The effect of dietary tocopherol supplementation on oxidative rancidity in turkey meat

Preston Loring Hayse
Iowa State University

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The effect of dietary tocopherol supplementation on oxidative rancidity in turkey meat

by

Preston Loring Hayse

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# TABLE OF CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>INTRODUCTION</strong></td>
<td>1</td>
</tr>
<tr>
<td><strong>LITERATURE REVIEW</strong></td>
<td>2</td>
</tr>
<tr>
<td>Oxidative Rancidity</td>
<td>2</td>
</tr>
<tr>
<td>Antioxidants and Stabilization</td>
<td>4</td>
</tr>
<tr>
<td>Properties of Vitamin E</td>
<td>8</td>
</tr>
<tr>
<td>Mechanically Deboned Turkey Meat</td>
<td>9</td>
</tr>
<tr>
<td>TBA Analysis</td>
<td>10</td>
</tr>
<tr>
<td><strong>METHODS AND MATERIALS</strong></td>
<td>13</td>
</tr>
<tr>
<td>Experiment I</td>
<td>13</td>
</tr>
<tr>
<td>Turkeys</td>
<td>13</td>
</tr>
<tr>
<td>Sample preparation</td>
<td>14</td>
</tr>
<tr>
<td>TBA analysis</td>
<td>15</td>
</tr>
<tr>
<td>Experiment II</td>
<td>15</td>
</tr>
<tr>
<td>Turkeys</td>
<td>15</td>
</tr>
<tr>
<td>Slaughter and roasting</td>
<td>16</td>
</tr>
<tr>
<td>Sensory evaluation</td>
<td>17</td>
</tr>
<tr>
<td>TBA analysis</td>
<td>17</td>
</tr>
<tr>
<td>Experiment III</td>
<td>18</td>
</tr>
<tr>
<td>Turkeys</td>
<td>18</td>
</tr>
<tr>
<td><strong>RESULTS AND DISCUSSION</strong></td>
<td>20</td>
</tr>
<tr>
<td>Experiment I</td>
<td>20</td>
</tr>
<tr>
<td>Experiment II</td>
<td>Page</td>
</tr>
<tr>
<td>---------------</td>
<td>------</td>
</tr>
<tr>
<td>TBA analysis</td>
<td>24</td>
</tr>
<tr>
<td>Sensory evaluation</td>
<td>27</td>
</tr>
<tr>
<td>Experiment III</td>
<td>28</td>
</tr>
<tr>
<td>Tocopherol analysis</td>
<td>28</td>
</tr>
<tr>
<td>SUMMARY AND CONCLUSIONS</td>
<td>30</td>
</tr>
<tr>
<td>LITERATURE CITED</td>
<td>33</td>
</tr>
<tr>
<td>APPENDIX</td>
<td>36</td>
</tr>
<tr>
<td>ACKNOWLEDGEMENTS</td>
<td>39</td>
</tr>
</tbody>
</table>
INTRODUCTION

With the increased production and consumption of further processed turkey products has come an increasingly difficult problem of quality control. The problem generally manifests itself in the form of off-flavors due primarily to oxidative rancidity within the turkey meat.

Oxidative rancidity and the subsequent development of off-flavors and off-odors are particularly acute where the meat has been mechanically deboned or precooked and held in frozen storage. Mechanical deboning predisposes the meat, which is high in unsaturated lipids and low in vitamin E (10 ug/g, Mecchi et al., 1956), to oxidation due to a severe disruption of cells and combination of the unsaturated lipids with atmospheric oxygen.

Recent work has shown that natural antioxidants added to the diet of growing turkeys can significantly retard the development of off-flavors in precooked turkey meat. The development of a safe, economical antioxidant administration program could be of great benefit to the turkey industry from the quality control standpoint. Such a program may be necessary if the demand for further processed turkey continues to grow.

The following project was conducted in an attempt to develop a practical antioxidant administration program. The project's objectives were to determine the extent (time and amount) of tocopherol supplementation required in controlling autoxidation in meat and to estimate accurately the amount of tocopherol present in the meat.
LITERATURE REVIEW

Oxidative Rancidity

Lipid oxidation is a major cause of deterioration in the quality of meat and meat products. Undesirable changes in color, flavor and nutritive value may occur as meat lipids oxidize and interact with other meat constituents.

Holman (1954) proposed that autoxidative rancidity of fats is primarily by the attack of oxygen on the unsaturated fractions. Lundberg (1962) reported that oxidation of lipid involves primarily autoxidative reactions having oxidative or nonoxidative characteristics. The basic mechanism is presented in Figure 1.

The extent of oxidation occurring in the meat is related to the extent of unsaturation of the lipid fraction in the tissue. Watts (1962) reported that there are two distinct categories of lipids are mainly intracellular and stored. The intracellular muscle lipids are mainly phospholipids while the stored lipids are primarily triglycerides. The phospholipids are very susceptible to oxidation due to their high content of unsaturated fatty acids (Hornstein et al., 1968).

Keskinel et al. (1964) stated that the very nature of lipids, especially the degree of unsaturation of fatty acids, was an important factor in rancidity. They found turkey meat to be higher in TBA\(^1\) numbers than beef, pork, or lamb. Turkey dark meat was higher in TBA numbers than light meat while cooked ground meat samples were reported to have higher TBA numbers than raw, ground meat samples.

\(^1\)TBA is mg malonaldehyde per 1000g meat.
Figure 1. Mechanism of autcatalytic autoxidation.

Initiation:
\[ RH + O_2 \rightarrow \text{free radicals} \]
\[ \text{ROOH} \rightarrow \text{free radicals (e.g. } R^*, \text{ RO}^*, \text{ RO}_2^*, \text{ HO}^* \) \]

Propagation:
\[ R^* + O_2 \rightarrow \text{RO}_2^* \]
\[ \text{RO}_2^* + RH \rightarrow R^* + \text{ROOH} \]

Termination:
\[ R^* + R^* \rightarrow \text{stable (non-radical) end products} \]
\[ \text{R}^* + \text{RO}_2^* \]
\[ \text{RO}_2^* + \text{RO}_2^* \]
A positive relationship between the amount of total lipid and oxidation rate of turkey meat stored for seven days at 4° C was reported by Marion and Forsythe (1964). They too reported higher TBA numbers for dark turkey meat compared with light meat. As the proportion of red to white meat in a mixture increased, TBA values increased and the turkey skin, which is high in total lipid, showed relatively low TBA numbers, similar to white meat.

Acosta et al. (1966) suggested off-odors and off-flavors which develop during storage of meat are due to the proteolipids, phospholipids and, to a lesser extent, the triglycerides involved in autoxidation. They also found that rate of oxidation of total lipid varied between cooked and uncooked turkey.

Antioxidants and Stabilization

Machlin (1962) stated that the more natural antioxidant a tissue contains the more stable the lipid will be. He also reported that in the live animal pro-oxidants will decrease the deposition of antioxidants or catalyze oxidation of lipids.

Research by Nutter et al. (1943) concluded that oxidative rancidity in turkey fat must be due to a lower content of natural antioxidants such as tocopherols. The authors found no other consistent differences between the characteristics of turkey and chicken fat to explain the difference in stability between commercial chicken and turkey fat.

Barnes et al. (1943) reported that the induction time of rat body fat was decreased while the animal was fed a vitamin E deficient diet,
but stability was restored after administration of large doses of tocopherols.

Griddle and Morgan (1947) reported that dietary tocopherols retarded rancidity development in turkey meat. Varying levels of mixed tocopherols were fed for 35 days prior to slaughter. After slaughter, reduced peroxide development in the fat of the treated birds was noted along with increased tocopherol content of the tissue. The control birds also developed off-odors during prolonged storage while the treated birds did not. Similar results were obtained by Kummerow et al. (1948) and Hood et al. (1950).

Major and Watts (1948) found protection against rancidity development in rabbit tissue could be achieved by feeding or injecting high levels of tocopherols.

Hite et al. (1949) concluded that supplementing a turkey's diet with choline and ethanolamine had a stabilizing effect on carcass fat during storage. The authors reported the greatest differences were noted between the supplemented and unsupplemented birds after four and nine months of storage.

Pool et al. (1950) studied the effects of spraying eviscerated turkey with nordihydroguaiaretic acid in propylene glycol. They reported that peroxide formation and off-odors were decreased during frozen storage; however, the antioxidant spray treatment had no effect on the quality of cooked meat.

Mecchi et al. (1953) concluded that the tocopherol content of the fat is the principal, if not the only, factor in chicken fat causing it
to be more stable than turkey fat. Mecchi et al. (1956) found that tocopherol deposition in carcass fat was much greater for chickens than for turkeys when they were fed a diet containing natural levels of tocopherols. The workers found that by adding supplemental (0.1%) tocopherol to the diets of chickens and turkeys, the tocopherol content and stability of carcass fat were increased in both species. The increase was four to six times the natural level in turkeys and two to three times the natural level in chickens; however, the lipids of chickens remained twice as stable as those of turkey. Additional work showed that the stability of turkey fat could be brought to equal that of chicken with the addition of enough tocopherol to the diet of the turkey. The authors concluded that tocopherols play an important part in maintaining the stability of poultry fat.

Watts (1962) demonstrated that catalysts (pro-oxidants) greatly accelerate the rate of oxidation. Peroxides, because of their anti-vitamin E effect, can be classified as pro-oxidants when present in the diet according to Machlin (1962).

Lineweaver et al. (1952) demonstrated that precooked frozen turkey, previously limited in its use by its susceptibility to rancidity, could be effectively utilized with the proper use of antioxidants. The use of propylene glycol, propyl gallate and butylated hydroxy anisole retarded rancidity development.

Zipser and Watts (1961) established that Mullet tissue lipids begin to oxidize rapidly after cooking as evidenced by off-odors and TBA values. It was also found that the oxidation could be decreased but not entirely stopped by freezing or limiting contact with atmospheric oxygen.
Fickett et al. (1968) studied the use of subcutaneous administration of vitamin E in turkeys. TBA numbers of turkey after two months of frozen storage indicated that the treated birds showed less change than those receiving no vitamin E. In contrast, Mickelberry (1970) reported that antioxidant injections (including tocopherol) 5 minutes or 24 hours prior to slaughter had no effect on the stability of turkeys.

Webb et al. (1971) investigated the feeding of dl alpha-tocopherol acetate, ethoxyquin and BHT on rancidity development in pre-cooked, frozen broiler parts through the use of TBA numbers and taste panels. Feeding BHT in amounts up to 0.04% of the diet did not significantly reduce rancidity development; however, 0.04% ethoxyquin did significantly reduce TBA numbers and was noted by the taste panel. Feeding broilers 5 or 10 IU of vitamin E per pound of diet for 36 days prior to slaughter held TBA numbers below those of the control (P<0.01) and, in this case, the effectiveness of vitamin E was confirmed by the taste panel scores.

Webb et al. (1972) studied the effects of tocopherol supplementation of turkeys on the stability of precooked frozen turkey parts and mechanically deboned turkey meat. They provided 10 or 100 IU of tocopheryl acetates per pound of ration and administered corresponding amounts intramuscularly on a weekly basis. Overall TBA numbers were significantly (P<0.01) affected by treatment, storage period, meat type (breast or thigh), treatment X storage interaction, treatment X meat type interaction and storage X meat type interaction. All tocopherol treatments except for the 10 IU oral resulted in lower TBA numbers than those of the control samples. Although no significant correlations were found between taste
panel responses and TBA numbers, panelists preferred breast meat to thigh meat samples when the latter was evaluated for flavor, off-flavor and off-odor.

Properties of Vitamin E

According to Draper and Crallany (1969) the "metabolically active form" of vitamin E in animal tissue may exist only in the minds of its pursuers. Out of research, however, has emerged a great deal of valuable information concerning vitamin E and several metabolites.

Tappel (1973) in a review of vitamin E stated that the biochemical properties of vitamin E are related directly to its important functions. It is completely fat-soluble, thus it occurs in the fatty portions of our food. It is absorbed and transported similarly to fat, is stored in adipose tissue, and functions in all tissues in the stabilization of the lipid portion of the cell's membranous parts.

Approximately 50 to 85 percent of dietary vitamin E at normal dietary levels (25 IU per day for adults) is absorbed in the intestine. After absorption, it is transported mainly in the beta-lipoproteins of the blood. It equilibrates readily between the beta-lipoproteins and the red blood cell membranes. All tissues in the animal body will in time reflect the average intake of the vitamin.

The distribution of vitamin E in the body tissues is determined mainly by its solubility in fats and other lipids; it is stored in the adipose tissue and mobilized with fat. Compared with the more rapid turnover of some of the water-soluble vitamins, vitamin E turns over very slowly. This means that considerable time is required to deplete the body stores and cause a vitamin E deficiency.
The main biochemical property of vitamin E is that it is a rather slow reducing compound. Vitamin E easily reacts with free radicals (compounds having unpaired electrons). A major source of fat-soluble free radicals is polyunsaturated fat that has been oxidized. Vitamin E converts the free radical into a less reactive form sparing the polyunsaturated fats from oxidative deterioration.

Demole (1939) established that alpha-tocopherol and alpha-tocopheryl acetate are virtually non-toxic when administered to mice in amounts of 50 g. of tocopherol per kilogram of body weight.

March et al. (1973) studied the effects of feeding excess amounts (220-2200 IU/kilogram of diet) of vitamin E to chicks. The results demonstrated that growth rate was not affected by a level of 1000 IU but was depressed by 2200 IU of vitamin E per kilogram of diet. Excess vitamin E had a detrimental effect on bone calcification and it increased the requirement for vitamin D. Lowered hematocrit values and lengthened prothrombin time were also noted. The increased prothrombin time was easily reversed by injecting vitamin K, indicating an increased dietary requirement for vitamin K in the presence of excess vitamin E. The findings suggest that excess vitamin E, like other fat-soluble vitamins, must be considered potentially harmful.

Mechanically Deboned Turkey Meat

Mechanically deboned turkey meat is a product of a changing turkey industry. Instead of being concerned with marketing only whole birds, the industry has begun to further process turkeys to meet the demand for convenience foods.
The turkey industry now further processes more than fifty percent of the birds marketed compared with only 13.5 percent in 1962 according to the U.S.D.A. statistics (1973).

The mechanical deboning machine was developed to help cut losses resulting from meat adhering to bones especially in the wings, neck and back.

With the deboning machines come problems of quality control. Maxon (1971) demonstrated that due to increased surface area, bone marrow, contact with metal and atmospheric oxygen, and other factors, the meat is very susceptible to lipid oxidation. His work indicated that a deboning machine could be adjusted to yield products of varying composition and that different machines produce quite variable products.

Maxon (1971) also found that TBA numbers of mechanically deboned meat stored in a cooler and a freezer increased linearly with time.

Essary and Ritchey (1968) performed amino acid analyses on mechanically deboned turkey meat and reported that the breast and leg meat were similar in amino acid composition to beef, chicken, pork, milk and eggs.

The authors suggested that the turkey product was suitable for use in further processed products such as salami, turkey rolls and weiners based on an analysis of moisture, fat, and protein. The dark meat contained 70.78% moisture, 12.34% lipid and 11.76% protein while the light meat contained 67.57% moisture, 15.03% lipid and 13.28% protein.

TBA Analysis

The TBA test is a color reaction resulting from the reaction of one molecule of malonaldehyde and two molecules of 2-thiobarbituric acid. In
1958, Sinnhuber and Yu found the reactive compound in the TBA test to be malonaldehyde, the resultant breakdown product of fatty acid oxidation. Malonaldehyde is a highly reactive dicarbonyl which exists to a very limited extent as a free compound even in highly oxidized fat. The decomposition of hydroperoxides of malonaldehyde derivatives by the acid TBA reagent gives rise to malonaldehyde which condenses with the TBA to form a red color.

Tarladgis et al. (1960) published a procedure for the determination of malonaldehyde in food containing oxidized fat which involves the distillation of malonaldehyde from an acidified slurry of meat. As suggested by Sinnhuber and Yu (1958), the TBA number is defined as the milligrams of malonaldehyde per 1000 grams of sample with 1,1,3,3-tetraethoxy propane as the standard.

Dahle et al. (1962) established that the TBA color developed from the oxidized lipids varied with the profile of polyunsaturated fatty acids in the sample. This was based on evidence that oxidized linolenic acid produced 60-100 times as much color as oxidized linoleic acid and oxidized oleic acid produced no color at all when all were measured at the same level of autoxidation as indicated by peroxide value. Thus, at similar stages of oxidation, a tissue containing greater quantities of highly unsaturated fatty acids would yield a higher TBA reading than would a tissue sample rich in saturated and less unsaturated fatty acids. The researchers, therefore, concluded that the TBA reaction can be used as a direct measure of the extent of oxidation of a particular highly unsaturated fatty acid.
Tarladgis et al. (1964) modified their distillation procedure (Tarladgis et al., 1960) for the quantitative determination of malonaldehyde in rancid food. The authors suggested that since free malonaldehyde is produced during the oxidative breakdown of the unsaturated fatty acids, the amount of malonaldehyde produced can be measured without acid-heat treatment. They concluded that the acid-heat treatment is unnecessary for the reaction of malonaldehyde with TBA.
METHODS AND MATERIALS

Experiment I

Turkeys

Sixteen-week-old Large White male turkeys of Nicholas breeding were obtained from a local turkey grower. The birds, selected at random, had been raised on a standard commercial starter and grower ration throughout the brooding and growing period.

The birds were placed in slat-floored pens with five birds per pen. The treatments were then randomly assigned to each pen (Table 1). All birds received the control ration (Table 2) when they were not receiving the treatment rations.

Table 1. Dietary treatments used in Experiment 1

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Characteristics</th>
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</thead>
<tbody>
<tr>
<td>Control</td>
<td>Control diet + no added vitamin E</td>
</tr>
<tr>
<td>1</td>
<td>Control diet + 100 IU vitamin E(^a) per pound of ration for 2 weeks prior to slaughter</td>
</tr>
<tr>
<td>2</td>
<td>Control diet + 100 IU vitamin E per pound of ration for 4 weeks prior to slaughter</td>
</tr>
<tr>
<td>3</td>
<td>Control diet + 100 IU vitamin E per pound of ration for 6 weeks prior to slaughter</td>
</tr>
<tr>
<td>4</td>
<td>Control diet + 100 IU vitamin E per pound of ration for 8 weeks prior to slaughter</td>
</tr>
</tbody>
</table>

\(^a\)Added as dl alpha-tocopheryl acetate in powder form directly to 100-lb. batches of feed.
Table 2. Composition of the control ration

<table>
<thead>
<tr>
<th>Ingredients, %</th>
<th>Soybean Meal</th>
<th>Dical Trace Vit. Cora Heal Phos. Limestone</th>
<th>Trace Salt</th>
<th>Vit. Mix^a</th>
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<td>Corn</td>
<td>81</td>
<td>15</td>
<td>2</td>
<td>1</td>
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</table>

^a Contained no vitamin E.

Sample preparation

When the turkeys reached 24 weeks of age they were taken to a commercial processor where they were slaughtered and hand deboned. The rack (sternum and rib cage) and neck were ground in a food chopper and mechanically deboned in a Beehive Deboner (Model AV 1259 MR). The deboned product from each pen was pooled into one common batch since the deboned product would not be identified with individual birds.

The deboned product was then returned to the Iowa State University poultry products laboratory where each treatment batch was divided into seven 100-gram samples. Each sample was placed in a 3" x 5" NASCO Whirl Pak bag. Two samples from each treatment were frozen immediately for testing after 90 and 120 days of storage at -15° C. The remainder of the samples were held at 5° C. On the day of slaughter and at two, four, six and eight days after slaughter, one 100-gram sample from each treatment was removed from the 5° C cooler and duplicate TBA determinations performed.
TBA analysis

Duplicate TBA determinations were performed on the deboned product from each treatment at each sampling period. The determinations were performed using the procedure of Tarladgis et al. (1960), with a few exceptions to standardize the procedure. The exceptions amounted to using a set amount of time for thawing, weighing, grinding, mixing and transferring the sample to the Kjeldahl flask. If the sample needed to be thawed it was allowed to stand at room temperature for five minutes before placing the sample bags in a pan of crushed ice. After thawing, the samples were ground twice (if not a deboned product) in a Hobart K5-A grinder, placed in a plastic bag and returned to the crushed ice. The grinding was completed within 10 minutes. The ground samples were removed from the crushed ice and weighed into 50 ml. beakers and then returned again to the crushed ice. The weighing step was also completed within ten minutes. The weighed samples were mixed with 50 ml. of deionized water in a 500 ml. Omni Mixer cup for 15 seconds prior to transfer to the Kjeldahl flask. Mixing and transfer were completed within 15 minutes. None of the steps were started until the previous time period had elapsed. Using such a schedule helped to standardize the procedure against time variation as the experiment advanced.

Experiment II

Turkeys

Sixteen-week-old Large White male turkeys were obtained from the Iowa State University Poultry Center. The turkeys, having been grown on
standard corn-soy turkey starter and grower diets, were selected at random for this experiment.

The birds were placed in cement floored pens with four inches of wood shavings as litter. Five birds were placed in each pen with duplicate pens for each treatment. The treatments (Table 3) were randomly assigned to each pen. All birds were maintained on the treatment diets for 8 weeks prior to slaughter at 24 weeks of age.

Table 3. Dietary treatments used in Experiment 2

<table>
<thead>
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<th>Treatment</th>
<th>Characteristics</th>
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<tr>
<td>Control</td>
<td>Control diet + no added vitamin E</td>
</tr>
<tr>
<td>1</td>
<td>Control diet + 25 IU vitamin E per pound of ration</td>
</tr>
<tr>
<td>2</td>
<td>Control diet + 50 IU vitamin E per pound of ration</td>
</tr>
<tr>
<td>3</td>
<td>Control diet + 75 IU vitamin E per pound of ration</td>
</tr>
<tr>
<td>4</td>
<td>Control diet + 100 IU vitamin E per pound of ration</td>
</tr>
</tbody>
</table>

^See Table 2.

Slaughter and roasting

The birds were removed at random, one per pen per day, and processed at the Iowa State University Poultry Center. A representative sample of both the right breast and thigh were removed and immediately used for TBA analysis. The remainder of the bird was placed in a rotary hearth oven and roasted at 162° C to an internal breast temperature of 82° C.
After the roasting was completed, a sample of the remaining right breast and thigh was taken for TBA analysis. The left breast and thigh were removed and vacuum packed in separate Cryovac bags. The bags were placed in a blast freezer and frozen at -25°C. The frozen, cooked samples were held in frozen storage at -15°C for seven months prior to subjecting them to sensory evaluation.

**Sensory evaluation**

Sensory evaluations with a panel of eight members were conducted after the meat had been in storage seven months. Two training sessions were held to acquaint the panelists with both fresh and slightly rancid turkey flavor. Half of the members had prior experience on turkey taste panels.

Five breast and five thigh samples were removed at random from the freezer daily and allowed to thaw overnight in a 5°C cooler. The samples were then heated at 162°C to an internal temperature of 55°C. A sample was then removed from each breast and thigh for TBA analysis.

Ten samples were presented to the panelists daily and each member was asked to evaluate the samples for flavor, off-flavor and off-odor on an ascending scale of 1 - 8. Flavor was scored with 1 indicating extremely undesirable and 8 indicating extremely desirable. For off-flavor and off-odor a score of 1 indicated imperceptible detection and a score of 8 indicated an extremely pronounced detection.

**TBA analysis**

The TBA determinations were performed as in Experiment I with the
exception that the samples in this experiment had to be ground. The meat was ground twice in a Hobart K5-A grinder, placed in a plastic bag, and then the bag was placed in a pan of crushed ice until needed for analysis.

Experiment III

Turkeys

Twenty-week-old Large White male turkeys were obtained from the Iowa State University Poultry Center. The birds were selected at random from a flock on range that had been reared on standard corn-soy starter, grower and finisher rations.

The birds were placed in slat-floored pens with five birds per pen. The treatments were then assigned to each pen (Table 4).

Table 4. Experiment III treatments

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Characteristics</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>Control diet$^a$</td>
</tr>
<tr>
<td>1</td>
<td>Control diet + 100 IU vitamin E per pound of ration</td>
</tr>
<tr>
<td>2</td>
<td>Control diet + 500 IU vitamin E per pound of ration</td>
</tr>
</tbody>
</table>

$^a$See Table 2.

After sixteen weeks on the experimental rations, two birds from each treatment were slaughtered. Fifty gram samples of breast and thigh meat were taken from each bird. The breast and thigh meat from each treatment were pooled, placed in Whirl-Fak bags, frozen in liquid nitrogen, packed
in dry ice and shipped to Hoffman-LaRoche Inc., Nutley, New Jersey for quantitative tocopherol analysis.
RESULTS AND DISCUSSION

Experiment I

Some mechanically deboned turkey meat samples were held at 5° C for a period of 8 days while those in frozen form were stored for 90 or 120 days. During that time the samples were subjected to TBA analysis seven times. The samples represented five different time periods in which the turkeys had been given supplemental dietary tocopherol. The results of the analysis of variance of the TBA values are presented in Table 1 (Appendix). Significant differences ($P < 0.01$) were found due to treatment, days and treatment by day interaction.

Figure 2 shows that compared with the control each treatment had a positive effect on controlling oxidative changes, as indicated by the magnitude of the TBA numbers. It is obvious that treatments 3 and 4 (100 IU of vitamin E for 6 and 8 weeks respectively) had the greatest beneficial effect. Treatment 2 (4 weeks) was also effective but it was more or less intermediate between treatments 1, and 3 and 4; treatment 1 was similar to the control.

Table 5 shows a linear increase in the TBA numbers over the initial 8-day test period. These results concur with those of Maxon (1971) and Webb et al. (1972) who also found that TBA numbers increased with time. It is interesting to note that the control samples reached a plateau on the second day of the test and remained there throughout the entire experiment. Possibly the meat had reached a point of maximum malonaldehyde production and all of the natural antioxidants present in the meat were utilized. Treatments 3 and 4 also seem to have reached a
Figure 2. The effect of vitamin E on TBA numbers of mechanically deboned turkey.
Table 5. Mean TBA numbers of mechanically deboned turkey meat

<table>
<thead>
<tr>
<th>Storage (days)</th>
<th>Overall Mean</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>0</td>
</tr>
<tr>
<td>Control</td>
<td>0.802</td>
</tr>
<tr>
<td>1</td>
<td>0.400</td>
</tr>
<tr>
<td>2</td>
<td>0.274</td>
</tr>
<tr>
<td>3</td>
<td>0.144</td>
</tr>
<tr>
<td>4</td>
<td>0.169</td>
</tr>
</tbody>
</table>

Tableau by the fourth day. Although the TBA numbers for treatment 3 (6 weeks) were slightly higher than those of treatment 4 (8 weeks), the two ran parallel throughout the experiment.

The results of the experiment are interesting in the fact that the tocopherol supplementation had its greatest effect during the first few days of the experiment. Between 8 and 90 days the tocopherol seems to have lost some of its effectiveness as evidenced by the dramatic increases in the TBA numbers. TBA numbers for treatment 1 (2 weeks) were too high initially to demonstrate a change after 90 days of storage. After 120 days of storage the TBA numbers seem to group together with the control, treatments 1 and 2 in one group and treatments 3 and 4 in another group (Figure 2).

From the above data, it can be concluded that, at a supplemental level of 100 IU of vitamin E per pound of diet, definite control of oxidative deterioration can be achieved if the supplementation is
continued for a sufficient period of time. Six to eight weeks of supplementation seems sufficient to achieve the desired results while 4 weeks is marginal; 2 weeks will not produce acceptable results.

Maxon (1971) found that meat from mechanical deboners varied with the adjustment of the machine and between machines. The question arises as to whether the differences noted in this experiment could be decreased or eliminated entirely depending on which deboning machine was used and how it was adjusted. Further testing with other experimental variables would be desirable.

Experiment II

TBA analysis

Representative samples of breast and thigh meat to be used for TBA analysis were taken from turkey carcasses in the raw state and after cooking. Cooked, intact breast meat and thighs were packed in plastic bags and frozen for subsequent storage tests and sensory evaluation. The initial mean TBA numbers are presented in Table 6.

Significant differences (P< 0.01) due to treatment, tissue type (breast or thigh) and cooking were found. As can be seen in Table 6, the treatments had their greatest effects on the cooked samples. The TBA values increased four-fold after cooking, confirming that heat is instrumental in the development of oxidative rancidity. Other workers, Zipser and Watts (1961) and Acosta et al. (1966) also reported that cooking meat increases the rate at which the lipids oxidize.
Table 6. Initial TBA numbers of cooked and raw turkey meat

<table>
<thead>
<tr>
<th>Sample</th>
<th>Control</th>
<th>Treatment 1 (25 IU/lb.)</th>
<th>Treatment 2 (50 IU/lb.)</th>
<th>Treatment 3 (75 IU/lb.)</th>
<th>Treatment 4 (100 IU/lb.)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breast (raw)</td>
<td>0.180&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.159&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.167&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.143&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.145&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>Breast (cooked)</td>
<td>0.841&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.853&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.653&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.467&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.391&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>Thigh (raw)</td>
<td>0.253&lt;sup&gt;a&lt;/sup&gt;</td>
<td>0.282&lt;sup&gt;b&lt;/sup&gt;</td>
<td>0.174&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.163&lt;sup&gt;cd&lt;/sup&gt;</td>
<td>0.156&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
<tr>
<td>Thigh (cooked)</td>
<td>1.161&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.134&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.010&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.696&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.639&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>1</sup>Values in each row not followed by a common superscript are different at the P<0.01 level of significance.

With the exception of Treatment 1 on cooked breast and raw thigh, each treatment showed a marked influence on the reduction of rancidity. Seventy-five and 100 IU of tocopherol per pound of feed produced the greatest decrease in rancidity while 50 IU per pound gave somewhat mediocre but significant (P<0.01) results compared with 25 IU per pound.

Treatment 1 (25 IU/lb.) gave significantly (P<0.01) lower TBA numbers than the controls, however, it was not consistent as indicated by the results on cooked breast and raw thigh samples. It appears that Vitamin E supplementation at 25 IU per pound of diet (Treatment 1) was not adequate to consistently give a reduction in the TBA numbers.

Looking at Treatment 2 (50 IU/lb.) there was a significant (P<0.01) reduction in TBA numbers in all instances except with the raw breast sample. Possibly there was some error in analysis of raw breast samples in Treatment 1 causing them to be lower than those in Treatment 2,
however, it is not likely with 10 samples being used.

Based on the data in Table 6, treatments of 75 and 100 IU per pound produced the greatest reduction in TBA numbers. Treatments 3 and 4 were not significantly different for each tissue type. They were significantly different, however, on both cooked samples. This indicates that Treatment 4 is the best overall supplementation level, but, if turkeys were not to be used in a precooked product, then Treatment 3 would be adequate for reducing rancidity.

Table 7 shows the TBA numbers, flavor, off-flavor and off-odor scores of precooked turkey meat held at -15° C for seven months.

Significant differences (P<0.01) in TBA numbers due to treatment and tissue were found. As expected the thigh meat had undergone more oxidative change than breast meat. Here again this is probably due to the amount of lipid in the meat plus the fact that it was cooked. This agrees with the work of Marion and Forsythe (1964) who found that red muscle oxidizes at a more rapid rate than white muscle.

The increase in TBA numbers due to cooking (Table 6) and storage (Table 7) appear to be fairly similar. The TBA number for cooked thigh meat was 0.91 greater than that for raw thigh while breast meat showed a corresponding increase of 0.66. During storage thigh meat increased in TBA number by an average of 0.57 and breast meat by 0.47.

It appears that the tocopherol treatments, especially treatments 3 and 4 (75 and 100 IU/lb.), had a very beneficial effect in minimizing oxidative rancidity in the meat. There appears to be a very definite breaking point between treatments 2 and 3 (25 and 50 IU/lb.). Although
Table 7. TBA numbers, flavor, off-flavor and off-odor scores of cooked turkey after seven months frozen storage

<table>
<thead>
<tr>
<th>Variable</th>
<th>Sample</th>
<th>Control</th>
<th>1 25 IU/lb.</th>
<th>2 50 IU/lb.</th>
<th>3 75 IU/lb.</th>
<th>4 100 IU/lb.</th>
</tr>
</thead>
<tbody>
<tr>
<td>TBA</td>
<td>breast</td>
<td>1.3&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.3&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.3&lt;sup&gt;c&lt;/sup&gt;</td>
<td>0.9&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.8&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>thigh</td>
<td>1.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.6&lt;sup&gt;b&lt;/sup&gt;</td>
<td>1.4&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1.1&lt;sup&gt;d&lt;/sup&gt;</td>
<td>0.9&lt;sup&gt;e&lt;/sup&gt;</td>
</tr>
<tr>
<td>Flavor</td>
<td>breast</td>
<td>5.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.4&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.3&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>thigh</td>
<td>4.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.9&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.8&lt;sup&gt;a&lt;/sup&gt;</td>
<td>5.0&lt;sup&gt;a&lt;/sup&gt;</td>
<td>4.9&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Off-flavor</td>
<td>breast</td>
<td>2.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.6&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.8&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>thigh</td>
<td>2.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.4&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.2&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>Off-odor</td>
<td>breast</td>
<td>1.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.8&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.4&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.7&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td></td>
<td>thigh</td>
<td>2.2&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.9&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.5&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values not followed by a common superscript are different at the P < 0.01 level of significance. This comparison is valid among means within a row.

Treatment 2 was beneficial, treatments 3 and 4 resulted in the greatest control of oxidative rancidity in the meat.

Sensory evaluation

Seven to nine panel members were present for each session. The panelists evaluated the meat for flavor, off-flavor and off-odor. The statistical analysis showed no significant differences due to treatment, replicate group, or tissue and treatment interaction (Tables 9, 10, and 11, Appendix). The panelists did, however, distinguish between tissues (breast or thigh).
The mean flavor scores are presented in Table 7. Significant differences (P<0.01) were found between breast and thigh samples. It is quite apparent that the panelists rated breast meat over thigh meat in flavor intensity as indicated by the scores in Table 7. The apparent treatment differences in oxidative change as shown by TBA numbers were not detected by the panelists. As noted by Webb et al. (1972) panelists may prefer turkey meat that is slightly rancid.

The off-flavor scores (Table 7) were similar within breast and thigh samples but there were differences between the tissue types, with thigh meat having the higher off-flavor scores. Treatment had no effect on the off-flavor scores. It is interesting to note, however, that there was a slight trend toward lower scores as the supplementation level of tocopherol increased. This was especially true for thigh meat.

As with flavor and off-flavor, the off-odor scores (Table 7) showed significant differences due only to tissue type and not treatment. The scores of the thigh meat showed slight but non-significant benefits from Treatments 3 and 4. Thus the off-odor scores agree with the TBA scores even though there are no significant differences.

The data in Table 7 seem to indicate that the results of the taste panel and TBA analysis follow the same general trend. In general, it appeared that the tocopherol treatments had a beneficial effect on the retardation of the rancidity in the meat, especially thigh.
Experiment III

Tocopherol analysis

Fifty grams of breast and thigh meat from the three different treatments were analyzed for tocopherol content by Hoffman-LaRoche Inc., Nutley, New Jersey. The results of the analysis are presented in Table 8.

Table 8. Alpha tocopherol content (mg/100 g.) of turkey breast and thigh meat

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Sample</th>
<th>Control</th>
<th>100 IU/lb.</th>
<th>500 IU/lb.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breast</td>
<td></td>
<td>0.15</td>
<td>0.52</td>
<td>1.06</td>
</tr>
<tr>
<td>Thigh</td>
<td></td>
<td>0.64</td>
<td>1.07</td>
<td>2.78</td>
</tr>
</tbody>
</table>

The results of this experiment show that tissue tocopherol is influenced by ration treatment. It also shows that the thigh muscle contains a greater amount of tocopherol than does the breast. This finding was expected due to the amount of fat present in thigh tissue.

Mecchi et al. (1956) reported that the natural tocopherol content of turkey carcass fat was 10 ug. per gram. They found that by adding 0.1% d alpha-tocopheryl acetate to the diet for 6 or 9 weeks prior to slaughter they could recover 1.0 ug. of tocopherol per 100 g. of extracted fat from thigh meat after 6 weeks and 1.80 ug. per 100 g. after 9 weeks of tocopherol supplementation.

The tocopherol levels found in this experiment and those found by Mecchi et al. (1956) compare quite favorably. It has been demonstrated
that the tocopherol level in the tissue can be increased through dietary supplementation. Thus, an increased tocopherol content in the tissue could be used to increase induction time and slow down oxidative rancidity.
SUMMARY AND CONCLUSIONS

Large White male turkeys were raised on standard commercial starter and grower rations throughout the brooding and growing periods. At 16 weeks of age the birds were assigned to pens and received supplementary dietary tocopherol (100 IU of dl alpha-tocopheryl acetate per pound of feed) for either 2, 4, 6, or 8 weeks prior to slaughter. The birds were commercially slaughtered and deboned. The deboned product was refrigerated and tested for TBA-reactive substances on the day of slaughter and 2, 4, 6 and 8 days after slaughter. A portion of the initial deboned product was frozen for testing after 90 or 120 days of frozen storage at -15°C.

Compared to the control, each treatment had a positive effect on controlling oxidative changes. Supplementation for 6 and 8 weeks had the greatest beneficial effect. Although the 4 week supplementation was effective, it was intermediate between the 2 and 6 week supplementation periods. Two weeks of supplementation produced an effect similar to that of the control.

TBA numbers increased linearly with time, however, the control samples reached a TBA plateau on the second day after slaughter. The tocopherol had its greatest effect during the first few days of the experiment as evidenced by the dramatic increase in TBA numbers after 90 days of storage. A supplemental level of 100 IU of tocopherol per pound of diet can have a definite beneficial effect on oxidative deterioration if continued for at least 6 weeks prior to slaughter.
Sixteen-week-old Large White male turkeys were supplemented with 25, 50, 75 or 100 IU of tocopherol per pound of diet for 8 weeks prior to slaughter. At 24 weeks of age, the birds were slaughtered and one breast and one thigh sampled immediately for TBA-reactive substances. The remainder of the bird was roasted at 162° C in a rotary hearth oven to an internal breast temperature of 82° C. A sample of the cooked breast and thigh meat was taken immediately after roasting for TBA analysis and the remainder frozen at -25° C. The samples were held at -15° C for seven months and then subjected to an eight-member taste panel. The samples were evaluated for flavor, off-flavor and off-odor. TBA analyses were conducted immediately after the taste panels.

Significant differences (P<0.01) due to treatment, tissue type and cooking were found. The treatments had their greatest effect on the cooked samples. With the exception of the treatment of 25 IU, each treatment showed a marked influence on the reduction of rancidity in cooked breast and thigh. Seventy-five and 100 IU produced the greatest decrease in rancidity. It appears that the 25 IU treatment level is not adequate to give a consistent reduction in TBA numbers. One hundred IU per pound is the best overall supplementation level if a bird is going to be further processed, but if it is not, the 75 IU level is adequate.

Significant differences (P<0.01) in TBA numbers due to treatment and tissue type were found in the pre-cooked meat. The statistical analysis of the sensory evaluations showed no significant differences in flavor, off-flavor or off-odor due to treatment, replicate group or tissue x treatment interaction. The panelists could, however, distinguish
between tissue types (breast and thigh). The results of the sensory evaluations seem to indicate that some people prefer slightly rancid meat. In general, it appeared that tocopherol treatments had a beneficial effect on retarding rancidity in the meat, especially in the thigh.

Fifty gram samples of turkey breast and thigh meat from Large White males were assayed quantitatively for tocopherol determination. The turkeys had been supplemented for 16 weeks with 100 or 500 IU of dl alpha-tocopheryl acetate per pound of diet. The data show a definite accumulation of tocopherol in the tissue. The 500 IU treatment produced tissue containing approximately twice as much tocopherol as the 100 IU level and four times as much as the control group.

The results of this work indicate that the use of dietary tocopherol supplementation is at least a partial answer to controlling oxidative stability in turkey meat.
LITERATURE CITED


Sinnhuber, R. O. and T. C. Yu. 1958. 2-thiobarbituric acid method for the measurement of rancidity in fishery products II. The quantitative determination of malonaldehyde. Food Technol. 12:9-12.


APPENDIX

Table 9. Analysis of variance table for flavor scores

<table>
<thead>
<tr>
<th>Source</th>
<th>D.F.</th>
<th>Mean Sq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>4</td>
<td>0.0239</td>
</tr>
<tr>
<td>Tissue</td>
<td>1</td>
<td>0.5864*</td>
</tr>
<tr>
<td>Tissue x treatment</td>
<td>4</td>
<td>0.0462</td>
</tr>
<tr>
<td>Residual</td>
<td>710</td>
<td>0.0486</td>
</tr>
</tbody>
</table>

*Significant at the 0.01 level of probability.

Table 10. Analysis of variance table for off-flavor scores

<table>
<thead>
<tr>
<th>Source</th>
<th>D.F.</th>
<th>Mean Sq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>4</td>
<td>0.1473</td>
</tr>
<tr>
<td>Tissue</td>
<td>1</td>
<td>0.6465*</td>
</tr>
<tr>
<td>Tissue x treatment</td>
<td>4</td>
<td>0.0594</td>
</tr>
<tr>
<td>Residual</td>
<td>710</td>
<td>0.0575</td>
</tr>
</tbody>
</table>

*Significant at the 0.01 level of probability.
Table 11. Analysis of variance table for off-odor scores

<table>
<thead>
<tr>
<th>Source</th>
<th>D.F.</th>
<th>Mean Sq.</th>
</tr>
</thead>
<tbody>
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<td>0.1405</td>
</tr>
<tr>
<td>Tissue</td>
<td>1</td>
<td>0.2887*</td>
</tr>
<tr>
<td>Tissue x treatment</td>
<td>4</td>
<td>0.1188</td>
</tr>
<tr>
<td>Residual</td>
<td>710</td>
<td>0.0428</td>
</tr>
</tbody>
</table>

*Significant at the 0.01 level of probability.

Table 12. Analysis of variance of stored meat TBA numbers

<table>
<thead>
<tr>
<th>Source</th>
<th>D.F.</th>
<th>Mean Sq.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>4</td>
<td>0.2882*</td>
</tr>
<tr>
<td>Tissue</td>
<td>1</td>
<td>0.2194*</td>
</tr>
<tr>
<td>Tissue x treatment</td>
<td>4</td>
<td>0.0161</td>
</tr>
<tr>
<td>Residual</td>
<td>100</td>
<td>0.0056</td>
</tr>
</tbody>
</table>

*Significant at the 0.01 level of probability.
ACKNOWLEDGEMENTS

I wish to express my sincere gratitude to Dr. W. W. Marion for his guidance and counsel during the course of the research. Appreciation is also expressed to the members of my graduate committee, especially Dr. S. L. Balloun. Finally I wish to express my sincere gratitude to my wife Katie for her patience and understanding.