS[1] values of raw dark and light meat during storage at 4°C.

Storage Time (days)	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
	Dark meat		
0	0.26 ± 0.04[a]	0.19 ± 0.01[a]	0.15 ± 0.02[a]
2	0.29 ± 0.03[a]	0.20 ± 0.04[ab]	0.14 ± 0.01[b]
4	0.80 ± 0.12[a]	0.23 ± 0.02[b]	0.22 ± 0.02[b]
6	1.14 ± 0.17[a]	0.42 ± 0.08[b]	0.22 ± 0.02[c]
8	1.38 ± 0.18[a]	0.53 ± 0.07[b]	0.31 ± 0.02[c]
	Light meat		
0	0.21 ± 0.02[a]	0.17 ± 0.01[a]	0.16 ± 0.01[a]
2	0.22 ± 0.01[a]	0.18 ± 0.01[ab]	0.15 ± 0.01[b]
4	0.31 ± 0.03[a]	0.22 ± 0.02[b]	0.18 ± 0.03[b]
6	0.45 ± 0.08[a]	0.24 ± 0.01[b]	0.22 ± 0.03[b]
8	0.49 ± 0.03[a]	0.28 ± 0.04[b]	0.21 ± 0.02[c]

TBARS values in the table are the group means of data from 12 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

[1]TBARS values are expressed as mg malonaldehyde/kg meat

levels had significantly lower ($p < 0.05$) TBARS values during storage than meat samples from the turkeys fed the control diet. These results confirmed those of previous studies for swine (Buckley et al., 1989; Monahan et al., 1990a; Asghar et al., 1991b), veal (Engeseth, 1990), and broilers (Lin et al., 1989b).

The effect of frozen storage on development of TBARS in raw turkey meat is shown in Table 8 and Appendix 4 and 5. Meat from the turkeys fed with the two higher dietary vitamin E levels had lower TBARS values than meat from the turkeys fed the control diet. There was no significant difference ($p < 0.05$) between treatments for light meat after 3 months of frozen storage, but dark meat. After 6 months, the dark and light meat from the two supplemented groups had significantly lower ($p < 0.05$) TBARS values than the meat from the control turkeys. These results demonstrate a positive relationship between tissue α-tocopherol concentration and stability of meat toward lipid oxidation during both refrigerated and frozen storage.

Sklan et al. (1983) reported that dietary vitamin E supplementation (60 IU/kg feed) resulted in increased tissue α-tocopherol concentrations and decreased oxidation of turkey meat tissues during storage. Bartov et al. (1983) also pointed out that tissue α-tocopherol levels significantly influenced the oxidative stability of poultry meat during frozen storage. Sheldon (1984) stated that tissue α-tocopherol levels and TBARS values during

Table 8. The effect of vitamin E supplementation on TBARS values of raw dark and light meat during storage at -20°C[1].

Storage (months)	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
	Dark meat		
0	0.26 ± 0.04[a]	0.19 ± 0.01[a]	0.15 ± 0.02[a]
3	0.65 ± 0.15[a]	0.42 ± 0.07[ab]	0.32 ± 0.01[b]
6	0.99 ± 0.14[a]	0.31 ± 0.09[b]	0.36 ± 0.02[b]
	Light meat		
0	0.21 ± 0.02[a]	0.17 ± 0.01[a]	0.16 ± 0.01[a]
3	0.26 ± 0.03[a]	0.16 ± 0.01[a]	0.16 ± 0.02[a]
6	0.34 ± 0.09[a]	0.17 ± 0.03[b]	0.16 ± 0.02[b]

The values in the table are the group means of data from 12 turkeys ± standard deviation.

Mean values having the same superscript in a row are not significantly different ($p < 0.05$)

[1]TBARS values are expressed as mg malonaldehyde/kg meat)

refrigerated and frozen storage were significantly influenced by dietary vitamin E levels. The present study also supports the findings of Lin et al. (1989b), who reported that vitamin E supplementation at the 100 IU/kg feed level significantly ($p < 0.05$) improved the stability of lipids in both the light and dark meat of broilers during frozen and refrigerated storage. It was also found that dark meat gave consistently higher TBARS values than light meat from all three treatments at the two storage temperatures. These results agreed with those of Sklan et al. (1983), Pikul et al. (1984) and Lin et al. (1989b) who demonstrated that the oxidative changes were much more extensive in dark meat than in light meat, possibly due to the higher phospholipid concentrations.

Similarly, the beneficial effects of dietary vitamin E on the oxidative stability of pork products have been demonstrated. Monahan et al. (1990a) reported that supplementing the diets of pigs fed with 200 IU vitamin E/kg feed significantly ($p < 0.01$) decreased TBARS values of pork during refrigerated storage. Asghar et al. (1991b) demonstrated that TBARS values of raw pork from pigs receiving vitamin E supplements (100 and 200 IU vitamin E/kg feed) were significantly ($p < 0.05$) lower than those for pork from swine fed a control diet during refrigerated storage. Furthermore, pork samples from the 200 IU vitamin E /kg supplemented group were more stable than pork from pigs receiving the feed supplemented with 100 IU vitamin

E/kg.

Drip loss

Fresh raw turkey meat from the three dietary treatments did not exhibit significantly different rates of drip loss ($p < 0.05$) during the first three days of refrigerated storage (Table 9). After 12 days, however, the dark meat from the 50 and 100 IU vitamin E/kg group had significantly ($p < 0.05$) less drip loss than meat from the control groups of turkeys. On the other hand, after 12 days, the light meat from the turkeys receiving 100 IU vitamin E/kg supplements had significantly less drip loss than meat from the control group ($p < 0.05$).

In the frozen storage study, the drip losses from the dark meat were not significantly different between the three groups after 3 and 6 months storage ($p <0.05$) (Table 10). On the other hand, the drip losses from the light meat were significantly ($p < 0.05$) different between the control and supplemented treatments during frozen storage. The results of both refrigerated and frozen storage showed that vitamin E supplementation had a greater effect on the drip loss of light meat than that of dark meat during storage. This might be due to the fact that light meat had greater concentrations of α-tocopherol per unit of fat, thus decreasing the rate of phospholipid changes than dark meat (Sheldon, 1984; Kameda et al., 1985; Erin et al. 1986). These data confirmed the results of Asghar et al. (1991b)

Table 9. Percentage drip loss from meat from turkeys fed vitamin E-supplemented diets when stored under fluorescent light at 4°C

Storage (days)	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
	Dark meat		
3	2.26 ± 0.06[a]	1.87 ± 0.12[a]	1.80 ± 0.25[a]
7	5.38 ± 0.34[a]	4.93 ± 0.11[ab]	4.32 ± 0.27[b]
12	10.84 ± 0.64[a]	8.52 ± 0.24[b]	8.44 ± 0.42[b]
	Light meat		
3	2.31 ± 0.03[a]	1.87 ± 0.41[a]	1.83 ± 0.11[a]
7	5.71 ± 0.22[a]	5.05 ± 0.55[a]	5.31 ± 0.43[a]
12	10.66 ± 0.44[a]	9.92 ± 0.38[ab]	9.14 ± 0.62[b]

Drip loss values are the group means of data from 12 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

Table 10. Percentage drip loss from turkey meat which had been frozen for 3 and 6 months, and subsequently stored under fluorescent light at 4°C

Storage (days)	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
	Dark meat (frozen for 3 months)		
2	4.97 ± 0.30[a]	4.35 ± 0.67[a]	4.28 ± 0.15[a]
4	6.20 ± 0.38[a]	5.74 ± 0.75[a]	5.42 ± 0.22[a]
6	7.43 ± 0.28[a]	6.99 ± 0.77[a]	7.14 ± 0.45[a]
	Dark meat (frozen for 6 months)		
2	5.11 ± 0.68[a]	5.87 ± 0.66[a]	5.81 ± 0.83[a]
4	7.94 ± 0.32[a]	7.92 ± 0.28[a]	7.26 ± 0.32[a]
6	8.18 ± 0.31[a]	8.60 ± 0.83[a]	8.84 ± 0.48[a]
	Light meat (frozen for 3 months)		
2	7.22 ± 0.56[a]	4.81 ± 0.47[b]	3.66 ± 0.24[b]
4	7.60 ± 0.52[a]	5.89 ± 0.21[b]	4.97 ± 0.57[b]
6	8.32 ± 0.42[a]	6.46 ± 0.20[b]	5.71 ± 0.32[b]
	Light meat (frozen for 6 months)		
2	10.78 ± 0.04[a]	6.72 ± 1.32[b]	7.76 ± 0.31[b]
4	12.11 ± 0.23[a]	8.55 ± 0.31[b]	8.67 ± 0.47[b]
6	12.62 ± 0.23[a]	9.74 ± 0.35[b]	9.24 ± 0.54[b]

The values in the table are the group means of data from 6 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different from one another ($p < 0.05$)

who showed that a higher α-tocopherol concentration in pork had a positive effect in minimizing drip losses in meat that had been frozen for 3 months, before thawing and storing at 4°C. Asghar et al. (1991b) also demonstrated that pork from pigs fed 200 IU vitamin E/kg feed exhibited significantly ($p < 0.05$) different rates of drip loss than meat from the control group (10 IU vitamin E/kg feed). Results of the present study revealed that vitamin E was more effective in controlling drip loss in the fresh dark meat samples than in samples that were frozen and then thawed. This might be due to frozen (long term storage) dark meat requiring a higher vitamin E content to prevent drip loss. As the study of Asghar et al. (1991b) indicated, dietary supplementation with 200 IU vitamin E/kg supplied enough vitamin E to show a significant difference in drip loss relative to the control diet (10 IU vitamin E/kg feed), but the 100 IU/kg supplementation level did not. In the present study, the light meat exhibited a greater drip loss than did the dark meat.

Although the exact reasons for the effect of vitamin E on drip loss are not clear, Asghar et al. (1991b) have suggested several possible explanations. First, α-tocopherol may protect the integrity (fluidity) of cell membranes from freeze injury which increases their permeability and results in leakage of sarcoplasmic fluid. Second, α-tocopherol has been shown to protect the membranes from the action of phospholipases (Erin et al., 1986) which

remove the fatty acid moiety from phospholipid molecules. The loss of phospholipids from membranes causes a substantial reduction in its fluidity (Kameda et al., 1985) and hence may increase the drip loss. Third, α-tocopherol may preserve the fluidity of membranes which could otherwise be adversely affected by oxidative changes in the phospholipids.

Color

Changes in color (expressed as L, a, b Hunter values) of turkey meat (dark and light) during storage at 4°C under fluorescent light for 12 days and at -20°C for 6 months are presented in Tables 11, 12, 13 and 14. In the refrigerated samples, L (luminance, whiteness) and b (yellowness) values of dark and light turkey meat of each treatment oscillated or increased very slightly during storage and did not show any apparent change with the length of storage and dietary treatment. This might be due to the difficulty in quantifying the very slight changes in surface color of turkey meat. The redness (a value) of turkey meat apparently decreased with the duration of exposure to fluorescent light. However, changes in redness were slower in the supplemented groups (50 and 100 IU vitamin E/kg feed) compared to those in the control group for both dark and light meat during the first 8 days of storage. There was a significant ($p < 0.05$) difference in the color of dark meat between the control group and the supplemented groups.

Table 11. Changes in the color characteristics of dark turkey meat stored at 4°C for 8 days.

Storage (days)	Hunter parameter	Dietary vitamin E level		
		Control	50 IU/kg	100 IU/kg
0	L-value	39.7 ± 0.6[a]	40.3 ± 0.6[a]	40.9 ± 1.0[a]
2		38.3 ± 0.8[a]	39.2 ± 0.5[a]	40.0 ± 1.3[a]
4		38.0 ± 0.3[a]	38.5 ± 0.4[a]	39.0 ± 0.9[a]
6		38.1 ± 0.6[a]	39.0 ± 0.3[a]	39.1 ± 0.8[a]
8		39.1 ± 0.7[a]	39.3 ± 1.0[a]	38.7 ± 0.7[a]
0	a-value	10.8 ± 0.3[a]	10.9 ± 0.2[a]	10.7 ± 0.5[a]
2		9.4 ± 0.4[b]	10.5 ± 0.2[a]	10.0 ± 0.4[ab]
4		8.8 ± 0.4[a]	9.8 ± 0.5[a]	9.8 ± 0.4[a]
6		8.1 ± 0.6[b]	9.3 ± 0.2[a]	8.8 ± 0.3[ab]
8		6.9 ± 0.7[b]	8.4 ± 0.4[a]	8.6 ± 0.2[a]
0	b-value	8.3 ± 0.4[a]	8.2 ± 0.4[a]	7.4 ± 0.2[a]
2		8.4 ± 0.2[a]	8.1 ± 0.2[a]	7.8 ± 0.3[a]
4		8.2 ± 0.3[a]	8.4 ± 0.3[a]	8.0 ± 0.4[a]
6		8.4 ± 0.3[a]	8.6 ± 0.1[a]	8.2 ± 0.2[a]
8		8.9 ± 0.2[a]	8.6 ± 0.2[a]	8.3 ± 0.1[a]

L: measure lightness and varies from 100 for perfect white to zero for black.
a: measures redness when positive, greyness when zero and greenness when negative.
b: measures yellowness when positive, greyness when zero, and blueness when negative.

The values in the table are the group means of data from 12 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

Table 12. Changes in the color characteristics of light turkey meat stored at 4°C for 8 days.

Storage (days)	Hunter parameter	Dietary vitamin E level		
		Control	50 IU/kg	100 IU/kg
0	L-value	46.6 ± 0.3[a]	46.1 ± 1.4[a]	45.2 ± 0.7[a]
2		48.2 ± 0.4[a]	47.2 ± 1.2[a]	47.6 ± 1.0[a]
4		48.1 ± 0.7[a]	47.5 ± 1.3[a]	47.7 ± 0.8[a]
6		48.0 ± 0.8[a]	47.3 ± 1.7[a]	47.6 ± 0.9[a]
8		48.1 ± 0.5[a]	47.6 ± 1.4[a]	47.9 ± 0.9[a]
0	a-value	9.0 ± 0.5[a]	9.1 ± 0.2[a]	9.1 ± 0.6[a]
2		8.2 ± 0.4[a]	8.9 ± 0.3[a]	8.4 ± 0.4[a]
4		8.0 ± 0.5[a]	8.5 ± 0.4[a]	8.3 ± 0.3[a]
6		7.7 ± 0.4[a]	8.2 ± 0.5[a]	8.0 ± 0.2[a]
8		7.2 ± 0.7[a]	7.5 ± 0.3[a]	7.7 ± 0.3[a]
0	b-value	8.4 ± 0.4[a]	8.3 ± 0.7[a]	8.2 ± 0.6[a]
2		9.6 ± 0.3[a]	9.6 ± 0.3[a]	9.5 ± 0.5[a]
4		9.8 ± 0.3[a]	9.8 ± 0.3[a]	9.6 ± 0.2[a]
6		10.1 ± 0.3[a]	10.2 ± 0.4[a]	10.1 ± 0.4[a]
8		10.6 ± 0.3[a]	10.3 ± 0.4[a]	10.2 ± 0.5[a]

L: measure lightness and varies from 100 for perfect white to zero for black.
a: measures redness when positive, greyness when zero and greenness when negative.
b: measures yellowness when positive, greyness when zero, and blueness when negative.

The values in the table are the group means of data from 12 turkeys ± standard deviation

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

Table 13. Changes in the color characteristics of dark turkey meat stored at -20°C for 3 and 6 months

Storage (months)	Hunter parameter	Dietary vitamin E level		
		Control	50 IU/kg	100 IU/kg
0	L-value	39.7 ± 0.6[a]	40.3 ± 0.6[a]	40.9 ± 1.0[a]
3		37.7 ± 1.3[a]	36.7 ± 0.5[a]	36.5 ± 1.0[a]
6		35.8 ± 0.9[a]	34.9 ± 0.9[a]	35.1 ± 2.0[a]
0	a-value	10.8 ± 0.3[a]	10.9 ± 0.2[a]	10.7 ± 0.5[a]
3		10.2 ± 0.5[a]	10.2 ± 0.4[a]	10.1 ± 0.2[a]
6		8.1 ± 0.4[a]	8.8 ± 0.2[a]	8.9 ± 0.4[a]
0	b-value	8.3 ± 0.4[a]	8.2 ± 0.4[a]	7.4 ± 0.2[a]
3		8.7 ± 0.3[a]	7.5 ± 0.6[a]	7.6 ± 0.5[a]
6		8.0 ± 0.4[a]	7.9 ± 0.3[a]	7.6 ± 0.6[a]

L: measure lightness and varies from 100 for perfect white to zero for black.
a: measures redness when positive, greyness when zero and greenness when negative.
b: measures yellowness when positive, greyness when zero, and blueness when negative.

The values in the table are the group means of data from 12 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

Table 14. Changes in the color characteristics of light turkey meat stored at -20°C for 3 and 6 months.

Storage (months)	Hunter parameter	Dietary vitamin E level		
		Control	50 IU/kg	100 IU/kg
0	L-value	46.6 ± 0.3[a]	46.1 ± 1.4[a]	45.2 ± 0.7[a]
3		46.9 ± 1.5[a]	43.9 ± 1.6[a]	46.4 ± 2.1[a]
6		40.5 ± 1.6[a]	41.2 ± 2.2[a]	40.6 ± 0.2[a]
0	a-value	9.0 ± 0.5[a]	9.1 ± 0.2[a]	9.1 ± 0.5[a]
3		10.3 ± 0.5[b]	12.2 ± 0.3[a]	9.5 ± 0.5[b]
6		7.1 ± 0.2[a]	7.5 ± 0.2[a]	7.8 ± 0.5[a]
0	b-value	8.4 ± 0.4[a]	8.3 ± 0.7[a]	8.2 ± 0.6[a]
3		10.6 ± 0.1[a]	10.3 ± 0.4[a]	10.6 ± 0.3[a]
6		8.1 ± 0.1[a]	8.0 ± 0.2[a]	8.5 ± 0.4[a]

L: measure lightness and varies from 100 for perfect white to zero for black.
a: measures redness when positive, greyness when zero and greenness when negative.
b: measures yellowness when positive, greyness when zero, and blueness when negative.

The values in the table are the group means of data from 12 turkeys ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$)

The L and b values of the turkey meat during frozen storage also did not show apparent changes with length of storage and treatment. The "a" value decreased after 6 months of frozen storage, but was not related to the vitamin E level in the diet of turkeys. This was possibly due to the fact that the level of vitamin E supplementation (50 and 100 IU/kg feed) was not high enough to deposit sufficient α-tocopherol in the meat to protect the pigments during 6 months of frozen storage.

Haurowitz et al. (1941) revealed that hemoglobin was destroyed in the presence of unsaturated fatty acids and oxygen. Lin and Hultin (1977) found that the oxidation of oxymyoglobin in vitro was enhanced by the addition of linoleate hydroperoxide; butylated hydroxyanisole (BHA) and glutathione plus glutathione peroxidase were both effective as antioxidants. Hutchins et al. (1967) also reported a positive correlation ($r=0.73$, $p< 0.005$) between metmyoglobin accumulation and malonaldehyde production in ground beef.

Lipid and pigment oxidation reactions are closely coupled in beef; an increase in one of these processes results in a similar increase for the other (Hutchins et al., 1967; Greene, 1969; Faustman et al., 1989b). The basis for this relationship is not clearly understood.

From the viewpoint of meat color, it is possible that radical species produced during lipid oxidation act directly to promote pigment oxidation, and/or indirectly by damaging pigment reducing systems (Faustman and Cassens, 1990).

Color stability is generally enhanced by the addition of antioxidants (ascorbic acid, butylated hydroxyanisole, propyl gallate and vitamin E) to meat (Greene, 1969; Greene et al., 1971; Govindarajan et al., 1977; Faustman et al., 1989a,b; Asghar et al., 1991b). Faustman et al. (1989a,b) reported that sirloin steaks from vitamin E-supplemented Holstein steers (370 IU/animal/day) had greater ($p < 0.01$) chroma and Hunter "a" values than steaks from control animals. Fresh ground beef from vitamin E-supplemented Holstein steers was more stable to lipid oxidation, and this may be responsible for the associated increased pigment stability. Asghar et al. (1991b) also demonstrated that changes in redness (a value) during the first 6 days of refrigerated storage under light were relatively slow in pork from vitamin E-supplemented pigs (100 and 200 IU vitamin E/ kg feed) compared to the pork from control group (10 IU vitamin E/kg feed).

In addition to inhibiting lipid oxidation and minimizing changes in color, α-tocopherol is also an efficient quencher of singlet oxidation which is normally initiated by fluorescent light (Foote, 1985). Alpha-tocopherol can maintain the reduced form of heme iron for a long time depending on its concentration in the meat system.

Effect of Vitamin E Supplementation on Lipid Stability and Cholesterol Oxidation in Cooked Meat

Lipid stability

Data in Table 15 show the influence of the different dietary levels of vitamin E on the oxidative stability of cooked ground turkey meat stored at 4°C under fluorescent light for 48 hours. TBARS values increased with storage time in all cases. The TBARS values of the cooked ground dark meat from the supplemented groups were slightly lower than the control group, but the differences in the treatments during storage were not significant ($p < 0.05$). The cooked ground light meat, however, did show significantly ($p < 0.05$) different results between the control group and the supplemented groups after 48 hours of refrigerated storage. That the vitamin E supplementation decreased the rate of lipid oxidation in light meat slightly more than in dark meat might be possibly due to the differences in concentrations of tocopherol per unit of fat between dark and light meat (Sheldon, 1984). Monahan et al. (1992b) reported that the formation of lipid oxidation products was significantly higher ($p< 0.01$) in pork from pigs fed a basal level of α-tocopherol acetate (10 mg/kg) compared to the pigs fed the supplemented levels (100 and 200 mg/kg feed). Differences in TBARS between the two supplemented groups were only significant ($p < 0.05$) after 4 days of storage. Vitamin E supplementation in the present study was not as effective in preventing lipid oxidation as

Table 15. The effect of dietary vitamin E supplementation on TBARS values of cooked turkey dark and light meat during storage at 4°C[1].

	Dietary level of vitamin E (IU/kg feed)		
	Control	50	100
Storage time (hr)		Dark meat	
0	3.06 ± 0.20[a]	3.18 ± 0.46[a]	3.14 ± 0.08[a]
24	8.60 ± 0.90[a]	7.37 ± 1.11[a]	7.25 ± 0.94[a]
48	11.83 ± 1.15[a]	11.53 ± 0.81[a]	9.81 ± 1.17[a]
		Light meat	
0	3.22 ± 0.60[a]	2.97 ± 0.48[a]	2.50 ± 0.38[a]
24	5.94 ± 0.64[a]	5.25 ± 0.53[a]	4.80 ± 0.28[a]
48	8.07 ± 0.20[a]	7.40 ± 0.47[a]	6.90 ± 0.43[a]

The values in tables are the group means of data from 12 animals ± standard deviation.

Mean values in a row with the same superscript are not significantly different from one another ($p < 0.05$)

[1]TBARS values are expressed as mg malonaldehyde/kg meat

was reported by Monahan et al. (1992b). This might be due to the fact that turkey lipids are more highly unsaturated than pork lipids and that the turkeys were fed less vitamin E than were the pigs.

The susceptibility of muscle tissue from different species to oxidize is related to the degree of unsaturation of muscle lipids (Allen and Foegeding, 1981; Tichivangana and Morrissey, 1985). The polyunsaturated fatty acid content of muscle varies between species and decreases in the order: fish > poultry > pork > beef > lamb (Pearson et al., 1977). Higher vitamin E supplementation might be necessary to supply a more beneficial antioxidative effect in those meats containing a higher proportion of unsaturated lipids (Rhee, 1988; Frigg et al., 1990; Monahan et al., 1992c). The present study also showed that the TBARS values of cooked dark meat were higher than the cooked light meat values in all cases during refrigerated storage. Jacobson and Koehler (1970) reported that cooked dark meat had higher TBARS values than cooked light meat because the former contains more heme pigment which can catalyze or enhance the lipid oxidation process. They also demonstrated a marked increase in TBARS value in the two types of meat during refrigeration.

Cholesterol oxidation

Four major COPS were consistently present in detectable quantities in all turkey samples: cholesterol α-epoxide,

cholesterol ß-epoxide, 7ß-hydroxycholesterol and 7-ketocholesterol (Table 16). The other minor cholesterol oxidation products were 7α-diol (7α-OH), 25-hydroxycholesterol and cholestane-triol. COPS in the cooked turkey samples increased during refrigerated storage (Table 16). The principal oxidation product was 7-ketocholesterol and constituted over 50% of the total COPS, as was reported earlier by Zubillaga and Maerker (1991). The secondary oxysterols did not increase over the 48 hour period in the manner reported by Pie et al. (1991). These investigators demonstrated that there was a greater increase in primary oxysterols (i.e, those formed by oxidation at carbon 7 or the side chain) compared to the secondary derivatives (cholesterol epoxides and cholestanetriol) during the cooking of fresh meat. The concentration of oxysterols doubled in the commercially available precooked frozen meals after 3 months at -20°C with the secondary oxysterols showing the highest increase (Pie et al., 1991).

Data in Table 17 and Appendix 6 and 7 show that the concentration of COPS, expressed as a percentage of total cholesterol in cooked turkey meat, increased during refrigerated storage, thus confirming the observation of Pie et al. (1991) that the COPS in cooked meat increase with storage time. In this study, total COPS were calculated as the sum of the concentrations of the four major COPS and the three minor COPS mentioned previously. Pie et al. (1991) demonstrated that all oxidized derivatives increased

Table 16. Effect of dietary α-tocopherol supplementation on the percentage of cholesterol oxide to total cholesterol in cooked ground dark and light turkey meat during refrigerated storage.

Dark meat

COPS	0 hour			24 hour			48 hour		
	G1	G2	G3	G1	G2	G3	G1	G2	G3
ß-epoxide	0.041	0.049	0.016	0.162	0.180	0.044	0.189	0.169	0.121
α-epoxide	0.084	0.060	0.017	0.199	0.191	0.141	0.261	0.317	0.166
7ß-OH	0.130	0.056	0.018	0.489	0.487	0.154	0.561	0.503	0.258
7-keto	0.256	0.210	0.099	1.318	0.983	0.661	1.921	1.650	1.044

Light meat

COPS	0 hour			24 hour			48 hour		
	G1	G2	G3	G1	G2	G3	G1	G2	G3
ß-epoxide	0.027	0.039	0.030	0.265	0.162	0.084	0.336	0.310	0.157
α-epoxide	0.064	0.094	ND	0.055	0.156	0.111	ND	ND	ND
7ß-OH	0.159	0.127	0.076	0.448	0.277	0.216	0.710	0.575	0.394
7-keto	0.159	0.174	0.028	1.075	0.667	0.482	1.277	1.102	0.772

The value in the table are the group means (6 animals)

Table 17. Effect of vitamin E supplementation on the development of total cholesterol oxidation products in cooked dark and light turkey meat during storage at 4°C.

	Total COP concentration (expressed as a percentage of initial cholesterol)		
Dietary Vitamin E Level (IU/kg feed)	12.5 (control)	50	100
Storage Time (hr)		Dark meat	
0	0.73 ± 0.16[a]	0.59 ± 0.06[ab]	0.23 ± 0.04[b]
24	2.38 ± 0.26[a]	1.79 ± 0.13[b]	1.00 ± 0.20[c]
48	3.12 ± 0.24[a]	2.44 ± 0.21[b]	1.53 ± 0.23[c]
		Light meat	
0	0.55 ± 0.03[a]	0.35 ± 0.04[a]	0.21 ± 0.03[a]
24	1.88 ± 0.32[a]	1.19 ± 0.07[b]	0.91 ± 0.17[b]
48	2.49 ± 0.18[a]	2.22 ± 0.18[a]	1.17 ± 0.15[b]

The values in the table are group means of data from 6 turkeys per treatment ± standard deviation.

Mean values in a row with the same superscript are not significantly different ($p < 0.05$).

progressively in meat samples stored for 3 months at -20°C in both raw and cooked meat. Table 17 also shows that the concentration of COPS in dark meat were slightly greater than those in light meat. Park and Addis (1987) reported that total COPS formed in dark turkey meat after 3 days of refrigerated storage at 4°C represented 1.20% of the total cholesterol. A slightly higher concentration of COPS (3.12%) in the dark meat after 2 days at 4°C were observed in the present study and can be explained by the fact that the meat had been stored at -20°C for 3 months before cooking.

The formation of COPS in cooked turkey meat during storage was significantly ($p < 0.05$) influenced by dietary vitamin E. Cholesterol oxide concentrations in the cooked ground dark turkey meat from the three groups were significantly ($p < 0.05$) different after 24 hours. Cholesterol oxide formation in cooked ground light meat from the control group was significantly ($P < 0.05$) different from that in meat from the higher vitamin E (100 IU/kg feed) supplemented group over the same storage period. There was no significant difference between the data (2.22% and 1.17%) of the two vitamin E (50 and 100 IU/ kg feed) supplemented groups or the data (2.49% and 2.22%) of the control group and lower vitamin E (50 IU/kg feed) supplemented group.

The results of this study agree with those of previous studies (Engeseth, 1990; Monahan et al., 1992b) in that COP formation in cooked meat is significantly influenced by

dietary vitamin E. Engeseth (1990) reported that the COP concentration in cooked veal from calves fed a basal diet and one containing 500 IU vitamin E/day were 1.53 and 0.53% of the total cholesterol after 4 days of refrigerated storage, respectively. Monahan et al. (1992b) also demonstrated that the total COPS present in cooked pork from pigs fed a basal level of vitamin E (10 IU/kg feed) represented 2.7% of the total cholesterol after 2 days of refrigerated (4°C) storage. In the case of pork from pigs fed diets containing 200 IU vitamin E, total COPS accounted for 1.6% of the total cholesterol.

Relationship between lipid oxidation and cholesterol oxidation

The relationship between lipid oxidation, as estimated by the TBA method, and COP formation was examined (Figures 6 and 7). The data indicate correlations of r= 0.79 and r= 0.55 between TBARS values and total COPS (expressed as a percentage of the total cholesterol) in cooked dark and light turkey meat stored for 2 days at 4°C, respectively. Park and Addis (1987) noted greater cholesterol oxide development in beef and turkey with increasing rancidity (as determined by TBARS values). Engeseth (1990) also showed that cholesterol oxidation followed similar trends as TBARS development. De Vore (1988) reported that TBARS values and 7-ketocholesterol were correlated for raw (r=0.91) and cooked (r=0.99) beef. Monahan et al. (1992b) also

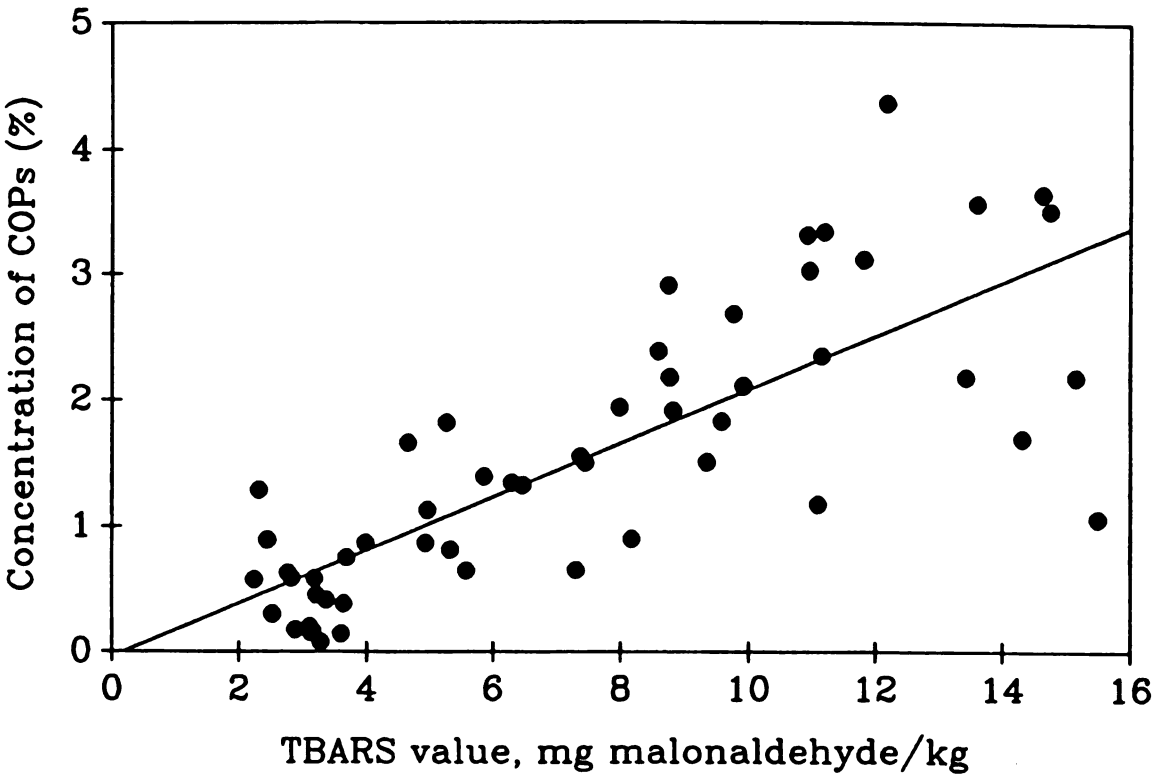


Figure 6. Relationship between TBARS and total COP formation in cooked ground dark turkey meat stored for up to 3 days at 4°C.

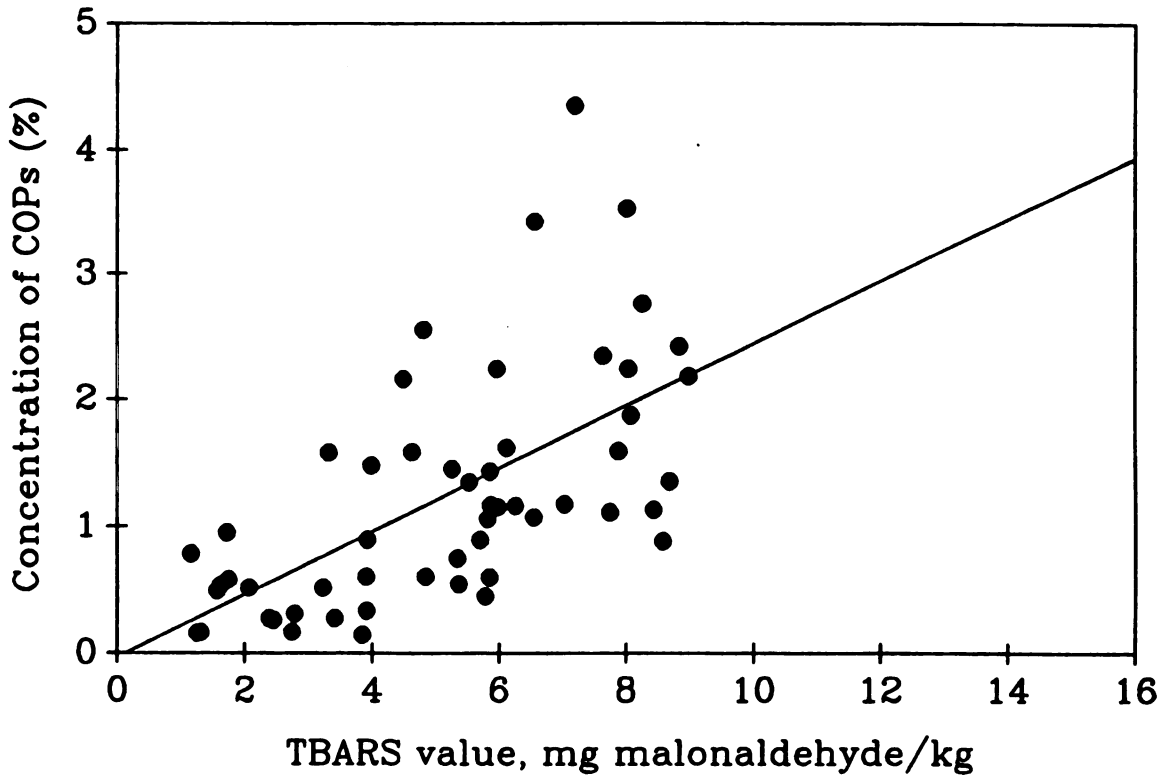


Figure 7. Relationship between TBARS and total COP formation in cooked ground light turkey meat stored for up to 3 days at 4°C.

demonstrated a strong correlation (r= 0.88) between TBARS and total COP concentration in cooked pork stored at 4°C for 4 days and the correlation coefficients for β-epoxide, 7β-OH and 7-keto with TBARS were +0.75, +0.78 and +0.84, respectively.

These studies demonstrated that the rate of cholesterol oxidation in meat is greatly accelerated by cooking and appears to follow the same trend as lipid oxidation in general. Polyunsaturated lipids are susceptible to oxidation and to the formation of highly reactive free radicals. Igene et al. (1980) established that membrane phospholipids are the major source of polyunsaturated fatty acids (PUFA). The structural membranes in lean beef muscle contain the major share of the cholesterol (Rhee et al., 1982b). PUFA and cholesterol are integral components of the membrane structure, free radicals from phospholipid oxidation may initiate cholesterol oxidation in the tissue membranes of ground beef. Dietary α-tocopherol supplementation has been shown to significantly increase the α-tocopherol content of muscle mitochondrial and microsomal cell membranes in pigs (Monahan et al., 1990b) and chickens (Asghar et al., 1989b). Thus, the localization of α-tocopherol at the proposed site of initiation of lipid oxidation provides a means of retarding lipid and cholesterol oxidation in meats through dietary supplementation with vitamin E.

Effect of Vitamin E Supplementation on Microsomal Lipid Stability

Concentration of α-tocopherol in the microsomal fractions of turkey meat

Alpha-tocopherol concentrations in the microsomal fraction isolated from the light and dark meat from turkeys receiving the vitamin E-supplemented diets were approximately 3-5 fold greater than those in comparable fractions from meat of the control animals (Table 18). The vitamin E concentration in the microsomes increased with increasing dietary vitamin E level.

Several researchers have investigated the deposition of α-tocopherol in the membranes as a consequence of feeding vitamin E-supplemented diets. Asghar et al. (1990) reported that the microsomes from the dark meat of control broilers and those receiving a vitamin E diet (100 IU vitamin E/kg), contained 3.3 and 126.0 μg α-tocopherol/g membrane, respectively. The microsomes from the light meat of the control group and the vitamin E group contained 0.9 μg/g membrane and 2.2 μg/g membrane, respectively. Microsomal α-tocopherol concentrations in the dark meat of turkeys were much lower than those reported in chicken microsomes by Asghar et al. (1990), however, light meat concentrations were similar.

The results of studies with pigs (Monahan et al., 1990b; Asghar et al., 1991b) and calves (Engeseth, 1990) also agreed with those of the present study in that vitamin E supplementation increased the α-tocopherol concentration

Table 18. Concentration of α-tocopherol in the microsomes of dark and light meat from turkeys fed control, and vitamin E-supplemented diets

Treatment	Alpha-tocopherol (μg/g membrane, wet basis)	
	Microsome	
Vitamin E dietary level (IU/kg feed)	Dark	Light
12.5	1.53 ± 0.84	0.68 ± 0.14
50	6.29 ± 0.54	2.32 ± 1.20
100	7.70 ± 2.72	2.51 ± 0.10

The values in the table are the means of data from the duplicate pooled sample (12 turkeys from each treatment) ± standard deviation.

of microsomes. Monahan et al. (1990b) reported that the microsomal α-tocopherol concentrations in the meat of pigs which were fed a basal diet (<40 IU vitamin E/kg feed) and 160 IU/kg feed were 62.4 and 164.8 μg/g protein. Asghar et al. (1991b) demonstrated that the deposition of α-tocopherol in the microsomes (12, 29.4, 53.1 μg/g protein) increased linearly with the level (10, 100, and 200 IU/kg feed) of vitamin E in the diet. Engeseth (1990) reported microsomal α-tocopherol concentrations in control and vitamin E-supplemented (500 IU/day) veal calves of 139 and 707 μg/g protein, respectively.

Metmyoglobin/hydrogen peroxide-catalyzed lipid peroxidation

Microsomal lipid oxidation in turkey dark and light meat is graphically presented in Figures 8 and 9, respectively. Generally, oxidative changes were more extensive in the microsomal fractions isolated from dark meat than those in the comparable fraction from light meat. These results confirm the findings of Harel and Kanner (1985) and Asghar et al. (1989b) who reported a higher rate of lipid peroxidation in microsomes from the dark muscle tissues of turkeys and broilers, respectively, compared to the light muscle microsomes. The reason for these differences was not explained by these investigators. However, greater total lipid contents may contribute to the larger TBARS values for the membranes from the dark meat (Lin, 1988).

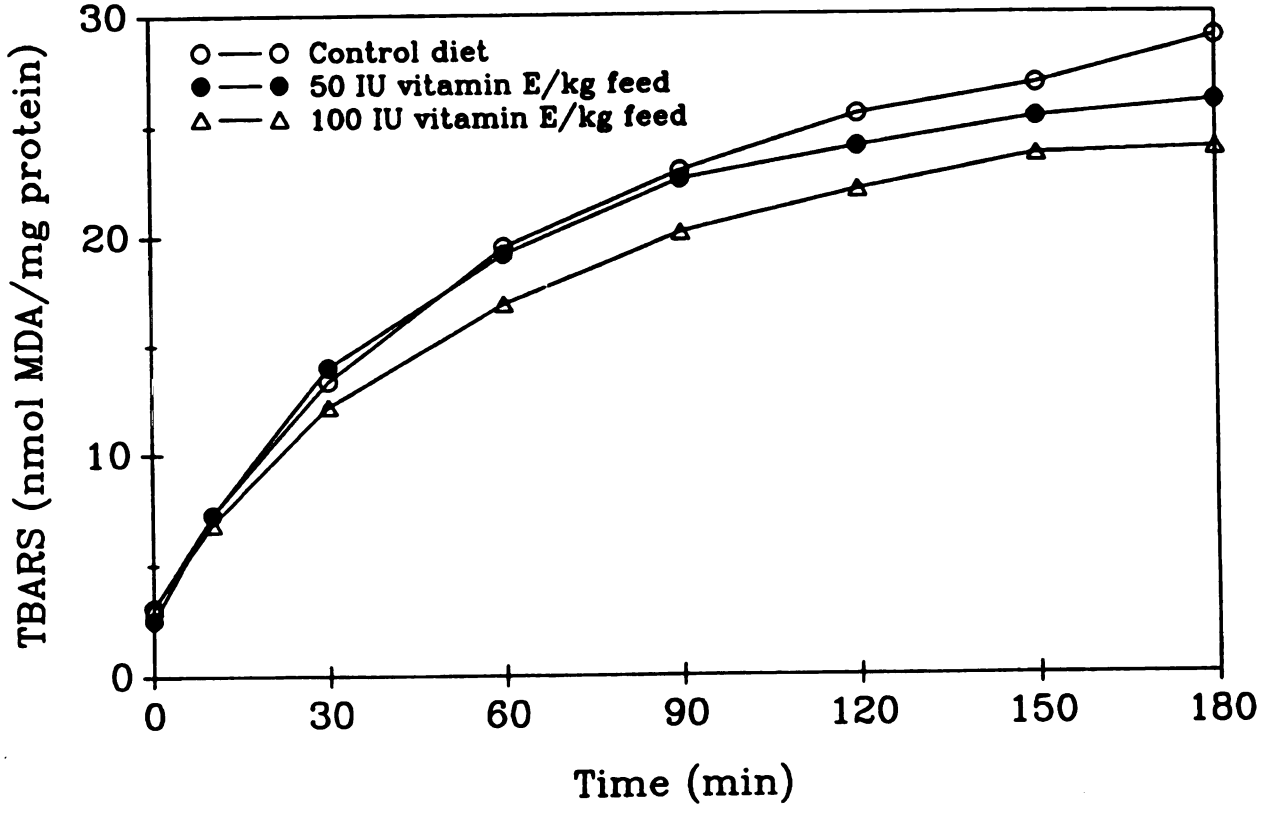


Figure 8. Metmyoglobin/hydrogen peroxide-catalyzed lipid peroxidation in the microsomal membranes from dark meat of turkeys fed a control and vitamin E-supplemented (50 and 100 IU/kg feed) diets

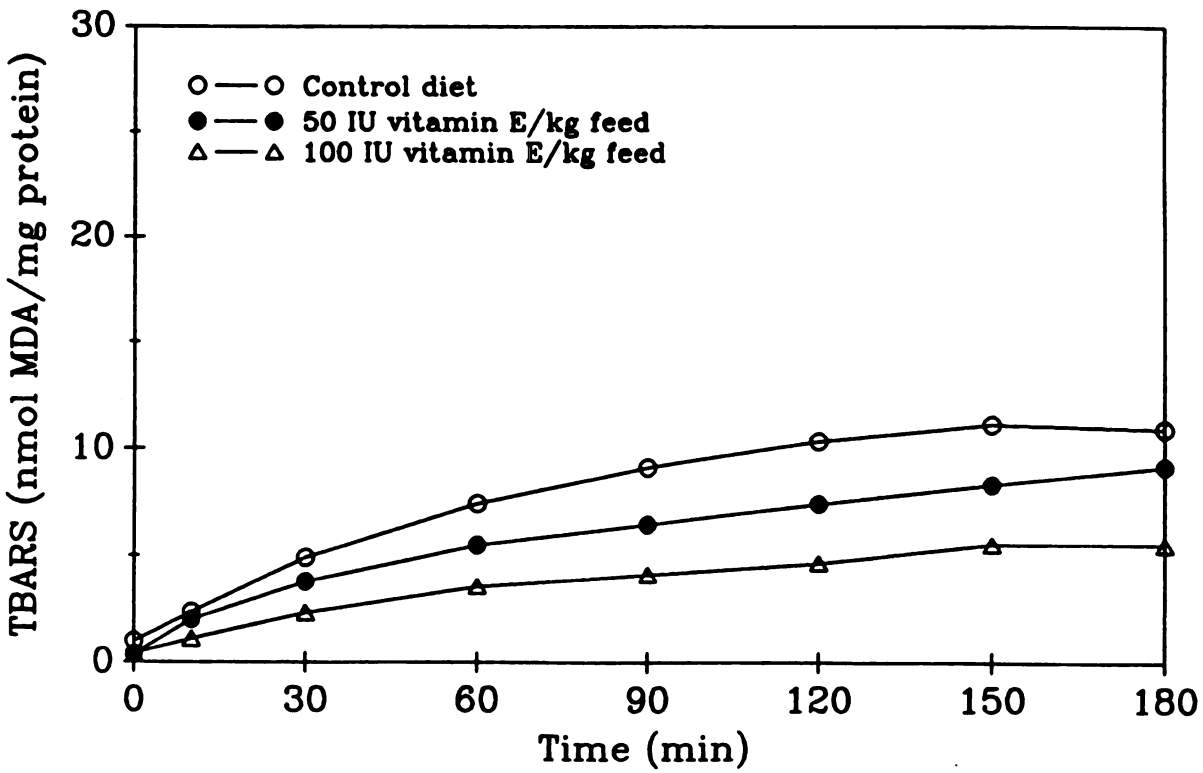


Figure 9. Metmyoglobin/hydrogen peroxide-catalyzed lipid peroxidation in the microsomal membranes from light meat of turkeys fed control vitamin E-supplemented (50 and 100 IU/kg feed) diets

Membranal oxidation in the supplemented groups (50 IU and 100 IU vitamin E/kg feed) were slower than the control group. Asghar et al. (1989b) demonstrated that membrane lipids from broilers receiving 200 IU vitamin E/kg showed significantly ($p < 0.01$) different rates of peroxidation to membrane lipids from control group. In the case of broiler microsomal lipids showed more significant difference between the supplemented group and control group than those of turkey membranal lipids did. The difference might be due to the fact that the broilers were fed higher levels of vitamin E than the turkeys in the present study. It is quite likely that turkeys need higher levels of dietary vitamin E than broilers to increase the α-tocopherol concentration of the microsomes of turkey meat in order to decrease the rate of oxidation (Harel and Kanner, 1985). In our study, the microsomes of the dark and light meat from the control group were the least stable, while the higher level of vitamin E supplementation (100 IU/kg feed) produced the more stable microsomes when reacted with the metmyoglobin/hydrogen peroxide catalytic system. These results confirmed that α-tocopherol can inhibit *in vitro* microsomal lipid peroxidation catalyzed by hydrogen peroxide-activated metmyoglobin as reported by Harel and Kanner (1985), Asghar et al. (1989b; 1991b), Buckley et al. (1989) and Monahan et al. (1990a). The latter group demonstrated that vitamin E stabilized the membrane-bound lipids in pork against metmyoglobin/H_2O_2-catalyzed oxidation and also significantly

($p<0.05$) improved the oxidative stability of rendered fat. Asghar et al. (1991b) reported that the differences in the concentration of α-tocopherol in the mitochondrial and microsomal fractions from *L. dorsi* muscle of the pig fed with 10, 100 and 200 IU vitamin E/kg feed were clearly reflected in the enhanced stability of the membrane-bound lipids.

SUMMARY AND CONCLUSION

The influence of three levels of dietary vitamin E (12.5, 50 and 100 IU/kg feed) on the deposition of α-tocopherol in the muscle, on selected quality characteristics of raw turkey meat (oxidative stability of lipids, color, drip loss) during refrigerated and frozen storage, and on the oxidative stability of lipid and cholesterol in cooked meat during refrigerated storage was studied. The effect of vitamin E supplementation on the oxidative stability of turkey microsomal lipids was also investigated.

Results indicated that the level of dietary vitamin E influenced the concentration of α-tocopherol in the muscle membranes and the resultant oxidative stability of turkey lipids. Dietary supplementation at the higher levels (50 IU and 100 IU/kg feed) resulted in 3-5 fold increases in muscle and microsomal α-tocopherol concentrations and enhanced the oxidative stability of muscle and microsomal lipids. Concentrations of α-tocopherol in the dark and light meat from turkeys fed 50 IU and 100 IU vitamin E/kg feed were 3.35 μg and 1.59 μg/g, and 4.59 μg and 2.13 μg/g, respectively. These values were significantly higher (p

<0.05) than those for the control group. Growth performance was not significantly ($p < 0.05$) improved by dietary supplementation with vitamin E.

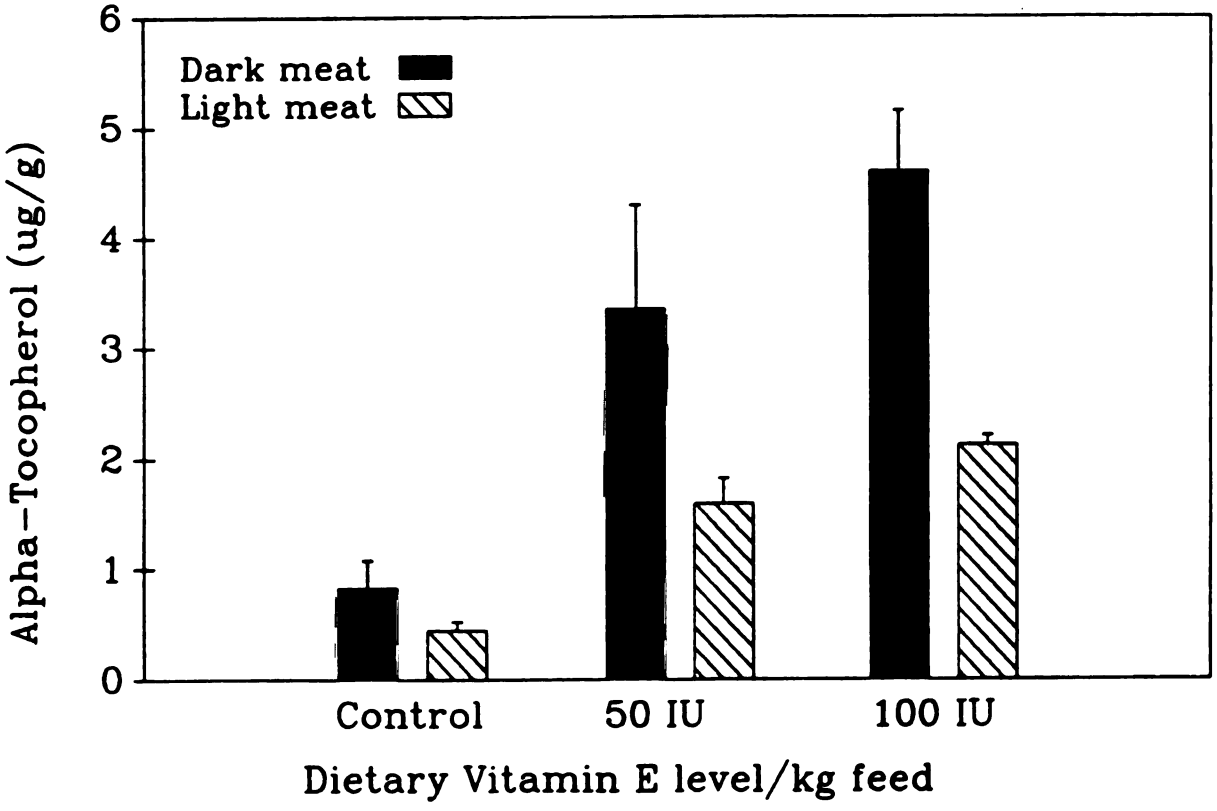
Dietary vitamin E significantly ($p < 0.05$) influenced the oxidative stability of raw meat during refrigerated and frozen storage. The increased oxidative stability of turkey meat through dietary vitamin E supplementation also improved color stability and decreased drip loss during refrigerated storage. During frozen storage, however, vitamin E supplementation did not produce any significant differences in the color of turkey meat. TBARS values of cooked dark meat were not significantly influenced by dietary treatment. However, vitamin E supplementation had a positive effect on the stability of cooked light meat.

The microsomes of the dark and light meat from the control group were the least stable while vitamin E supplementation (100 IU/kg feed) produced the more stable microsomes when reacted with the metmyoglobin/hydrogen peroxide catalyst. The oxidative changes were much more extensive in the microsomes from dark meat compared to those from light meat.

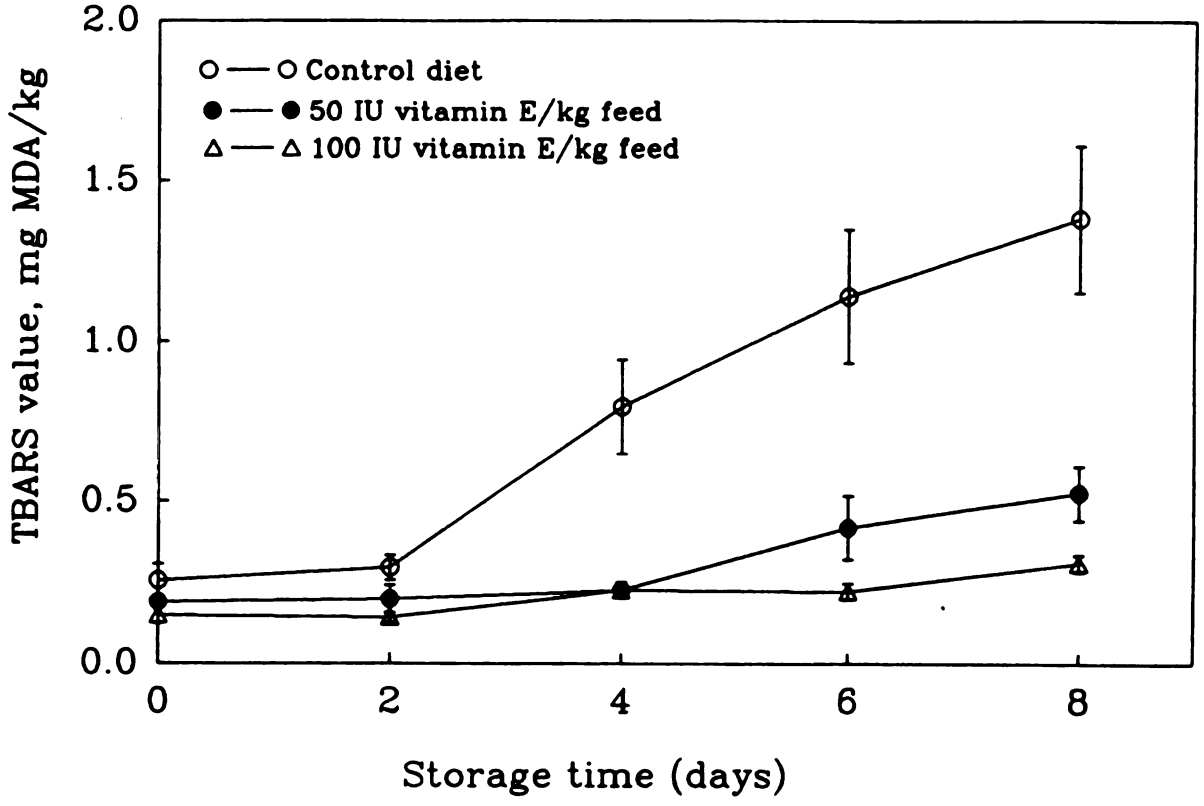
Cholesterol oxidation occurred in cooked turkey meat stored for 3 days at 4°C. The formation of COPS in cooked turkey meat during storage was significantly ($p < 0.05$) influenced by dietary vitamin E. The relationship between lipid oxidation indicated correlations of $r = +0.79$ and $r = +0.55$ between TBARS values and cholesterol oxide

concentrations in cooked dark and light turkey meat, respectively.

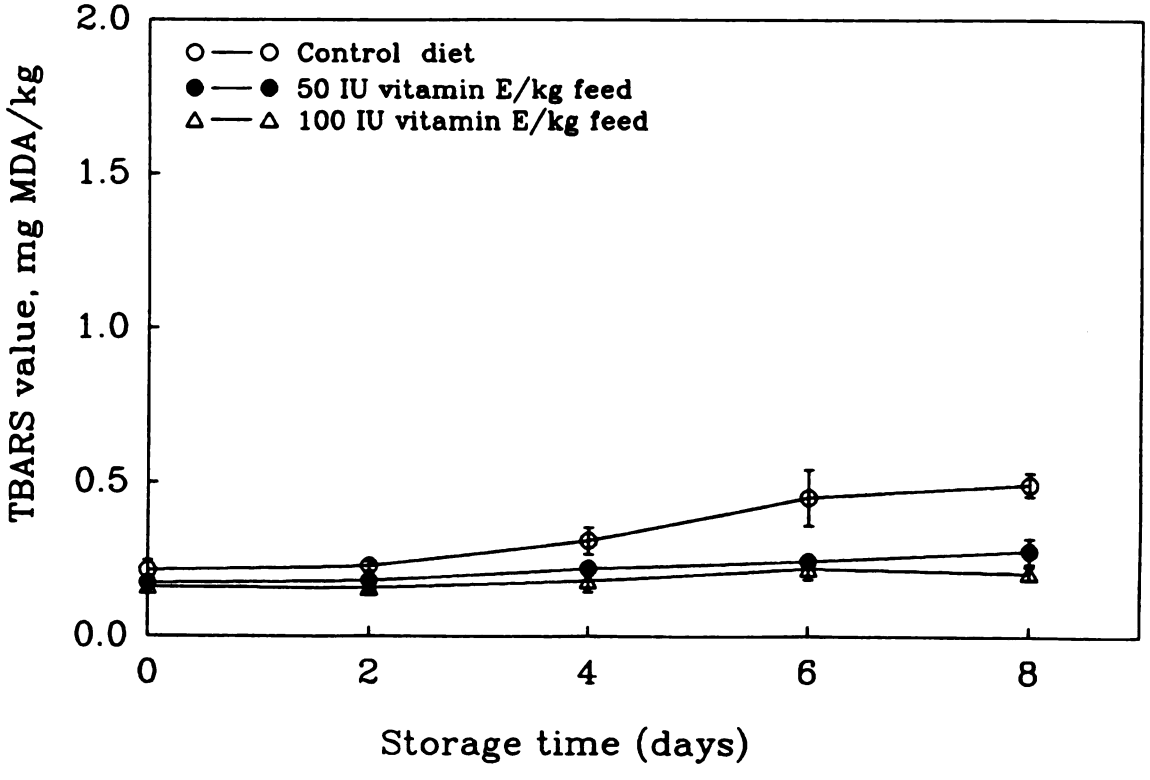
APPENDICES



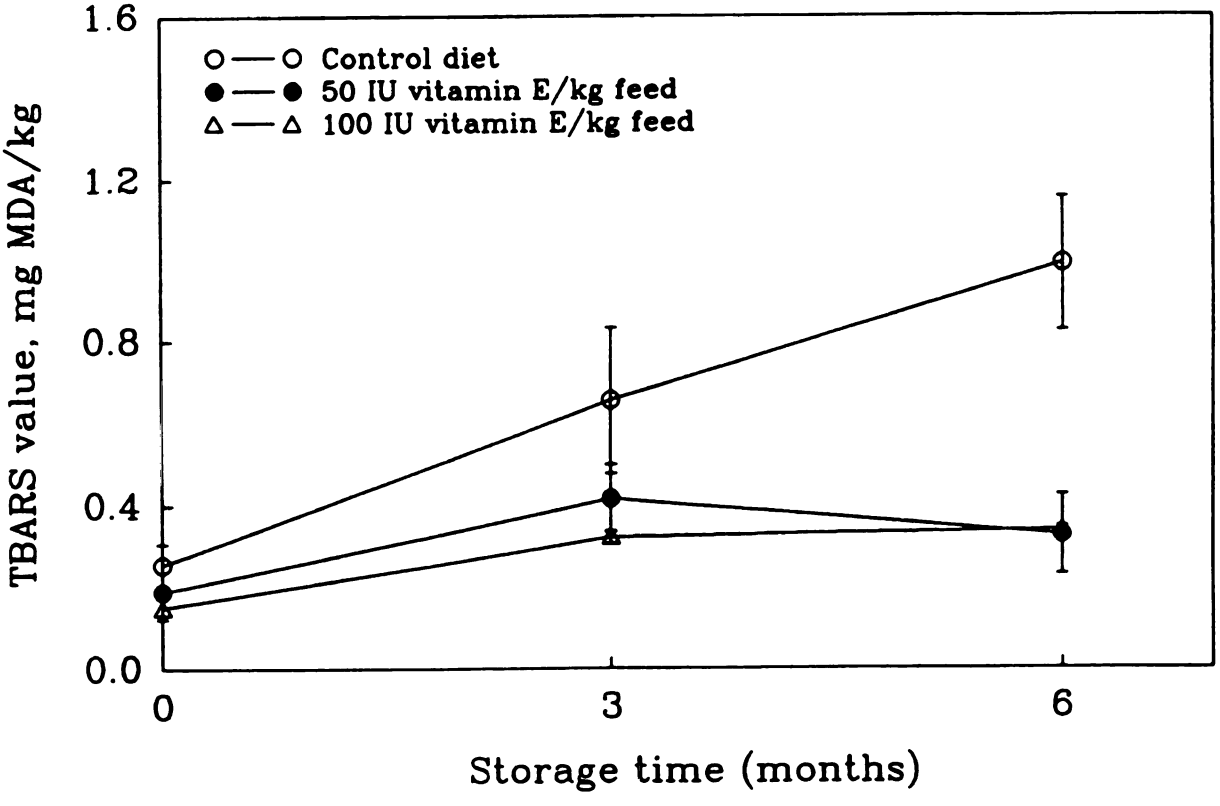
Appendix 1. Concentrations of α-tocopherol (μg/g meat) in dark and light meat from turkeys fed diets containing three levels of vitamin E.



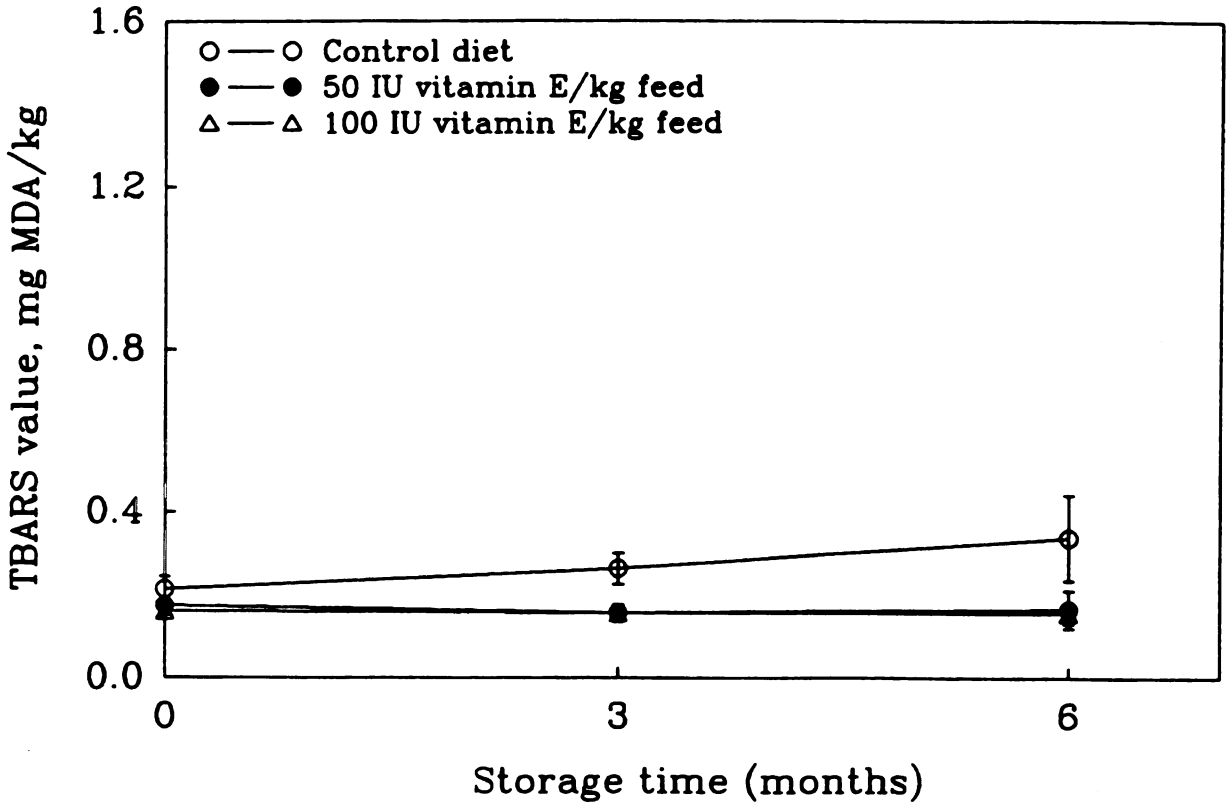
Appendix 2. The effect of vitamin E supplementation on TBARS values (mg malonaldehyde/kg sample) of raw dark turkey meat stored at 4°C for 8 days.



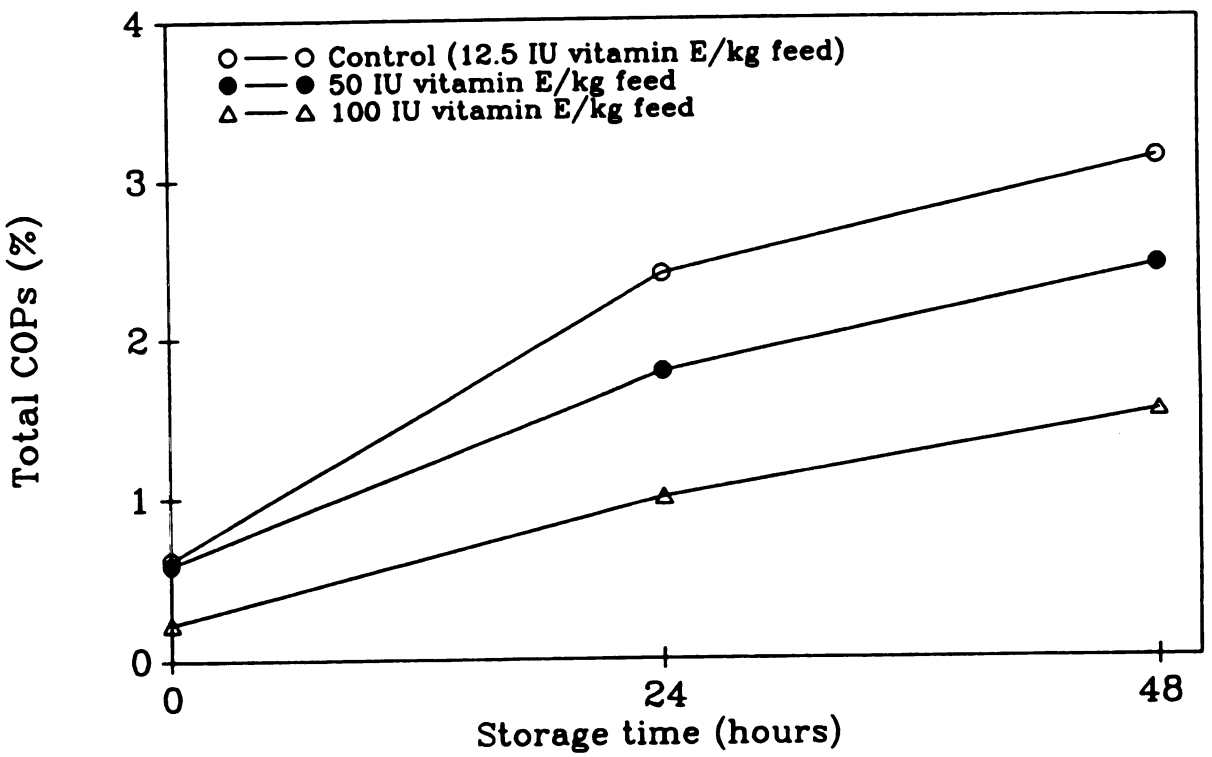
Appendix 3. The effect of vitamin E supplementation on TBARS values (mg malonaldehyde/kg sample) of raw light turkey meat stored at 4°C for 8 days.



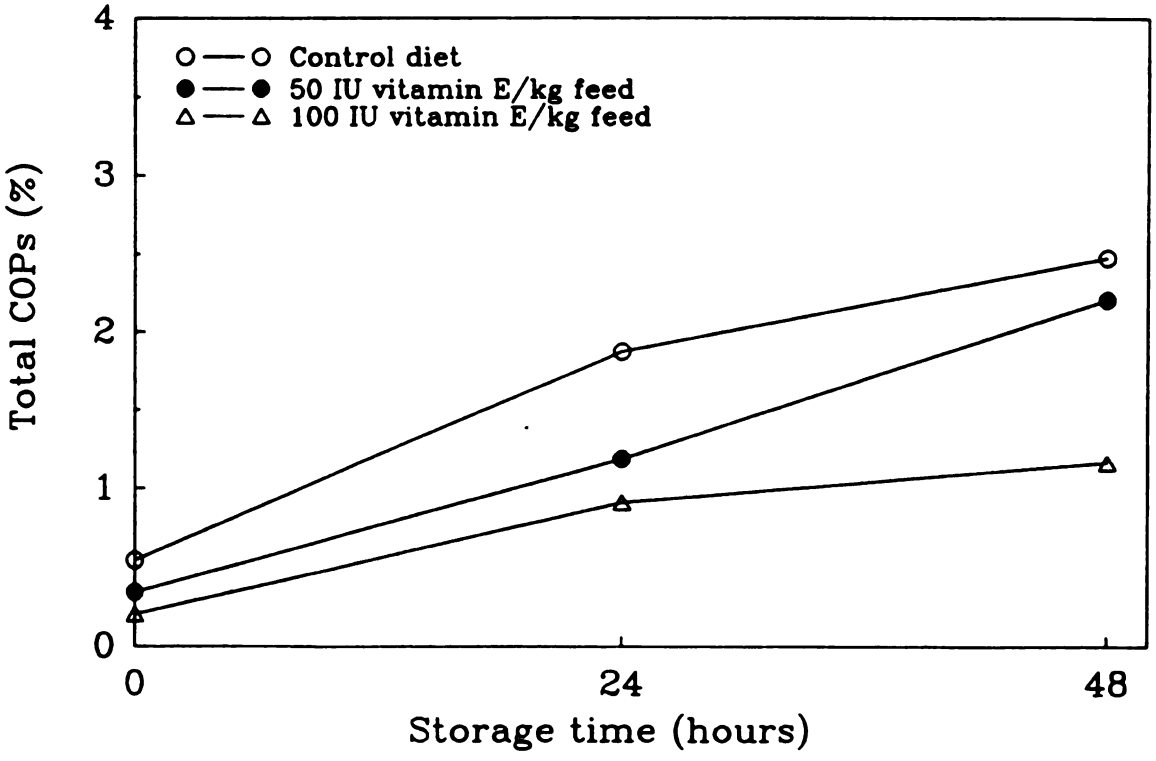
Appendix 4. Effect of different levels of dietary vitamin E on TBARS values (mg malonaldehyde/kg sample) of raw dark meat stored at -20°C for 6 months



Appendix 5. Effect of different levels of dietary vitamin E on TBARS values (mg malonaldehyde/kg sample) of raw light meat stored at -20°C for 6 months



Appendix 6. Effect of vitamin E supplementation on the development of cholesterol oxidation products (expressed as a percentage of the initial cholesterol concentration) in cooked ground dark turkey meat during storage at 4°C.



Appendix 7. Effect of vitamin E supplementation on the development of cholesterol oxidation products (expressed as a percentage of the initial cholesterol concentration) in cooked ground light turkey meat during storage at 4°C.

Appendix 8. Analysis of variance for TBARS values of raw dark turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	2.05	0.01	174.32	< 0.05
E(1) =pens/T	6	0.04	1.02		
P = periods	4	1.71	0.43	168.06	< 0.05
TP = interaction	8	0.96	0.12	47.30	< 0.05
E(2) = residual error	24	0.06	0.00		

Appendix 9. Analysis of variance for TBARS values of raw light turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.21	0.10	78.91	< 0.05
E(1) =pens/T	6	0.01	0.00		
P = periods	4	0.16	0.04	45.53	< 0.05
TP = interaction	8	0.08	0.01	11.27	< 0.05
E(2) = residual error	24	0.02	0.00		

Appendix 10. Analysis of variance for "a" values of color of raw dark turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	8.14	4.07	5.04	< 0.05
E(1) =pens/T	6	4.85	0.81		
P = periods	4	44.14	11.03	111.92	< 0.05
TP = interaction	8	3.11	0.39	3.94	< 0.05
E(2) = residual error	24	2.37	0.10		

Appendix 11.Analysis of variance for " a" values of color of raw light turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	1.46	0.73	0.71	> 0.05
E(1) =pens/T	6	6.12	1.02		
P = periods	4	12.47	3.12	44.32	< 0.05
TP = interaction	8	0.46	0.06	0.82	> 0.05
E(2) = residual error	24	1.69	0.07		

Appendix 12.Analysis of variance for "L" values of color of raw dark turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	6.44	3.22	1.04	> 0.05
E(1) =pens/T	6	18.66	3.11		
P = periods	4	17.42	4.36	17.76	< 0.05
TP = interaction	8	4.02	0.50	2.05	> 0.05
E(2) = residual error	24	5.89	0.25		

Appendix 13.Analysis of variance for "L" values of color of raw light turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	4.02	2.01	0.28	> 0.05
E(1) =pens/T	6	43.07	7.18		
P = periods	4	22.60	5.65	53.09	< 0.05
TP = interaction	8	2.10	0.26	2.46	< 0.05
E(2) = residual error	24	2.55	0.11		

Appendix 14. Analysis of variance for "b" values of color of raw dark turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	2.16	1.08	4.17	> 0.05
E(1) =pens/T	6	1.55	0.26		
P = periods	4	2.39	0.60	8.76	< 0.05
TP = interaction	8	0.70	0.09	1.29	> 0.05
E(2) = residual error	24	1.64	0.07		

Appendix 15. Analysis of variance for "b" values of color of raw light turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.23	0.12	0.11	> 0.05
E(1) =pens/T	6	6.48	1.08		
P = periods	4	23.41	5.85	159.76	< 0.05
TP = interaction	8	0.22	0.03	0.76	> 0.05
E(2) = residual error	24	0.88	0.04		

Appendix 16. Analysis of variance for total cholesterol oxidation product concentrations in cooked dark turkey meat (cooked study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	6.10	3.05	53.52	< 0.05
E(1) =pens/T	6	0.34	0.06		
P = periods	2	15.87	7.94	162.33	< 0.05
TP = interaction	4	0.99	0.25	5.07	< 0.05
E(2) = residual error	12	0.59	0.05		

Appendix 17. Analysis of variance for total cholesterol oxidation product concentrations in cooked light turkey meat (cooked study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	4.30	2.15	30.67	< 0.05
E(1) =pens/T	6	0.42	0.07		
P = periods	2	11.70	5.85	62.22	< 0.05
TP = interaction	4	1.27	0.32	3.37	< 0.05
E(2) = residual error	12	1.13	0.09		

Appendix 18. Analysis of variance for TBARS values of cooked dark turkey meat (cooked study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	5.43	2.71	1.71	> 0.05
E(1) =pens/T	6	9.52	1.59		
P = periods	2	285.65	142.83	170.32	< 0.05
TP = interaction	4	5.03	1.26	1.50	> 0.05
E(2) = residual error	12	10.06	0.84		

Appendix 19. Analysis of variance for TBARS values of cooked light turkey meat (cooked study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	4.62	2.31	4.35	> 0.05
E(1) =pens/T	6	0.42	0.07		
P = periods	2	93.89	46.94	210.89	< 0.05
TP = interaction	4	0.25	0.06	0.29	> 0.05
E(2) = residual error	12	2.67	0.22		

Appendix 20. Analysis of variance for drip loss of raw dark turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	8.59	4.30	17.30	< 0.05
E(1) =pens/T	6	1.49	0.25		
P = periods	2	242.57	121.28	1131.06	< 0.05
TP = interaction	4	4.59	1.15	10.69	< 0.05
E(2) = residual error	12	1.29	0.11		

Appendix 21. Analysis of variance for drip loss of raw light turkey meat (refrigerated study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	3.17	1.58	2.80	< 0.05
E(1) =pens/T	6	3.40	0.57		
P = periods	2	283.30	141.65	1952.08	< 0.05
TP = interaction	4	1.42	0.35	4.88	< 0.05
E(2) = residual error	12	0.87	0.07		

Appendix 22. Analysis of variance for drip loss of raw dark turkey meat (3 months frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	1.81	0.91	0.96	> 0.05
E(1) =pens/T	6	5.65	0.94		
P = periods	2	31.66	15.83	210.07	< 0.05
TP = interaction	4	0.26	0.06	0.85	> 0.05
E(2) = residual error	12	0.90	0.08		

Appendix 23. Analysis of variance for drip loss of raw light turkey meat (3 months frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	40.25	20.12	32.41	< 0.05
E(1) =pens/T	6	3.73	0.62		
P = periods	2	11.62	5.81	74.88	< 0.05
TP = interaction	4	0.95	0.24	3.06	> 0.05
E(2) = residual error	12	0.93	0.08		

Appendix 24. Analysis of variance for drip loss of raw dark turkey meat (6 months frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	3.84	1.92	4.71	> 0.05
E(1) =pens/T	6	2.45	0.41		
P = periods	2	45.82	22.91	86.35	< 0.05
TP = interaction	4	4.14	1.04	3.90	< 0.05
E(2) = residual error	12	3.18	0.27		

Appendix 25. Analysis of variance for drip loss of raw light turkey meat (6 months frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	69.15	34.57	49.57	< 0.05
E(1) =pens/T	6	4.19	0.70		
P = periods	2	20.68	10.34	32.14	< 0.05
TP = interaction	4	2.00	0.50	1.56	> 0.05
E(2) = residual error	12	3.86	0.32		

Appendix 26. Analysis of variance for TBARS values of raw dark turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.89	0.44	24.30	< 0.05
E(1) =pens/T	6	0.11	0.02		
P = periods	1	0.03	0.03	4.47	< 0.05
TP = interaction	2	0.15	0.07	10.37	< 0.05
E(2) = residual error	6	0.04	0.01		

Appendix 27. Analysis of variance for TBARS values of raw light turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.08	0.04	12.64	< 0.05
E(1) =pens/T	6	0.02	0.00		
P = periods	1	0.00	0.00	1.87	< 0.05
TP = interaction	2	0.01	0.00	1.30	< 0.05
E(2) = residual error	6	0.01	0.00		

Appendix 28. Analysis of variance for "a" value of color of raw dark turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.55	0.28	1.47	> 0.05
E(1) =pens/T	6	1.13	0.19		
P = periods	1	11.38	11.38	47.68	< 0.05
TP = interaction	2	0.71	0.35	1.48	> 0.05
E(2) = residual error	6	1.43	0.24		

Appendix 29. Analysis of variance for "a" values of color of raw light turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	5.18	2.59	37.88	< 0.05
E(1) =pens/T	6	0.41	0.07		
P = periods	1	45.25	45.25	119.25	< 0.05
TP = interaction	2	6.98	3.49	9.19	< 0.05
E(2) = residual error	6	2.28	0.38		

Appendix 30. Analysis of variance for "L" value of color of raw dark turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	3.74	1.87	0.87	> 0.05
E(1) =pens/T	6	12.94	2.16		
P = periods	1	12.92	12.92	6.69	< 0.05
TP = interaction	2	0.15	0.07	0.04	> 0.05
E(2) = residual error	6	11.60	1.93		

Appendix 31. Analysis of variance for "L" values of color of raw light turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	4.55	2.28	0.61	> 0.05
E(1) =pens/T	6	22.48	3.75		
P = periods	1	111.50	111.50	25.01	< 0.05
TP = interaction	2	11.64	5.82	1.30	> 0.05
E(2) = residual error	6	26.75	4.46		

Appendix 32. Analysis of variance for "b" value of color of raw dark turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	1.87	0.93	1.92	> 0.05
E(1) =pens/T	6	2.92	0.49		
P = periods	1	0.05	0.05	0.39	> 0.05
TP = interaction	2	1.07	0.53	3.91	> 0.05
E(2) = residual error	6	0.82	0.14		

Appendix 33. Analysis of variance for "b" values of color of raw light turkey meat (frozen study).

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	0.57	0.29	2.98	> 0.05
E(1) =pens/T	6	0.57	0.10		
P = periods	1	23.80	23.80	182.53	< 0.05
TP = interaction	2	0.10	0.05	0.37	> 0.05
E(2) = residual error	6	0.78	0.13		

Appendix 34. Analysis of variance for α-tocopherol contents in plasma samples of turkey during feeding period.

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	53.31	26.65	217.31	< 0.05
E(1) =pens/T	6	0.74	0.12		
P = periods	3	0.87	0.29	2.22	> 0.05
TP = interaction	6	1.47	0.24	1.88	> 0.05
E(2) = residual error	18	2.34	0.13		

Appendix 35. Analysis of variance for body weights of turkey during feeding period.

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	1.56	0.78	1.23	> 0.05
E(1) =pens/T	6	3.81	0.64		

Appendix 36. Analysis of variance for α-tocopherol contents in raw dark meat of turkeys fed three dietary levels of vitamin E

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	22.13	11.07	26.27	< 0.05
E(1) =pens/T	6	2.53	0.42		

Appendix 37. Analysis of variance for α-tocopherol contents in raw light meat of turkeys fed three dietary levels of vitamin E

Source of Variation	Degree of Freedom	Sum of Square	Mean Square	F Value	Probability
T = Treatments	2	4.47	2.24	99.49	< 0.05
E(1) =pens/T	6	0.14	0.02		

REFERENCES

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

Adams, M.L., Sullivan, D.M., Smith, R.L. and Richter, E.F. 1986. Evaluation of direct saponification method for determination of cholesterol in meats. J. Assoc. Off. Anal. Chem. 69:844

Addis, P.B. 1986. Occurrence of lipid oxidation products in foods. Food Chem. Toxic. 24:1021.

Addis, P.B., Csallany, A.S. and Kindom, S.E. 1983. Some lipid oxidation products as xenobiotics. In "Xenobiotics in Foods and Feeds," Chapter 5, p. 85, Eds. Finley, J.W. and Schwass, D.E. Am. Chem. Symp. Ser. 234. J. Am. Chem. Soc., Washington, D.C.

Addis, P.B. and Park, S.W. 1989. Role of lipid oxidation products in atherosclerosis. In "Food Toxicology: A Perspective on the Relative Risks," Chapter 11. Eds. Taylor, S.L. and Scanlan, R.A. Marcel Dekker, Inc., New York.

Allen, C.E. and Foegeding, E.A. 1981. Some lipid characteristics and interactions in muscle foods - a review. Food Technol. 35(5):253.

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.

Asghar, A., Gray, J.I., Booren, A.M., Miller, E.R., Pearson, A.M.,Ku, P.-K. and Buckley, D.J. 1989a. Influence of dietary vitamin E on swine growth and meat quality. Proceedings of the 35th International Congress of Meat Science and Technology, Vol. 3, p. 1065. Copenhagen, Denmark.

Asghar, A., Lin, C.F.,Gray, J.I., Buckley, D.J., Booren, A.M., Crackel, R.L. and Flegal, C.J. 1989b. Influence of oxidised dietary oil and antioxidant supplementation on membrane-bound lipid stability in broiler meat. Brit. Poultry Sci. 30:815.

Asghar, A., Lin, C.F., Gray, J.I., Buckley, D.J., Booren, A.M. and Flegal, C.J. 1990. Effects of dietary oils and α-tocopherol supplementation on membranal lipid oxidation in broiler meat. J. Food Sci. 55:46.

Ashgar, A., Gray, J.I., Miller, E.R., Ku, P.-K., Booren, A.M. and Buckley, D.J. 1991a. Influence of vitamin E supplementation in the feed on swine growth performance and deposition in different tissues. J. Sci. Food Agric. 57:19.

Asghar, A., Gray, J.I., Booren, A.M., Gomaa, E.A., Abouzied, M.M. and Miller, E.R. 1991b. Effects of supranutritional dietary vitamin E levels on subcellular deposition of α-tocopherol in the muscle and on pork quality. J. Sci. Food Agric. 57:31.

Aust, S.D. and Svingen, B.A. 1982. The role of iron in enzymatic lipid peroxidation. In "Free Radicals in Biology," Vol. 5. p. 1. Ed. Pryor, W.A. Academic Press, Orlando, FL.

Badings, H.T. 1970. Cold storage defects in butter and their relation to the autoxidation of unsaturated fatty acids. Ned. Melk Zuiveltijdschr. 24:147.

Bailey, M.E. 1983. The Maillard reaction and meat flavor. In "The Maillard Reaction in Foods and Nutrition" p. 169, Eds. Waller, G.R., and Feather, M.S. ACS Symposium Series 215, J. Am. Chem. Soc., Washington, DC.

Bartov, I., Basker, D. and Angel, S. 1983. Effect of dietary vitamin E on the stability and sensory quality of turkey meat. Poultry Sci. 62:2119.

Bascoul, J., Domergue, N., Olle, M. and Crastes de Paulet, A. 1986. Autoxidation of cholesterol in tallows heated under deep frying conditions: evaluation of oxysterols by GLC and TLC-FID. Lipids. 21:383.

Bauernfeind, J.C. 1980. Tocopherols in foods. In "Vitamin E, A Comprehensive Treatise", p. 99, Eds. Machlin, L.J. Marcel Dekker, New York.

Bendich, A., Gabriel, E. and Machlin, L.J. 1986. Dietary vitamin E requirement for optimum immune responses in the rat. J. Nutr. 166:675.

Bieri, J.G. 1972. Kinetics of tissue α-tocopherol depletion and repletion. Ann. N.Y. Acad. Sci. 203:181.

Bieri, J.G., Corash, L. and Hubbard, V.S. 1983. Medical uses of vitamin E. New Engl. Med. 308:1063.

Bloor, W.R. 1943. "Biochemistry of The Fatty Acids." Van Nostrand-Reinhold. Princeton, NJ.

Body, D.R., Shorland, F.B. and Gass, J.P. 1966. Foetal and maternal lipids of Romney sheep. 1. The composition of the lipids of the total tissues. Biochem. Biophys. Acta 125:207.

Body, D.R., Shorland, F.B. and Czochanska, Z. 1970. Changes in the composition of the rumen and abomasum lipids of sheep from birth to maturity. J. Sci. Food Agric. 21:220.

Brue, R.N. and Latshaw, J.D. 1985. Energy utilization by the broiler chicken as affected by various fats and fat levels. Poultry Sci. 64:2119.

Buckley, J. and Connolly, J.F. 1980. Influence of alpha-tocopherol (vitamin E) on storage stability of raw pork and bacon. J. Food Protect. 43:265.

Buckley, D.J., Gray, J.I., Asghar, A., Price, J.F., Crackel, R.L., Booren, A.M., Pearson, A.M. and Miller, E.R. 1989. Effects of dietary antioxidants and oxidized oil on membranal lipid stability and pork product quality. J. Food Sci. 54:1193.

Buege, J.A. and Aust, S.D. 1978. Microsomal lipid peroxidation. In "Methods in Enzymology". 52:302.

Burr, G.O. and Barnes, R.H. 1943. Non-calorie functions of dietary fats. Physiol. Rev. 23:256.

Burton, G.W., Doba, T., Gabe, E.J., Hughes, L., Lee, F.L., Prasad, L. and Ingold, K.U. 1985. Autoxidation of biological molecules. 4.Maximizing the antioxidant activity of phenols. J. Am. Chem. Soc. 107:7053.

Burton, G.W. and Ingold, K.U. 1981. Autoxidation of biological molecules. 1.The antioxidant activity of vitamin E and related chain-breaking phenolic antioxidants in vitro. J. Am. Chem. Soc. 130:6472.

Burton, G.W., and Ingold, K.U. 1986. Vitamin E: Application of the principles of physical organic chemistry to the exploration of its structure and function. Accounts of Chem. Res. 19:194.

Caygill, C.P., Lucy, J.A. and Diplock, A.T. 1971. The effect of vitamin E on the intracellular distribution of different oxidation stages of selenium in rat liver. Biochem. J. 125:407.

Century, B. and Horwitt, M.K. 1959. Effect of fatty acids on chick encephalomalacia. Proc. Soc. Exp. Biol. Med. 102:375.

Chang, S.S. and Peterson, R.J. 1977. Recent developments in the flavour of meat. J. Food Sci. 42:298.

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

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

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.

Daniels, D.G.H. and Martin, H.F. 1968. Antioxidants in oats:glyceryl esters of caffeic and ferulic acids. J. Sci. Food Agric. 19:710.

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

Dawson, P.L., Sheldon, B.W., Ball, H.R., JR. and Larick, D.K. 1990. Fatty acid composition of neutral lipid and phospholipid fractions of mechanically deboned chicken meat. Poultry Sci. 69:1414.

De Vore, V.R. 1988. TBA values and 7-ketocholesterol in refrigerated raw and cooked ground beef. J. Food Sci. 53:1058.

Drumm, T.D. and Spanier, A.M. 1991. Changes in the content of lipid autoxidation and sulfur-containing compounds in cooked beef during storage. J. Agric. Food Chem. 39(2):336.

Dugan, L.R. Jr., 1971. Fats. In "The Science of Meat and Meat Products." 2nd ed. p. 133. Eds. Price, J.F. and Schweigett, B.S. W.H. Freeman, San Francisco, California.

Dugan, L.R. Jr., 1976. Lipids. In "Principles of Food Science, Part 1. Food Chemistry". p. 182, Eds. Fennema, O.R. Marcel Dekker, Inc. New York and Basel.

Dziezak, J.D. 1986. Preservatives: antioxidants. Food Technol. 40(9):94.

Einerson, M.A. and Reineccius, G.A. 1978. Characterization of antioxidants responsible for inhibition of warmed-over flavor in retorted turkey. J. Food Process. Preserv. 1:279.

Engeseth, N.J. 1990. Membranal lipid oxidation in muscle tissue: mechanism and prevention. Ph.D. Dissertation, Michigan State Univ., E. Lansing, MI.

Enser, M. 1987. What is lipid oxidation. Food Sci. Technol. Today 1:151.

Erin, A.N., Skrypin, V.I., Prilipko, L.L. and Kagan, V.E. 1986. Stabilization effect of α-tocopherol on synaptosomes exposed to the action of phospholipase A_2. Byll. Eksp. Biol. Med. 102:25. (Cited in Chem Abstr 101 Abstr No. 148302g)

Faustman, C. and Cassens, R.G. 1990. The biochemical basis for discoloration in fresh meat: a review. J. Muscle Foods 1:217.

Faustman, C., Cassens, R.G., Schaefer, D.M., Buege, D.R. and Scheller, K.K. 1989a. Vitamin E supplementation of Holstein steer diets improves sirloin steak color. J. Food Sci. 54:485.

Faustman, C., Cassens, R.G., Schaefer, D.M., Buege, D.R., Williams, S.N. and Scheller, K.K. 1989b. Improvement of pigment and lipid stability in Holstein steer beef by dietary supplementation with vitamin E. J. Food Sci. 54:858.

Finocchiaro, E.T., Lee, K. and Richardson T. 1984. Identification and quantification of cholesterol oxides in grated cheese and bleached butter oil. J. Am. Oil Chem. Soc. 61:877.

Folch, J., Lees, M. and Stanley, G.H. 1957. A simple method for the isolation and purification of total lipids from animal tissues. J. Biol. Chem. 226:497.

Fong, K.L., McCay, P.B., Poyer, J.L., Misra, H.P., and Keele, B.B. 1976. Evidence for superoxide-dependent reduction of Fe^{3+} and its role in enzyme-generated hydroxyl radical formation. Chem. Biol. Intl. 15:77.

Foote, C.S. 1985. Chemistry of reactive oxygen species. In: Chemical Changes in Food During Processing. Ed. Richardson T., Finley, J.W. AVI Publishing Company, Inc. Westport, CT, p. 17.

Frankel, E.N. 1962. Hydroperoxides. In "Symposium on Foods and Lipids and Their Oxidation,", Eds. Schultz, H.W., Day, E.A. and Sinnhubuer, R.O. AVI Publishing CO., Westport, CT, p. 51

Frankel, E.N. 1984. Lipid oxidation: mechanisms, products and biological significance. J. Am. Oil Chem. Soc. 61:1908.

Frankel, E.N. 1985. Chemistry of autoxidation: mechanism, products and flavor significance. In "Flavor Chemistry of Fats and Oils," p.1, Eds. Min, D.B., Smouse, T.H. J. Am. Oil Chem. Soc., Champaign, IL.

Frigg, M., Prabucki, A.L., Ruhdel, E.U. 1990. Effect of dietary vitamin E levels on oxidative stability of trout fillets. Aquaculture. 84:145.

Gallo-Torres, H.E. 1980. Biochemistry. In "Vitamin E, A Comprehensive Treatise", p. 170, Eds. Machlin, L.J. Marcel Dekker, New York.

Gardner, H.W. 1979. Lipid hydroperoxide reactivity with proteins and amino acids: a review. J. Agric. Food Chem. 27(2):220.

Gardner, H.W. 1983. Effects of lipid hydroperoxides on food components. In "Xenobiotics in Foods and Feeds," p. 63, Eds. Finley, J.W., Schwass, D.E. ACS Symposium Series 234, J. Am. Chem. Soc., Washington, DC.

German, J.B. and Kinsella, J.E. 1985. Lipid oxidation in fish tissue. Enzymatic initiation via lipoxygenase. J. Agric. Food Chem. 33:680.

Golovnja, R.V. and Rothe, M. 1980. Sulfur containing compounds in the volatile constituents of boiled meat. Nahrung 24:141.

Govindarajan, S., Hultin, H.O. and Kotula, A.W. 1977. Myoglobin oxidation in ground beef: mechanistic studies. J. Food Sci. 42:517.

Gray, J.I. and Pearson, A.M. 1987. Rancidity and warmed-over flavor. In "Advances in Meat Research," Vol. 3, Chapter 6, p.221, Eds. Pearson, A.M. and Dutson, T.R. Van Nostrand Reinhold Co., New York.

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

Greene, B.E., Hsin, I. and Zipser, M.W. 1971. Retardation of oxidative color changes in raw ground beef. J. Food Sci. 36:941.

Grossman, S., Bergman, M. and Sklan, D. 1988. Lipoxygenase in chicken muscle. J. Agric. Food Chem. 36:1268.

Gunstone, F.D. and Norris, F.A. 1983. Oxidation. In "Lipids in Foods: Chemistry, Biochemistry and Technology, "Chapter 7. Pergamon Press, New York.

Gutteridge, J.M.C. 1984. Lipid peroxidation by superoxide-dependent hydroxyl radicals using complexed iron and hydrogen peroxide. FEBS Lett. 172:245.

Hall, G. 1987. Interactions between products of lipid oxidation and proteins. Food Sci. Technol. Today 1:155.

Harel, S. and Kanner, J. 1985. Muscle membranal lipid peroxidation initiated by H_2O_2-activated metmyoglobin. J. Agric. Food Chem. 33:1188.

Haurowitz, F., Schwerin, P. and Mutahhar Yenson, M. 1941. Destruction of hemin and hemoglobin by the action of unsaturated fatty acids and oxygen. J. Biol. Chem. 140:353.

Higley, N.A., Taylor, S.L., Herian, A.M. and Lee, K. 1986. Cholesterol oxides in processed meats. Meat Sci. 16:175.

Hochstein, P. and Ernster, L. 1963. ADP-activated lipid peroxidation coupled to the TPNH oxidase system of microsomes. Biochem. Biophys. Res. Comm. 12:388.

Hoelscher, L.M., Savell, J.W., Smith, S.B. and Cross, H.R. 1988. Subcellular distribution of cholesterol within muscle and adipose tissues of beef loin steaks. J. Food Sci. 53:718.

Houlihan, C.M. and Ho, C. 1985. Natural antioxidants. Ch. 6. In "Flavor Chemistry of Fats and Oils," p.139, Eds. Min, D.B. and Smouse, T.H. J. Am. Oil Chem. Soc., Champaign, IL.

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

Hulan, H.W., Proudfoot, F.G. and Nash, D.M. 1984. The effects of different dietary fat sources on general performance and carcass fatty acid composition of broiler chickens. Poultry Sci. 63:324.

Hultin, H.O. 1980. Enzyme-catalyzed lipid oxidation in muscle microsomes. In "Autoxidation in Food and Biological Systems," p. 505, Eds. Simic, M.G. and Karel, M. Plenum Press, New York.

Hutchins, B.K., Liu, T.H.P. and Watts, B.M. 1967. Effect of additives and refrigeration on reducing activity, metmyoglobin, malonaldehyde of raw ground beef. J. Food Sci. 32:214.

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

Igene, J.O., 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.

Imai, H. 1980. Arterial wall injury and repair due to oxygenated sterols and possible procursors. In "Autoxidation in Food and Biological Systems", p. 613, Eds. Simic, M.G. and Karel, M. Plenum Press, New York.

Jacobson, M. and Koehler, H.H. 1970. Development of rancidity during short time storage of cooked poultry meat. J. Agric. Food Chem. 18:1069.

Johns, A.M., Birkinshaw, L.H. and Ledward, D.A. 1989. Catalysts of lipid oxidation in meat products. Meat Sci. 25:209.

Kameda, K., Imai, M. and Senjo, M. 1985. The effect of vitamin E deficiency on some erythrocyte membrane properties. J. Nutr. Sci. Vitaminol. 31:481.

Kandutsch, A.A., Chen, H.W. and Heiniger, H.J. 1978. Biological activity of some oxygenated sterols. Science. 201:498.

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

Kanner, J., Harel, S. and Hazan, B. 1986. Muscle membranal lipid peroxidation by an "iron redox cycle" system :Initiation by oxyradicals and site-specific mechanisms. J. Agric. Food Chem. 34:506.

Kanner, J., German, J.B. and Kinsella, J.E. 1987. Initiation of lipid peroxidation in biological systems. CRC Crit. Rev. Food Sci. Nutri. 25:317.

Kanner, J. German, B.J. and Doll,L. 1988a. Catalytic "free" iron ions in muscle foods. J. Agric. Food Chem. 36:412.

Kanner, J., Shegalovich, I., Harel, S. and Hazan, B. 1988b. Muscle lipid peroxidation dependent on oxygen and free metal ions. J. Agric. Food Chem. 36:409.

Kanner, J., Sofer, F., Harel, S. and Doll, L. 1988c. Antioxidant activity of ceruloplasmin in muscle membrane and in-situ lipid peroxidation. J. Agric. Food Chem. 36:415.

Kanner, J., Salan, M.A., Harel, S. and Shegalovich, I. 1991. Lipid peroxidation of muscle food: the role of the cytosolic fraction. J. Agric. Food Chem. 39:242.

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.

Kaucher, M., Galbraith, H., Button, V. and Williams, H.H. 1944. The distribution of lipids in animal tissues. Arch. Biochem. 3:203

Keeney, M. 1962. Secondary degradation products. In "Symposium on Foods and Lipids and Their Oxidation," p. 79, Eds. Schultz, H.W., Day, E.A. and Sinnhubuer, R.O. AVI Publishing CO., Westport, CT.

Koch, D.E., Pearson, A.M., Magee, W.T., Hoefer, J.A., and Schweigert, B.S. 1968. Effect of diet on the fatty acid composition of pork fat. J. Anim. Sci. 27:360.

Krishnamurthy, S. and Bieri, J.G. 1963. The absorption, storage, and metabolism of α-tocopherol-C^{14} in the rat and chicken. J. Lipid Res. 4:330.

Kritchevsky, D. and Tapper, S.A. 1961. The free and ester sterol content of various foodstuffs. J. Nutr. 74:441.

Kubow, S. 1990. Toxicity of dietary lipid peroxidation products. Trends Food Sci. Technol. 1(3):67.

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

Ladikos, D. and Lougovois, V. 1990. Lipid oxidation in muscle foods: a review. Food Chem. 35:295.

Lai, S.M. 1989. Effects of oleorestin rosemary on the oxidative quality of restructured chicken products. M.S. Dissertation, Michigan State Univ., E. Lansing, M.I.

Lea, C.H. 1962. The oxidative deterioration of food lipids. In "Symposium on Foods and Lipids and Their Oxidation," p. 3, Eds. Schultz, H.W., Day, E.A. and Sinnhubuer, R.O. AVI Publishing CO., Westport, CT.

Lillard, D.A. 1987. Oxidative deterioration in meat, poultry, and fish. In "Warmed-Over Flavor of Meat," p. 41, Eds. St. Angelo, A.J., Bailey, M. E. Academic Press, Orlando, FL.

Lin, C.F. 1988. Lipid peroxidation in poultry meat and its subcellular membranes as influenced by dietary oils and antioxidant supplementation. Ph.D. Dissertation, Michigan State University, E. Lansing, MI.

Lin, C.F., Asghar, A., Gray, J.I., Buckley, D.J., Booren, A.M., Crackel, R.L. and Flegal, C.J. 1989a. Effects of oxidized dietary oil and antioxidant supplementation on broiler growth and meat stability. Brit. Poultry Sci. 30:855.

Lin, C.F., Gray, J.I., Asghar, A., Buckley, D.J., Booren, A.M. and Flegal, C.J. 1989b. Effects of dietary oils and α-tocopherol supplementation on lipid composition and, stability of broiler meat. J. Food Sci. 54(6):1457.

Lin, T-S. and Hultin, H.O. 1976. Enzymic lipid peroxidation in microsomes of chicken skeletal muscle. J. Food Sci. 41:1976.

Lin, T-S. and Hultin, H.O. 1977. Oxidation of myoglobin in vitro mediated by lipid oxidation in microsomal fractions of muscle. J. Food Sci. 42:136.

Linder, M.C. 1985. Nutrition and metabolism of vitamins. In "Nutritional Biochemistry and Metabolism with Clinical Application," Chapter 5, Ed. Linder, M.C. Elsevier Science Publishing Co., New York.

Liu, H.-P. 1970a. Catalysts of lipid peroxidation in meats. 1. Linoleate peroxidation catalyzed by MetMb or Fe(II)-EDTA. J. Food Sci. 35:590.

Liu, H.-P. 1970b. Catalysts of lipid peroxidation in meats. 2. Linoleate peroxidation catalyzed by tissue homogenates. J. Food Sci. 35:593.

Love, J.D. 1987. Mechanism of iron catalysis of lipid autoxidation in warmed-over flavor of meat. In "Warmed-Over Flavor of Meat," p. 19, Eds. St. Angelo, A.J., Bailey, M. E. Academic Press, Orlando, FL.

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. 32:1032.

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

Love, R.M. and Elerian, M.K. 1974. Protein denaturation in frozen fish. 8. The temperature of maximum denaturation in cod. J. Sci. Food Agric. 15:805.

Lowry, O.H., Rosebrough, N,J., Farr, A.L. and Randall, R.J. 1951. Protein measurement with the Folin phenol reagent. J. Biol. Chem. 193:265.

Luby, J.M. 1982. Photooxidation of cholesterol in butter. M.S. Dissertation, Michigan State Univ., E. Lansing, MI.

Lundberg, W.O. 1962. Mechanisms. In "Symposium on Foods and Lipids and Their Oxidation," p. 31, Eds. Schultz, H.W., Day, E.A. and Sinnhubuer, R.O. AVI Publishing CO., Westport, CT.

Machlin, L.J. 1980. Vitamin E, A Comprehensive Treatise. Ed. Machlin, L.J. Marcel Dekker Inc., New York.

Machlin, L.J. 1984. Vitamin E. In "Handbook of Vitamins," Chapter 3, Ed. Machlin, L.J. Marcel Dekker,Inc., New York.

Machlin, L.J. 1991. Vitamin E. In "Handbook of Vitamins," Chapter 3, 2nd Ed. Machlin, L.J. Marcel Dekker,Inc., New York.

Maerker, G. 1987. Cholesterol autoxidation, Current status. J. Am. Oil Chem. Soc. 64:388.

Marion, J.E. and Woodroof, J.G. 1966. Composition and stability of broiler carcasses as affected by dietary 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 16:5.

Marusich, W.L., DeRitter, E., Ogrinz, E.F., Keating, J., Mitrovic, M. and Bunnell, R.H. 1975. Effect of supplemental vitamin E in control of rancidity in poultry meat. Poultry Sci. 54:831.

McCay, P.B. and King, M.M. 1980. Vitamin E: Its role as a biolobical free radical scavenger and its relationship to the microsomal mixed-function oxidase system. In "Vitamin E", p. 289. L.J. Machlin, Ed., Marcel Dekker, Inc., New York.

McDonald, R.E. and Hultin, H.O. 1987. Some characteristics of the enzymic lipid peroxidation system in the microsomal fraction of flounder skeletal muscle. J. Food Sci. 52:15.

McDonald, R.E., Kelleher, S.D. and Hultin, H.O. 1979. Membrane lipid oxidation in a microsomal fraction of red hake muscle. J. Food Biochem. 3:125.

McGregor, J.T. and Lard, L. 1978. Mutagenicity of plant flavonoids: Structural requirements for mutagenicity in Salmonella typhimurium. Mutation Res. 54:297.

Mead, J.F. 1976. Free radical mechanisms of lipid damage and consequences for cellular membranes. In "Free Radicals", Vol. I, p. 51, Eds. Pryor, W.A. Academic Press, New York.

Mellors, A. and Barnes, M. McC. 1966. The distribution and metabolism of α-tocopherol in rat. Brit. J. Nutr. 20:69.

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

Metha, A.C. and Seshadri, T.R. 1959. Flavonoids as antioxidants. J. Sci. Ind. Res. (India), 18B:24.

Min, D.B.S., Ina, K., Peterson, R.J. and Chang, S.S. 1979. Preliminary identification of volatile flavor compounds in the neutral fraction of roast beef. J. Food Sci. 44:639.

Missler, S.R., Wasilchuk, B.A. and Merritt, C., Jr. 1985. Separation and identification of cholesterol oxidation products in dried egg preparations. J. Food Sci. 50:595.

Molenaar, I., Vos, J. and Hommes, F.N. 1972. Effect of vitamin E deficiency on cellular membranes. Vitam. Horm. New York 30:45.

Molenaar, I., Hulstaert, C.E. and Hardonk, M.J. 1980. Role in function and ultrastructure of cellular membranes. In "Vitamin E: A Comprehensive Treatise," Chapter 5, Part G., Ed. Machlin, L.J. Marcel Dekker, Inc., New York.

Monahan, F.J., Buckley, D.J., Gray, J.I., Morrissey, P.A., Hanrahan, T.J., Asghar, A. and Lynch, P.B. 1989. The effect of dietary α-tocopherol levels in the pig and on oxidation in pork products. Proceedings of the 35th International Congress of Meat Science and Technology, Vol. 3, p. 1071. Copenhagen, Denmark.

Monahan, F.J., Buckley, D.J., Gray, J.I., Morrissey, P.A., Asghar, A., Hanrahan, T.J., and Lynch, P.B. 1990a. The effect of dietary vitamin E on stability of raw and cooked pork. Meat Sci. 27:99.

Monahan, F.J., Buckley, D.J., Morrissey, P.A., Lynch, P.B. and Gray, J.I. 1990b. Effect of dietary α-tocopherol supplementation on α-tocopherol levels in porcine tissues and on susceptibility to lipid peroxidation. Food Science and Nutrition 42F:203.

Monahan, F.J., Crackel, R.L., Gray, J.I., Buckley, D.J. and Morrissey, P.A. 1992a. Catalysis of lipid oxidation in muscle model systems by haem and inorganic iron. Meat Sci. (in press).

Monahan, F.J., Gray, J.I., Booren, A.M., Miller, E.R., Buckley, D.J., Morrissey, P.A. and Gomaa, E.A. 1992b. Influence of dietary treatment on cholesterol oxidation in pork. J. Agric. Food Chem. (in press)

Monahan, F.J., Buckley, D.J., Morrissey, P.A., Lynch, P.B., and Gray, J.I. 1992c. Influence of dietary fat and α-tocopherol supplementation on lipid oxidation in pork. Meat Sci. 31:229.

Morgan, J.N. and Armstrong, D.J. 1987. Formation of cholesterol-5,6-epoxides during spray-drying of egg yolk. J. Food Sci. 52:1224.

Morrissey, P.A. and Apte, S. 1988. Influence of species, haem and non-haem iron fractions and nitrite on hexanal production in cooked muscle systems. Sciences des Aliments. 8:3.

Nair, P.O. 1972. Vitamin E and metabolic regulation. Ann. N.Y. Acad. Sci. 203:53.

Nourooz-Zaeh, J. and Appelqvist, L.A. 1987. Cholesterol oxides in Swedish foods and food ingredients: fresh eggs and dehydrated egg products. J. Food Sci. 52:57.

Nourooz-Zadeh, J. and Appelqvist, L.A. 1988. Cholesterol oxides in Swedish foods and food ingredients: milk powder products. J. Food Sci. 53:74.

Nourooz-Zadeh, J. and Appelqvist, L.A. 1989. Cholesterol oxides in Swedish foods and food ingredients: lard and bacon. J. Am. Oil Chem. Soc. 66:856.

Olcott, H.S. 1962. Marine products. In "Symposium on Food Lipids and Their Oxidation," p. 173, Eds. Schultz, H.W., Day, E.A. and Sinnhubuer, R.O. AVI Publishing CO., Westport, CT.

Packer, J.E., Slater, T.F., and Willson, R.L. 1979. Direct observation of a free radical interaction between vitamin E and vitamin C. Nature (London) 278:737.

Park, S. Won and Addis, P.B. 1985a. HPLC determination of C-7 oxidized cholesterol derivatives in foods. J. Food Sci. 50:1437.

Park, S. Won and Addis, P.B. 1985b. Capillary column gas-liquid chromatographic resolution of oxidized cholesterol derivatives. Anal. Biochem. 149:275.

Park, S. Won and Addis, P.B. 1986. Identification and quantitative estimation of oxidized cholesterol derivative in heated tallow. J. Agric. Food Chem. 34:653.

Park, S. Won and Addis, P.B. 1987. Cholesterol oxidation products in some muscle foods. J. Food Sci. 52:1500.

Park, G.L., Avery, S.M., Byers, J.L. and Nelson, D.B. 1983. Identification of bioflavonoids from citrus. Food Technol. 37(12):98.

Patnaik, P.N. and Nair, P.O. 1977. Studies on the binding of d-alpha-tocopherol to rat liver nuclei. Arch. Biochem. Biophys. 178:333.

Pearson, A.M. 1981. Meat and health. In "Developments in Meat Science-2," p. 241, Eds. Lawrie, R. Applied Science Publishers London, England.

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

Peng, S.K., Taylor, C.B., Tham, P., Werthessen, N.T. and Mikkelson, B. 1978. Effect of autoxidation products from cholesterol on aortic smooth muscle cell. Arch. Pathol. Lab. Med. 102:57.

Peng, S.K., Tham, P., Taylor, C.B. and Mikkelson, B. 1979. Cytotoxicity of oxidation derivatives of cholesterol on cultured aortic smooth muscle cells and their effect on cholesterol biosynthesis. Am. J. Clin. Nutr. 32:1033.

Peng, S.K., Morin, R.J., Tham, P. and Taylor, C.B. 1985. Effects of oxygenated deterives of cholesterol on cholesterol uptake by cultured aortic smooth muscle cells. Artery 13:144.

Peng, S.K., Phillips, G.A., Guang-Zhi, X. and Morin, R.J. 1987. Transport of cholesterol autoxidation products in rabbits lipoproteins. Atherosclerosis 64:1.

Pie, J.E., Spahis, K. and Seillan, C. 1991. Cholesterol oxidation in meat products during cooking and frozen storage. J. Agric. Food Chem. 39:250.

Pikul, J. and Kummerow, F.A. 1991. Thiobarbituric acid reactive substance formation as affected by distribution of polyenoic fatty acids in individual phospholipids. J. Agric. Food Chem. 39:451.

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

Player, T.J. and Hultin, H.O. 1977. Some characteristics of the NAD(P)H-dependent lipid peroxidation system in the microsomal fraction of chicken breast muscle. J. Food Biochem. 1:153.

Popekhina, P. and Tveritnev, M. 1982. Premixes for replacement hogs. Svinovodstov (Moscow) 1982:(1) 34. In "Vitamin E Abstracts 1982" pp. 124. Horwitt, M.K. (Ed). Distributed by Henkel Corporation. Minneapolis, MN.

Pratt, D.E. 1965. Lipid antioxidants in plant tissues. J. Food Sci. 30:737.

Pratt, D.E. 1972. Water soluble antioxidant activity in soybeans. J. Food Sci. 45:1058.

Pryor, W.A. 1976. Free Radicals in Biology. W.A. Pryor Ed. Vol. 1. p.1. Academic Press, New York.

Reindl, B. and Stan, H.J. 1982. Determination of volatile aldehydes in meat as 2,4-dinitrophenylhydrazones using reverse-phase high performance liquid chromatography. J. Agric. Food Chem. 30:849.

Rhee, K.S. 1987. Natural antioxidants for meat products. In "Warmed-Over Flavor of Meat" p.267, Eds. Angelo, A.J. and Bailey, M.E. Academic Press, Inc., Orlando.

Rhee, K.S. 1988. Enzymic and nonenzymic catalysis of lipid oxidation in muscle foods. Food Technol. 42(6):127.

Rhee, K.S., Dutson, T.R., Smith, G.C., Hostetler, R.L. and Reiser, R. 1982a. Cholesterol content of raw and cooked beef longissimus muscles with different degrees of marbling. J. Food Sci. 47:716.

Rhee, K.S., Dutson, T.R. and Smith, G.C. 1982b. Effect of changes in intermuscular and subcutaneous fat levels on cholesterol content of raw and cooked beef steaks. J. Food Sci. 47:1638.

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

Rhee, K.S., Dutson, T.R. and Smith, G.C. 1984. Enzymic lipid peroxidation in microsomal fractions from beef skeletal muscle. J. Food Sci. 49:675.

Rhee, K.S., Dutson, T.R. and Savell, J.W. 1985. Enzymic lipid peroxidation in beef muscle microsomes as affected by electrical stimulation and postmortem muscle excision time. J. Food Biochem. 9:27.

Rhee, K.S., Seideman, S.C. and Cross, H.R. 1986. Enzymic and nonenzymic factors affecting lipid peroxidation in raw beef muscles. J. Agric. Food Chem. 34:308.

Rhee, K.S., Ziprin, Y.A., and Ordonez, G. 1987. Catalysis of lipid oxidation in raw and cooked beef by metmyoglobin-H_2O_2, non- heme iron, and enzyme systems. J. Agric. Food Chem. 35:1013.

Roubal, W.T. and Tappel, A.L. 1966. Polymerization of proteins induced by free-radical lipid peroxidation. Arch. Biochem. Biophys. 113:150.

Ruenger, E.L., Reineccius, G.A. and Thompson, D.R. 1978. Flavor compounds related to the warmed-over flavor of turkey. J. Food Sci. 43:1199.

Salmon, R.E. and O'Neil, J.B. 1973. The effect of dietary fat and storage temperature on the storage stability of turkey meat. Poultry Sci. 52:314.

Sander, B.D., Smith, D.E., Addis, P.B. and Park, S.W. 1989. Effects of prolonged and adverse storage conditions on levels of cholesterol oxidation products in dairy products. J. Food Sci. 54:874.

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

Sato, K. and Herring, H.K. 1973. The chemistry of warmed-over flavor in cooked meats. In Proceedings of 26th. Ann. Reciprocal Meat Conference of the American Meat Science Assoc. Chicago, IL. p. 64.

Schenkman, J.B. and Cinti, D.L. 1978. Preparation of microsomes with calcium. Methods Enzymol. 52:83

Senate Select Committee. 1977a. Dietary Goals for the United States. No. 052-070-03916-2. Senate Select Committee on Nutrition and Human Needs. U.S. Govt. Print. Office, Washington, D.C.

Senate Select Committee. 1977b. Dietary Goals for the United States, 2nd ed., No. 052-070-04376-8. Senate Select Committee on Nutrition and Human Needs. U.S. Govt. Print. Office, Washington, D.C.

Shahidi, F., Rubin, L.J. and D'Souza, L.A. 1986. Meat flavor volatiles: A review of the composition, techniques of analysis, and sensory evaluation. CRC Crit. Rev. Food Sci. Nutr. 24:141.

Sheldon, B.W. 1984. Effect of dietary tocopherol on the oxidative stability of turkey meat. Poultry Sci. 63:673.

Shermer, W.D. and Calabotta, D.F. 1985. Controlling feed oxidation can be rewarding. Feedstuffs. p.24. Nov. 25, 1985.

Shibamoto, T. 1980. Heterocyclic compounds found in cooked meats. J. Agric. Food Chem. 28:237.

Shorland, F.B. 1962. The comparative aspects of fatty acid occurrence and distribution. Comp. Biochem. 3:1.

Shorland, F.B., Igene, J.O., Pearson, A.M., Thomas, J.W., McGuffey, R.K. and Aldridge, A.E. 1981. Effects of dietary fat and vitamin E on the lipid composition and stability of veal during frozen storage. J. Agric. Food Chem. 29:863.

Simpson, T.H. and Uri, N. 1959. Hydroxyflavones as inhibitors of the aerobic oxidation of unsaturated fatty acids. Chem. and Ind. p. 956.

Sklan, D., Tenne, E. and Budowski, P. 1983. The effect of dietary fat and tocopherol on lipolysis and oxidation in turkey meat stored at different temperatures. Poultry Sci. 62:2017.

Slabyj, B.M. and Hultin, H.O. 1982. Lipid peroxidation by microsomal fractions isolated from white and dark muscles of herring (Clupea harengus). J. Food Sci. 47:1395.

Smith, E.L., Hill, R.L., Lehman, I.R. and Lefkowitz, R.J. 1983. Membranes and subcellular organelles. In "Principles of Biochemistry:General Aspects." p. 272. Eds. Smith, E.L. McGraw-Hill Book Company. New York.

Smith, L.L. 1980. The autoxidation of cholesterol. In "Autoxidation in Food and Biological Systems", p.119, Eds. Simic, M.G. and Karel, M. Plenum Press, New York.

Smith, L.L. 1981. "Cholesterol Autoxidation". Plenum Press, New York.

Smith, L.L. 1987. Cholesterol autoxidation 1981-1986. Chem. Phys. Lipids 44:87.

Smith, L.L. and Kulig, M.J. 1975. Sterol metabolism. XXXIV. On the derivation of carcinogenic sterols from cholesterol. Cancer Biochem. Biophys. 1:79.

Smith, L.L., Kulig, M.J. and Teng, J.I. 1973a. Sterol metabolism XXVI. Pyrolysis of some sterol allylic alcohols and hydroperoxides steroids. J. Chromatog. 22:627.

Smith, L.L., Teng, J.I., Kulig, M.J. and Hill, F.L. 1973b. Sterol metabolism. XXIII. Cholesterol oxidation by radiation-induced processes. J. Org. Chem. 38:1763.

Smith, L.L., Teng, J.I., Lin, Y.Y., Seitz, P.K. and McGehee, M.F. 1982. Lipid peroxidation of cholesterol. In "Lipid Peroxides in Biology and Medicine," p.89, Ed. K. Yagi. Academic Press, Inc., London.

St. Angelo, A.J., Crippen, K.L., Dupuy, H.P. and James, Jr. C. 1990. Chemical and sensory studies of antioxidant-treated beef. J. Food Sci. 55:1501.

Stoick, S.M., Gray, J.I., Booren, A.M. and Buckley, D.J. 1991. Oxidative stability of restructured beef steaks processed with oleoresin rosemary, tertiary butylhydroquinone, and sodium tripolyphosphate. J. Food Sci. 56:597.

Stromer, M.H., Goll, D.E. and Roberts, J.H. 1966. Cholesterol in subcutaneous and intramuscular lipid depots from bovine carcasses of different maturity and fatness. J. Anim. Sci. 25:1145.

Stuber, D.C., Bechtel, P.J., Lan, Y.H., McLaren, D.G., Mckeith, F.K., Easter, R.A. and Coelho, M.B. 1990. Effect of vitamin E supplementation on swine growth performance and pork storage properties. J. Anim. sci. 68(Suppl):342.

Svingen, B.A., Buege, J.A., O'Neal, F.O. and Aust, S.D. 1979. The mechanism of NADPH-dependent lipid peroxidation. J. Biol. Chem. 254:5892.

Sweeten, M.K., Cross, H.R., Smith, G.C. and Smith, S.B. 1990. Subcellular distribution and composition of lipids in muscle and adipose tissues. J. Food Sci. 55:43.

Tahin, Q.S., Blum, M. and Carafoli, E. 1981. The fatty acid composition of subcellular membranes of rat liver, heart, and brain:diet-induced modifications. Eur. J. Biochem. 122:5.

Tappel, A.L. 1952. Linoleate oxidation catalyzed by hog muscle and adipose tissue extracts. Food Res. 17:550.

Tappel, A.L. 1953. Linoleate oxidation catalysts occurring in animal tissues. Food Res.18:104.

Tappel, A.L. 1962. Vitamin E as the biological lipid antioxidant. Vitamin and Hormones. Vol. 20. p. 493. Academic Press, New York.

Tappel, A.L. 1963. Lipoxygenase. In "The Enzymes" Eds. Boyer, P.D., Lardy, H., and Myrback, K. Academic. New York, London.

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

Taylor, C.B., Peng, S.K., Werthessen, N.T., Tham, P. and Lee, K.T. 1979. Spontaneously occurring angiotoxic derivatives of cholesterol. Am. J. Clin. Nutr. 32:40.

Teng, J.I., Kulig, M.J., Kan, G. and Van Lier, J.C. 1973a. Sterol metabolism. XX. Cholesterol 7α-hydroperoxide. J. Org. Chem. 38:119.

Teng, J.I., Kulig, M.J. and Smith, L.L. 1973b. Sterol metabolism XXVI. Gas. chromatographic differentiation among cholesterol hydroperoxides. J. Chromatog. 75:108.

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

Tien, M. and Aust, S.D. 1982. Comparative aspects of several model lipid peroxidation systems. In "Lipid Peroxides in Biology and Medicine," p. 23. Ed. Yagi, K. Academic Press, New York.

Tsai, K.S. 1984. Quantification of cholesterol oxidation products by capillary gas chromatography. M.S. Thesis, University of Minnesota, Minneapolis-St. Paul, MN.

Uebersax, M.A., Dawson, L.E., and Uebersax, K.L. 1978. Storage stability (TBA) of meat obtained from turkeys receiving tocopherol supplementation. Poultry Sci. 57:937.

Ullrich, F. and Grosch, W. 1987. Identification of the most intense volatile flavour compounds formed during autoxidation of linoleic acid. Z. Lebensm. Unters. Forsch. 184:277.

Van Lier, J.E. and Smith, L.L. 1970a. Sterol metabolism XI. Thermal decomposition of some cholesterol hydroperoxides. Steroids. 15:485.

Van Lier, J.E. and Smith, L.L. 1970b. Autoxidation of cholesterol via hydroperoxide intermediates. J. Org. Chem. 35:2627.

Verma, M.M., Ledward, D.A., and Lawrie, R.A. 1984. Lipid oxidation and myoglobin formation in sausages containing chick pea flour. Meat Sci. 11:171.

Waku, K. and Uda, U. 1971. Lipid composition in rabbit sarcoplsmic reticulum and occurrence of alkyl ether phospholipids. J. Biochem. 69:483.

Waldroup, P.W., Douglas, C.R., McCall, J.T. and Harms, R.H. 1960. The Effects of santoquin on the performance of broilers. Poultry Sci. 39:1313.

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.

Watts, B.M. 1961. The role of lipid oxidation in lean tissues in flavor deterioration of meat and fish. In "Proceedings of Flavor Chemistry Symposium," p. 83. Campbell Soup Co., Camden, NJ.

Watts, B.M. 1962. Meat products. In "Symposium on Foods: Lipids and Their Oxidation," p. 202, Eds. Shultz, H.W., Day, E.A., and Sinnhuber, R.O. Academic Press, New York.

Webb, J.E., Booren, C.C. and Yates, J.D. 1972. Effects of feeding antioxidants on rancidity development in pre-cooked, frozen broiler parts. Poultry Sci. 51:1601.

Whittern, C.C., Miller, E.E., and Pratt, D.E. 1984. Cottonseed flavonoids as lipid antioxidants. J. Am. Oil Chem. Soc. 61:1075.

Wilson, B.R. 1974. Effect of turkey lipids and phospholipids on warmed-over flavor measured by TBA analysis in muscle from several species. M.S. Thesis, Michigan State University, East Lansing.

Wilson, R.L. 1984. Ill-placed iron, oxygen free radicals and disease: Some recent and not so recent radiation studies. In "The Biology and Chemistry of Activated Oxygen," p. 238. Ed. Bannister, J.V. and Bannister, W.H., Elsevier, New York.

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.

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

Younathan, M.T. and Watts, B.M. 1959. Relationship of meat pigments to lipid oxidation. Food Res. 24: 728.

Zubillaga, M.P. and Maerker, G. 1991. Quantification of three cholesterol oxidation products in raw meat and chicken. J. Food Sci. 56:1194.

MICHIGAN STATE UNIV. LIBRARIES
31293007935806