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The Effect of Unidentified Factor Sources, Antibiotics and D-a-Tocopheryl Acetate on the Reproductive Performance of Broad Breasted Bronze and Beltsville Small White Turkeys

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SUMMARY

This bulletin presents a review of the work done in nutrition of turkey breeder hens at the Texas Agricultural Experiment Station since 1950. These investigations include tests with Broad Breasted Bronze and Beltsville Small White turkeys reared to maturity on the range or on wire floors. Various combinations of unknown hatchability factor sources, vitamins, antibiotics and antioxidants have been fed in an attempt to improve the reproductive performance of the turkey breeder hen.

Increased hatchability of fertile eggs was obtained by the addition of unidentified hatchability sources (fish meal, liver "L", distillers dried solubles, condensed fish solubles, dehydrated alfalfa meal, dried whey and dried brewers yeast) at levels of 3 to 5 percent to the diets of Broad Breasted Bronze and Beltsville Small White turkey hens.

The highest hatchability values were obtained when combinations of two or three of the above sources of unidentified factors were included as supplements in the breeder diet. Such combinations maintained hatchability at high levels for as long as 17 to 24 weeks, while the hatchability without these supplements dropped to extremely low levels after the first 8 to 12 weeks of the laying period.

The addition of supplemental vitamin E (D-alpha-tocopheryl acetate) to turkey breeder rations, in the presence of unidentified factor sources, produced an additional increase in the hatchability of eggs.

Unidentified hatchability factor sources included in the diet resulted in a decreased embryonic mortality during the first and last week of the incubation period.

The amount of tocopherol per egg yolk, interpreted as a measure of vitamin E in the diet, is reported for several experiments. The results indicate that unidentified hatchability factors and antioxidants included in the diet do not affect the tocopherol content of the yolk. The addition of vitamin E (in a stable form of D-alpha-tocopheryl acetate) improved the hatchability of eggs from hens fed an all-vegetable protein diet, without unidentified factors. These data indicate that natural feed ingredients may not contain vitamin E in a sufficient amount or in a form which is available for use by the turkey hen.

Embryos from hens fed an all-vegetable protein diet, without supplemental vitamin E, had cataracts and bulging eyes, and a high percent of these embryos died during the last week of the incubation period. The addition of vitamin E to the turkey hen diet reduced this embryonic mortality and prevented the blindness.

The addition of dried brewers yeast, alfalfa meal and fish solubles, singly and in various combinations, as supplements to an all-vegetable protein diet resulted in increased egg production in two experiments. The average amount of feed required to produce an egg ranged from 1.12 to 1.66 pounds. The combination of dried brewers yeast with condensed fish solubles produced the best results in these two studies.

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APPROXIMATELY 3½ TO 4 MILLION turkey breeder hens are maintained in the United States each year, with a total of 80 to 100 million poults being hatched from eggs produced by these hens. At an average value of 75 cents per poult, the expected income would be 60 to 75 million dollars annually from this phase of the 350 million dollar turkey industry. Approximately 65 to 75 million turkeys, consuming approximately 3 million tons of feed, are raised annually from these poults.

The feeding of turkey breeder hens for maximum egg production, fertility and hatchability has changed considerably during recent years as a result of research in this field. The hatchability of eggs from turkey hens usually is expected to be fairly good during the first 8 to 12 weeks of the production period. Since hatchability and fertility normally decline after this time, it has not been considered economically desirable to maintain breeder flocks for a longer period of time.

Nestler *et al.* (1936) reported that an all-vegetable protein diet would not maintain high hatchability unless the diet was supplemented with pork liver meal, dessicated meat meal, fish meal or dried buttermilk. These workers also reported that dried whey would not maintain high hatchability. Dickens *et al.* (1941) later reported that distillers dried grains with solubles would not replace dried skim milk, fish meal or meat scraps in the New England Conference laying ration for turkeys as far as hatchabiltiy was concerned. Stadelman *et al.* (1950) reported that vitamin D had no effect on egg production but that the absence of vitamin D from the turkey breeder diet resulted in a decreased hatchability. It was also shown by these workers that the level of vitamin D supplied to dams had a direct influence on both body weight and calcification of the bones of progeny that received no vitamin D to four weeks of age. Jensen (1953) found that vitamin E was necessary for high hatchability of turkey eggs. This was substantiated by Adkinson *et al.* (1955). A 36-percent increase in hatchability was produced by the addition of D-alpha-tocopheryl acetate to an all-vegetable protein diet fed Beltsville Small White turkey hens. Scott *et al.* (1955) found that supplements of 5 or 10 mg. of D-alpha-tocopheryl acetate per pound of feed would improve the hatchability in both a practical and semipurified diet. Each of these three reports indicated that natural feed ingredients may not contain sufficient vitamin E for the maintenance of high hatchability of turkey eggs. Jensen and McGinnis (1956) found that between 12 and 24 I.U. of vitamin E per pound of feed were required as a supplement to normal feed ingredients in order to maintain adequate hatchability during an 18-week study.

These investigations have been conducted with various types of turkeys and under varied experimental conditions. Ingredients known as unidentified factor sources have been included in many combinations in an attempt to improve the hatchability of turkey eggs. These substances include such feedstuffs as condensed fish solubles, alfalfa meal, dried brewers yeast, dried whey, grass juice and distillers dried solubles.

This bulletin presents a review of the work done in turkey breeder hen nutrition at the Texas Agricultural Experiment Station since 1950. These investigations include tests with Broad Breasted Bronze and Beltsville Small White turkeys reared to maturity on the range or on wire floors. Various combinations of unknown hatchability factor sources, vitamins, antibiotics and antioxidants have been fed in an attempt to improve the reproductive performance of the turkey breeder hen.

PROCEDURE

The turkeys used in the experiments being reported were from Broad Breasted Bronze or Beltsville Small White stock of the Department of Poultry Science and the Texas Agricultural Experiment Station. All experiments were conducted at the Texas A. and M. College Poultry Farm, College Station, Texas.

All Broad Breasted Bronze experiments were conducted with groups of 12 to 17 birds per group, maintained in pens with wooden or concrete floors covered with 2 to 4 inches of cane pulp litter. The Beltsville Small White turkeys were reared to maturity in batteries and on wire floors to prevent access to droppings. Some experiments with Beltsville Small White hens were conducted on

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wire floors, and in some tests birds were kept in individual cages throughout the laying period. Feed and water were supplied *ad libitum*. Artificial insemination with pooled semen was used in the cage studies. Natural matings or a combination of natural and artificial matings were used in the other experiments.

Eggs were gathered daily, stored in a cooler at 55° F. and set at weekly intervals. Eggs were candled at 7, 14, 21 and 24 days of incubation. All eggs removed in candling were broken and the time of embryonic death was estimated. Embryos removed in candling and those failing to hatch were examined for gross changes in appearance. In some of the studies, live embryos were removed and preserved in an appropriate solution for microscopic studies of the tissues. Eggs were analyzed for tocopherol content at various stages of the experimental periods by Distillation Products Industries, Rochester, New York.

Specific alterations in these above procedures are indicated in the discussion of individual experiments or treatments. Since D-alpha-tocopheryl acetate is a compound used in feeds to provide vitamin E activity, this compound often is used interchangeably with vitamin E in the discussion which follows. Substitutions of the supplements to the all-vegetable-protein basal diet were made at the expense of corn or milo and soybean oil meal in order to maintain the same calculated protein level within any given experiment. Hatchability data in all tables refer to percentage hatch of fertile eggs.

RESULTS AND DISCUSSION

Hatchability

Experiment 1. Broad Breasted Bronze turkeys were reared to maturity in batteries on an all-vegetable protein diet (Atkinson and Couch, 1951 a,b). At 24 weeks, the hens were divided into groups of four birds each and placed in individual laying cages. One group received the basal (all-vegetable protein) diet unsupplemented. Supplements of 25 mg. aureomycin per kilogram, 66 mg. streptomycin per kilogram plus 13 mcg. vitamin B_{12} per kilogram, six percent fish meal, 2 percent aureomycin APF or 4 percent Liver "L" respectively, were added to the basal (all-vegetable protein) diet and fed to various groups of hens. One group of hens received weekly injections of vitamin B_{12} (10 mcg. per bird per week). The vitamin B_{12} was supplied in a crude form since it was not commercially available in pure quantities at the time of this experiment. The aureomycin APF was a combination of aureomycin and the Animal Protein Factor (APF) which was the term applied to vitamin B_{12} at this time. Egg production was increased considerably by supplementing the basal ration with aureomycin (25 mg. per kg.) or liver "L" (4 percent). Hatchability was very low (16 percent) in the basal group, and was improved considerably by various supplements as follows: aureomycin (25 mg. per kg.) 43.5 percent, vitamin B_{12} injected (10 mcg. per bird per week) 53.8 percent, fish meal (6 percent) 41.7; and liver "L" (4 percent) 66.1 percent.

Experiment 2. Atkinson *et al.* (1953) reported a definite improvement in hatchability of eggs from Beltsville Small White turkey hens which had been reared to maturity on wire and fed an all-vegetable protein diet, by the addition of 5 percent Biopar C to the basal ration. Beltsville Small White turkeys were maintained on wire from hatching to maturity. At maturity 60 hens were placed in a laying house with a wire floor and were mated with Beltsville Small White toms which had been reared on range. Egg production was normal and fertility averaged approximately 90 per cent. Hatchability was poor (40 percent) and steadily decreased to approximately 10 percent by the eighth week.

After 10 weeks the house was divided into two pens of 30 birds each and 5 percent Biopar was substituted for 5 percent milo in the ration of one pen. Egg production remained normal and fertility was maintained at a high level (90 to 95 percent). Hatchability remained low (10 to 20 percent) for the basal group throughout the entire experimental period. The addition of Biopar C (5 percent) to the basal diet caused a significant and immediate improvement in hatchability (50 to 60 percent) which was maintained for approximately 8 weeks after which time a steady decrease was noted.

A number of embryos from the basal group which died between the 24th and 28th day of incubation or were alive but unhatched on the 28th day had shortened legs, hemorrhages under the skin and a bulging eye condition in which the lens and/or cornea appeared cloudy. These conditions were thought to be due to a deficiency of unknown hatchability factors of which Biopar C appeared to be a fairly good source.

Experiment 3. Couch *et al.* (1954) conducted a study to determine the value of distillers dried solubles and/or fish meal as supplements to a turkey breeder ration. Apparently there was no beneficial effect on hatchability or fertility when the hens were fed either fish meal or distillers dried solubles singly or in combination as supplements to the basal ration. Fertility remained at a fairly high level (65 to 90 percent) in all groups throughout the 18-week experimental period. Hatchability declined to levels of 0 to 10 percent after the 12th week of the experimental period. Egg production was slightly improved when both fish meal and distillers dried solubles were included in the diet but was not maintained at a desirable level in any of the experimental groups. This study shows clearly the need for more information regarding the nutrient requirements of the turkey breeder hen.

Experiment 4. The effect of adding dehydrated alfalfa meal, condensed fish solubles, dried whey and distillers dried solubles to the basal diet

TABLE 1. BASAL DIET

Ingredient[1]	Percent
Ground yellow corn	35
Ground milo	34
Soybean oil meal	25
Dicalcium phosphate	3
Oyster shell flour	2.5
Salt	.5

[1]Supplements added per lb. of diet: 160 mg. $MnSO_4$ (Technical grade), 4,500 I.U. vitamin A, 1,200 I.C.U. vitamin D_3, 2 mg. riboflavin, 12.5 mg. calcium pantothenate, 20 mg. niacin, 400 mg. choline chloride, 6 mcg. vitamin B_{12}.

(Table 1) on fertility, hatchability and tocopherol content of yolks of Broad Breasted Bronze turkey eggs is shown in Table 2 (Atkinson *et al.* 1955).

The hatchability of fertile eggs from these groups of Broad Breasted Bronze turkey hens ranged from 76.0 to 85.9 percent during the first 11 weeks of the 17-week experiment and was not changed appreciably by the addition of dehydrated alfalfa meal, condensed fish solubles, dried whey, distillers dried solubles or combinations of these supplements to the basal diet (Table 1). The basal diet included 2.5 mg. bacitracin, 1 mg. penicillin and 2.5 mg. terramycin per pound of diet.

Fertility for the first 11 weeks apparently was not affected by dietary supplements (Table 2). There was a rather low fertility (47.5 percent) in the group fed alfalfa meal, dried whey and fish solubles (Table 5). There was no apparent explanation for this low fertility. During the 12th through the 17th weeks of the experimental period, a 23.3 percent increase in hatchability of fertile eggs over that of the basal group was obtained when dehydrated alfalfa meal was added to the basal diet. A combination of dehydrated alfalfa meal and condensed fish solubles added to the basal ration improved hatchability 50 percent above that obtained with the basal group during this period. A 9.7 percent increase was obtained with distillers dried solubles and dehydrated alfalfa meal added as supplements to the basal ration over that obtained with feeding alfalfa meal alone. In each instance the combination of condensed fish solubles with one of the other unidentified factor sources produces an increase in the hatchability of fertile eggs (Table 2), above that obtained with the unidentified factor source alone.

The fertility of eggs from hens fed the basal diet was only 48.6 percent during the final 6-week period of the experiment (Table 2). In the remaining groups, except for group 5 as previously mentioned, the fertility was greater in the groups fed condensed fish solubles and dehydrated alfalfa than in the groups fed dried whey and distillers dried solubles supplemented with alfalfa meal.

The various supplements which were added to the diets of the hens had very little influence on embryonic mortality during the first 11 weeks of the experimental period. However, embryonic mortality during the first week of incubation in the unsupplemented group increased from 6.3 percent during the first 11 weeks to 44.8 percent (Table 3) during the last 6 weeks. An increase in embryonic mortality (8.5 percent to 20.7 percent) was observed in the basal group during the last 4 days of the incubation period, between the 12th and 17th week of the experimental period. The addition of the unidentified factor sources to the diet of the hens in groups 2 through 8 decreased embryonic mortality during the experimental period (Table 3).

Embryonic mortality during the 24th to the 28th day of incubation for the last 6 weeks of the

TABLE 2. EFFECT OF ALFALFA, FISH SOLUBLES, DRIED WHEY AND DISTILLERS DRIED SOLUBLES ON FERTILITY, HATCHABILITY AND TOCOPHEROL CONTENT OF YOLKS OF BROAD BREASTED BRONZE TURKEY EGGS

Group	Supplements to basal diet[1]	Weeks				
		0-11		12-17		Tocopherol/egg yolk mcg.[2]
		— — — Percent — — —				
		Fertility	Hatchability	Fertility	Hatchability	
1	None	79.4	83.7	48.6	31.0	590
2	5% DAM	72.5	80.3	83.6	54.3	670
3	5% DAM + 5% CFS	74.3	76.0	90.9	81.0	486
4	5% DAM + 3% DW	82.6	82.1	69.5	58.4	676
5	5% DAM + 3% DW + 5% CFS	47.5	80.1	44.0	64.5	349
6	5% DAM + 5% DDS	76.7	85.9	68.8	64.0	1,073
7	5% DAM + 5% DDS + CFS	87.6	83.0	83.5	76.5	781
8	5% DAM + 5% CFS + 3% DW + 5% DDS	81.6	82.4	79.2	75.0	699

[1]DAM = dehydrated alfalfa meal; CFS = condensed fish solubles; DW = dried whey; DDS = distillers dried solubles.
[2]Tocopherol content of egg yolks collected at the end of the 17th week.

TABLE 3. EFFECT OF ALFALFA, FISH SOLUBLES, DRIED WHEY AND DISTILLERS DRIED SOLUBLES ON EMBRYONIC MORTALITY IN BROAD BREASTED BRONZE TURKEY EGGS DURING THE 12TH THROUGH THE 17TH WEEK OF EXPERIMENT

Group	Supplement to basal diet[1]	Time of embryonic deaths	
		0-7 days	24-28 days
		Mortality rate, percent	
1	None	44.8	20.7
2	5% DAM	14.2	30.7
3	5% DAM + 5% CFS	13.8	5.9
4	5% DAM + 3% DW	26.9	12.4
5	5% DAM + 3% DW + 5% CFS	20.9	11.3
6	5% DAM + 5% DDS	19.8	14.0
7	5% DAM + 5% DDS + 5% CFS	12.3	9.8
8	5% DAM + 5% CFS + 3% DW + 5% DDS	9.5	15.5

[1]DAM = dehydrated alfalfa meal; CFS = condensed fish solubles; DW = dried whey; DDS = distillers dried solubles.

TABLE 4. EFFECT OF D-ALPHA-TOCOPHERYL ACETATE AND UNIDENTIFIED FACTOR SOURCES ON EGG PRODUCTION, FERTILITY, HATCHABILITY AND TOCOPHEROL CONTENT OF EGG YOLKS OF BELTSVILLE SMALL WHITE TURKEY EGGS[1]

Group	Supplement to basal diet	Production, %	Eggs set	Fertility, %	Hatch fertile eggs, %	Tocopherol/egg yolk, μg.
1	None	48.3	202	98.5	54.3	195
2	D-alpha-tocopheryl acetate (20 mg./lb.)	34.9	198	98.5	88.2	823
3	Fish solubles (5%)	38.6	187	65.8	47.2	273
4	Fish solubles (5%) + D-alpha-tocopheryl acetate (20 mg./lb.)	37.0	198	96.0	93.2	736
5	Dried whey (3%)	33.7	159	98.7	51.6	156
6	Dried whey (3%) + D-alpha-tocopheryl acetate (20 mg./lb.)	39.7	167	92.8	85.8	922
7	Dried whey (3%) + fish solubles (5%)	44.4	174	99.4	52.0	174
8	Dried whey (3%) + fish solubles (5%) + D-alpha-tocopheryl acetate (20 mg./lb.)	33.7	172	97.7	83.9	870
	Effect of supplement other than D-alpha-tocopheryl acetate					
1,2	None	41.6	400	98.5	71.1	
3,4	Fish solubles (5%)	37.8	385	81.3	75.1	
5,6	Dried whey (3%)	35.4	326	95.7	68.6	
7,8	Fish solubles (5%) + dried whey (3%)	39.1	346	98.6	67.7	
	Effect of D-alpha-tocopheryl acetate					
1,3,5,7	None	41.3	722	90.3	51.7	200
2,4,6,8	D-alpha-tocopheryl acetate (20 mg./lb.)	36.3	735	96.3	88.0	838

[1]Data from 1st to 9th week, inclusive. Tocopherol content is of eggs collected during the ninth week.

test varied from 5.9 to 30.7 percent (Table 3). The embryonic mortality in group 2, fed 5 percent dehydrated alfalfa meal only, was greater than the basal group during this part of the incubation period. When alfalfa was combined with condensed fish solubles, the embryonic mortality was reduced to 5.9 percent.

Jacobs *et al.* (1953) reported that dehydrated alfalfa meal increased the hatchability and fertility of eggs from chicken hens, when fed in the presence of vitamin B_{12}. These data suggest that dehydrated alfalfa meal and condensed fish solubles contain greater amounts of the unidentified factors necessary to maintain normal turkey fertility and hatchability after 11 weeks of production than distillers dried solubles or dried whey.

Experiment 5. Supplements of D-alpha-tocopheryl acetate, condensed fish solubles and dried whey were added to the basal diet (Table 1) of Beltsville Small White turkey hens which had been reared to maturity on wire floors (Atkinson *et al.* 1955). Vitamin A was increased to 9,000 I. U. per pound at the start of the 17-week experimental period and menadione was added at a level of 2 mg. per pound. D-alpha-tocopheryl acetate additions to the rations were reversed after 9 weeks of the test period (Tables 4 and 5).

The addition of 20 mg. of D-alpha-tocopheryl acetate per pound of feed increased hatchability approximately 36 percent the first 9 weeks (Table 4). When the hatchability data (Table 4) were treated statistically by analysis of variance, a significant difference was found to exist between treatments. An F value of 13.82 was obtained for treatments whereas only 3.02 was needed for

TABLE 5. EFFECT OF D-ALPHA-TOCOPHERYL ACETATE AND UNIDENTIFIED FACTOR SOURCES ON EGG PRODUCTION, FERTILITY, HATCHABILITY AND TOCOPHEROL CONTENT OF EGG YOLKS OF BELTSVILLE SMALL WHITE TURKEY EGGS[1]

Group	Supplement to basal diet	Production, %	Eggs set	Fertility, %	Hatch fertile eggs, %	Tocopherol/egg yolk, μg.
1	D-alpha-tocopheryl acetate (20 mg./lb.)	39.5	164	89.6	63.3	590
2	None	31.1	108	99.1	79.4	208
3	Fish solubles (5%) + D-alpha-tocopheryl acetate (20 mg./lb.)	28.2	112	96.4	72.2	777
4	Fish solubles (5%)	24.7	82	98.8	80.2	265
5	Dried whey (3%) + D-alpha-tocopheryl acetate (20 mg./lb.)	28.7	102	37.3	65.8	980
6	Dried whey (3%)	37.7	145	55.9	71.6	187
7	Dried whey (3%) + fish solubles (5%) + D-alpha-tocopheryl acetate (20 mg./lb.)	27.8	89	74.2	75.8	642
8	Dried whey (3%) + fish solubles (5%)	28.7	123	44.7	85.5	200
	Effect of supplement other than D-alpha-tocopheryl acetate					
1,2	None	35.3	272	93.4	70.1	
3,4	Fish solubles (5%)	26.5	194	97.4	75.7	
5,6	Dried whey (3%)	33.3	247	48.2	69.7	
7,8	Fish solubles (5%) + dried whey (3%)	28.3	212	57.1	80.2	
	Effect of D-alpha-tocopheryl acetate					
1,3,5,7	D-alpha-tocopheryl acetate (20 mg./lb.)	31.1	467	76.9	68.5	747
2,4,6,8	None	30.6	458	70.7	78.7	215

[1]Data from 10th to 17th week, inclusive. Tocopherol content is of eggs collected during the 17th week.

significance at the 0.01 level of probability. On further analysis an F value of 90.35 was obtained for vitamin E versus no vitamin E, whereas, only 7.17 was needed for significance at the 0.01 level of probability. This indicated that under the conditions of the experiment vitamin E was required to maintain an optimum level of hatchability.

Hatchability of eggs from groups not fed supplemental vitamin E during the first 9 weeks (groups 1, 3, 5 and 7) was increased approximately 17 percent by supplementation of the diet with D-alpha-tocopheryl acetate at the beginning of the 10th week of the experimental period (Table 5).

The average hatchability of eggs from groups 2, 4, 6 and 8 was 88 percent (Table 4) for the first 9-week period, during which time all birds in these groups received a supplement of 20 mg. of D-alpha-tocopheryl acetate per pound. Removal of the D-alpha-tocopheryl acetate from the diet at the end of the 9th week caused the hatchability to decrease from 88 to 78.7 percent (Tables 4 and 5).

All embryos found dead during the incubation period were examined in order to determine time of death and to check for any abnormalities which might have occurred. There were 309 dead embryos out of 652 fertile eggs during the first 9-week period from hens that did not receive D-alpha-tocopheryl acetate; however, there were only 83 dead embryos out of 708 fertile eggs from hens fed D-alpha-tocopheryl acetate. A large number of embryos from the unsupplimented groups died between the 24th and 28th days of incubation. This was not true for those groups supplemented with D-alpha-tocopheryl acetate. All deficient embryos dying after the 17th day of incubation were found to be smaller than normal as determined by body measurement. Most of the deficient embryos which died during incubation appeared to be blind and were found to have a cloudy lens or a cloudy spot under the cornea. Some of the embryos were found to have both of these conditions.

Failure to obtain a rapid reversal of hatchability upon reversal of the vitamin E treatments (Table 5) indicates that the turkey hen may be able to store vitamin E in the body tissues to a greater extent than was indicated by the report of Jensen (1953). He reported that the hatchability of eggs from birds fed D-alpha-tocopheryl acetate was reduced from 80 to 45 percent by the removal of the supplemental vitamin E from the diet for 3 weeks.

Within 2 weeks after the diets were reversed, the above-mentioned abnormalities ceased to occur and none were found in any group during the last 6 weeks of the 17-week experimental period. This would also indicate that there was considerable storage of the vitamin in the tissues of the birds.

It is apparent from Table 4 that the addition of D-alpha-tocopheryl acetate to the diet of turkey hens had increased the tocopherol content of egg yolks by the end of the first 9 weeks of the experiment. Reversal of the diets at the beginning of the 10th week resulted in a reversal, also, of the tocopherol content of the egg yolks, at the end of the 17th week (Table 5) of the experimental period. Such a decrease in the tocopherol content of the egg yolk did not result in as low a percentage hatchability in eggs from groups 2, 4, 6 and 8 as had been observed earlier in eggs from groups 1, 3, 5 and 7, even during the final week of the experiment. No explanation is immediately apparent for the hatchability remaining at a higher level in groups 2, 4, 6 and 8 since the tocopherol content of the eggs had decreased to a point where much lower hatchability might have been expected. The average hatchability of eggs from groups 2, 4, 6 and 8 was 78.7 percent from the 10th to the 17th week of the test, while it was 51.7 percent for eggs from the same birds during the 9 weeks prior to reversal of the diets. It is possible that unidentified factors, independent of or interacting with alpha-tocopheryl acetate, may have caused the hens to be depleted more slowly of tocopherol stores.

Late in the season 200 Broad Breasted Bronze turkey eggs were obtained from a commercial turkey breeder who was feeding a commercial turkey breeder mash and had experienced a considerable drop in both fertility and hatchability. These eggs were incubated in our laboratory. Only 30 percent of the 200 eggs were found to be fertile, and only 41.7 percent of the fertile eggs hatched. Eight of the embryos which died between the 24th and 28th day of incubation were found to have a cloudy lens, were smaller in size and were quite similar in appearance to embryos obtained from hens in experiment 5 that had not received supplemental D-alpha-tocopheryl acetate.

Experiment 6. Broad Breasted Bronze turkeys were fed the basal diet (Table 1) unsupplemented and supplemented with Vitamin E (D-alpha-tocopheryl acetate), dehydrated alfalfa meal, condensed fish solubles and combinations of these substances, (Table 6) (Ferguson *et al.* 1956). The basal ration used was the same as shown in Table 1 except that an additional 4,500 I. U. of vitamin A were added per pound of ration. These hens were reared on range and had been fed a practical type ration containing sources of unidentified factors. The hens were maintained on litter during the experimental period, rather than on wire floors, in order to determine the effectiveness of feeding supplements of unidentified factor sources and vitamin E to turkey hens maintained under practical conditions.

The addition of Vitamin E, dehydrated alfalfa meal and condensed fish solubles to the basal diet resulted in hatchability varying from 71 to 79 percent during the first 8 weeks of the test (Table

TABLE 6. HATCHABILITY OF FERTILE EGGS AND TOCOPHEROL CONTENT OF EGGS FROM BROAD BREASTED BRONZE TURKEY HENS

Group	Supplements to basal diet[1]	Hatchability (%)		Tocopherol (mcg./yolk)[2]		Embryonic mortality[3]
		1-8 wks.	9-20 wks.	13 wks.	21 wks.	
1	None	74	56	430	540	24.3
2	Vitamin E	77	60	1,820	920	30.4
3	DAM	71	60	520	370	26.3
4	DAM + vitamin E	79	68	630	1,250	21.6
5	CFS	76	68	520	840	19.9
6	CFS + vitamin E	78	68	1,170	1,100	18.8
7	CFS + DAM	74	68	690	580	18.5
8	CFS + DAM + vitamin E	76	77	1,180	1,050	8.0

[1]Vitamin E = D-alpha-tocopheryl acetate, 20 mg./lb.; DAM = dehydrated alfalfa meal 5%; CFS = condensed fish solubles 5%.
[2]Pre-experimental tocopherol levels: 471 mcg./yolk.
[3]Fourth week embryonic mortality in eggs laid during weeks 9th to 20th.

6). In each instance where vitamin E was added (groups 2, 4, 6 and 8) hatchability was increased 2 to 8 percent over that observed in groups 1, 3, 5 and 7. The latter groups represent the groups fed unidentified factor sources without supplemental vitamin E.

The hatchability of eggs during the last 12 weeks of the experiment was improved from 4 to 12 percent by the addition of either vitamin E, dehydrated alfalfa leaf meal or condensed fish solubles, fed singly or in combination of any two of these supplements. However, when all three of the supplements were fed (group 8), 77 percent hatchability was maintained which represented an increase of 21 percent over that of the unsupplemented group (group 1). The hens in this group (group 8) were the only ones which did not show a decrease in hatchability between the 9th and the 20th week of the experimental period. Analysis of the data for the 9 to 20-week period showed that the addition of vitamin E (D-alpha-tocopheryl acetate) to the diet caused an increased hatchability which was statistically significant at the 5 percent level of probability. The feeding of condensed fish solubles produced an increase in hatchability which was significant at the 1 percent level of probability. This increased hatchability due to vitamin E is in agreement with reports by Jensen (1953), Atkinson *et al.* (1955) and Ferguson *et al.* (1954).

Embryonic mortality was essentially the same for all groups during the first 8 weeks. There was also very little difference in the embryonic mortality between groups during the first 3 weeks of the incubation period for eggs laid during the 9th through the 20th week. Only 8 percent embryonic mortality was found during the 4th week of incubation of eggs from the hens in group 8 which had been fed supplements of alfalfa meal, fish solubles and vitamin E (D-alpha-tocopheryl actate) (Table 6), while the embryonic mortality ranged from 18.5 to 30.4 percent for the remaining groups during this same period.

Experiment 7. Feldman *et al.* (1957a) reported results of supplementing an all-vegetable protein ration with dehydrated alfalfa meal, dried brewers yeast and condensed fish solubles. Range-reared Broad Breasted Bronze turkeys were used in this study, and the hens were maintained on litter during the laying season. Vitamin E (20 mg. per pound), chlortetracycline (20 mg. per pound), penicillin (5 mg. per pound) and 2-methyl naphtho quinone (2 mg. per pound) were also added to the basal diet shown in Table 1. In addition, the levels of dicalcium phosphate and oyster shell flour were changed so that each was added at a level of 3.5 percent. The additional dicalcium phosphate and oyster shell flour were added at the expense of corn.

TABLE 7. EFFECT OF DEHYDRATED ALFALFA MEAL, CONDENSED FISH SOLUBLES AND DRIED BREWERS YEAST ON THE HATCHABILITY OF BROAD BREASTED BRONZE TURKEY EGGS

Group	Supplement to basal diet	0-11 weeks		12-17 weeks		12-24 weeks	
		Total eggs set	% hatchability[1]	Total eggs set	% hatchability[1]	Total eggs set	% hatchability[1]
1	None	428	75.0	182	24.8	352	36.9
2	5% dehydrated alfalfa meal	525	82.9	161	62.2[3]	311	55.9
3	10% dehydrated alfalfa meal	562	77.5	167	63.0[3]	284	67.0[3]
4	5% condensed fish solubles	478	75.9	120	73.5[3]	246	56.8
5	5% condensed fish solubles + 5% dehydrated alfalfa meal	673	83.8	208	78.0[3]	378	73.3[3]
6	5% condensed fish solubles + 10% dehydrated alfalfa meal	558	82.9	194	71.2[3]	300	62.4[2]
7	5% dried brewers yeast	593	74.4	200	64.1[3]	383	57.9
8	5% dried brewers yeast + 5% condensed fish solubles	593	84.6	254	78.6[3]	462	74.5[3]

[1]Percent of fertile eggs.
[2]Significant at .05 level.
[3]Significant at .01 level.

The hatchability of fertile eggs from Broad Breasted Bronze turkey hens varied from 74.4 percent to 84.6 percent during the first 11 weeks of the 24-week study. The hatchability of fertile eggs from hens fed diets supplemented with condensed fish solubles in combination with either dehydrated alfalfa leaf meal or dried brewers yeast was observed to be slightly higher than that of hens fed the unsupplemented basal ration or of hens fed rations supplemented with only dehydrated alfalfa meal or dried brewers yeast (Table 7).

The hatchability of fertile eggs from dams fed the unsupplemented basal diet (group 1) exhibit-

ed a decided drop which was evident by the end of the 12th week. The hatchability for this group averaged only 24.8 percent between the 12th and 17th week of the study. The addition of dehydrated alfalfa meal, condensed fish solubles or dried brewers yeast to the basal ration or condensed fish solubles fed in combination with dehydrated alfalfa meal or dried brewers yeast produced a significant increase in the hatchability of fertile eggs during this same period. Dehydrated alfalfa meal was the only supplement which was fed at more than one level. A 10-percent level of this supplement failed to produce any further increase in the hatchability of fertile eggs over the 5-percent level from the 12th through the 17th week.

There was an indication from the data in Table 7 that a depletion of the hatchability factor(s) occurred between the 12th and 17th weeks in groups fed supplements of 5 or 10 percent dehydrated alfalfa meal or 5 percent dried brewers yeast.

The highest hatchability of fertile eggs was obtained by supplementation of the basal diet with combinations of 5 percent condensed fish solubles and 5 percent dehydrated alfalfa meal (group 5) or with 5 percent condensed fish solubles and 5 percent dried brewers yeast (group 8). These differences were not found to be statistically significant.

The hatchability of fertile eggs from Broad Breasted Bronze turkey hens fed diets containing 5 percent dehydrated alfalfa meal (group 2), 5 percent condensed fish solubles (group 4) or 5 percent dried brewers yeast (group 7) decreased still further as shown in Table 7 for the period beginning with the 12th week and ending with the 24th week. Dehydrated alfalfa meal (group 3) at the 10-percent level, on the other hand, was able to maintain hatchability at a satisfactory level during this same period. This would indicate that 5 percent dehydrated alfalfa meal did not supply sufficient quantities of the unidentified hatchability factor(s). Hens fed 5 percent dehydrated alfalfa meal, 5 percent condensed fish solubles and 5 percent dried brewers yeast exhibited almost identical average hatchability for the period of the 12th to the 24th week.

Highly significant increases in the hatchability of fertile eggs were obtained in groups 5 and 8. These hens were fed combinations of condensed fish solubles (5 percent) and dehydrated alfalfa meal (5 percent) and condensed fish solubles (5 percent) and dried brewers yeast (5 percent) as shown in Table 7.

Two principal peaks of embryonic mortality were observed in eggs from the hens in this study. The first occurred during the first 7 days of the incubation period and the second occurred during the last 7 days of the incubation period.

TABLE 8. EFFECT OF DEHYDRATED ALFALFA MEAL, CONDENSED FISH SOLUBLES AND DRIED BREWERS YEAST ON THE EMBRYONIC MORTALITY OF BROAD BREASTED BRONZE TURKEY EGGS

Group	Supplement to basal diet	First week incubation			Fourth week incubation		
		0-11 wks.	12-17 wks.	12-24 wks.	0-11 wks.	12-17 wks.	12-24 wks.
		— — Embryonic mortality, percent — —					
1	None	8.9	70.0	48.2	13.4	2.9	13.1
2	5% dehydrated alfalfa meal	7.0	24.8	28.9	9.2	11.4	13.3
3	10% dehydrated alfalfa meal	7.0	13.9	14.9	14.1	25.3	18.4
4	5% condensed fish solubles	10.3	19.5	35.5	8.9	6.2	6.0
5	5% condensed fish solubles + 5% dehydrated alfalfa meal	3.3	13.7	17.9	5.6	6.3	7.5
6	5% condensed fish solubles + 10% dehydrated alfalfa meal	9.5	14.1	26.5	7.3	10.8	9.0
7	5% dried brewers yeast	5.7	21.7	28.0	15.7	13.0	12.5
8	5% dried brewers yeast + 5% condensed fish solubles	5.8	13.6	15.3	7.6	7.0	8.2

Slight differences in embryonic mortality were observed (Table 8) during the first 11 weeks of the experimental period which could not be attributed to the dietary treatments. Supplementation of the basal diet with 10 percent dehydrated alfalfa meal reduced the first week embryonic mortality below that obtained on the diet supplemented with 5 percent dehydrated alfalfa meal from the 12th through the 17th week. Condensed fish solubles fed in combination with either 5 percent or 10 percent dehydrated alfalfa meal reduced the first week embryonic mortality below that of the condensed fish solubles group. The feeding of 5 percent dried brewers yeast was no more effective in reducing the mortality of embryos during this period than was 5 percent dehydrated alfalfa meal or 5 percent condensed fish solubles, but when fed in combination with 5 percent condensed fish solubles, a slight improvement was noted.

TABLE 9. TOCOPHEROL CONTENT OF EGG YOLKS FROM TURKEY HENS FED A GRAIN-TYPE OF DIET SUPPLEMENTED WITH AN ANTIOXIDANT, DIPHENYL-P-PHENYLENEDIAME (DPPD) VITAMIN E AND CONDENSED FISH SOLUBLES

Diet[1]	Weeks on diet	Tocopherol per yolk, mcg.
Pre-experimental	5	1360
Basal	10	430
Basal + E	10	925
Basal + DPPD	10	470
Basal + E + DPPD	10	990
Basal + E[2]	4	1135
Basal + E + CFS[2]	4	1360

[1]D-alpha-tocopheryl acetate (vitamin E); added at level of 20 mg./lb.; DPPD added at 0.25 lb. per ton.
[2]See text for complete explanation.

The first week embryonic mortality, during the 12 to 24 weeks of the test, was greatly reduced when the hens were fed diets containing 10 percent dehydrated alfalfa meal (group 3), 5 percent condensed fish solubles and 5 percent dehydrated alfalfa meal (group 5), or 5 percent condensed fish solubles and 5 percent dried brewers yeast (group 8).

There were practically no differences in the 4th week embryonic mortality between experimental groups. However, it should be pointed out that in this case, the groups which were fed rations containing condensed fish solubles (groups 4, 5, 6 and 8) consistently exhibited the lowest 4th week embryonic mortality throughout the entire 24 weeks of the study.

Experiment 8. Thirty-six Beltsville Small White hens from groups 2, 4, 6 and 8 of experiment 5 were maintained on the basal diet (Table 1), (Ferguson *et al.* 1954b). Experiment 5 had terminated in July when the hens stopped laying. After about 8 weeks the hens were placed under lights and mated with toms similarly treated. The first eggs were laid approximately 12 weeks after the hens had gone out of production. Analysis for tocopherol was carried out on eggs collected during the fifth week of a pre-experimental period. Hatchability of fertile eggs during this 5-week period was 0, 45, 70, 56 and 66 percent, respectively. At this time the birds were divided into four groups and fed the basal diet supplemented with 20 mg. per pound vitamin E (D-alpha-tocopheryl acetate) and an antioxidant, 0.0125 percent DPPD (N, N'-Diphenyl-p-phenylenediamine) as shown in Table 9.

Hatchability of fertile eggs from the basal group was only 25 percent for the following 10-week period. Supplementation of the basal diet with Vitamin E (D-alpha-tocopheryl acetate) increased hatchability to 50.8 percent. The addition of DPPD and a combination of DPPD plus vitamin E resulted in hatchability values of 43 percent and 36 percent, respectively.

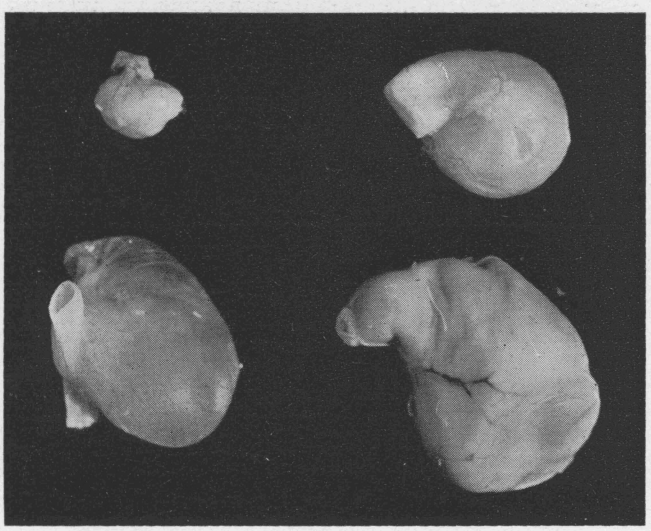

Figure 1. Normal-sized crop (top left) and gizzard (top right) from Beltsville Small White turkey, removed immediately after hatching. Enlarged thin-walled crop (bottom left) and gizzard (bottom right) from 27-day embryo. The lower pictures were made of preserved material and the gizzard is smaller than it appeared in the live embryo.

TABLE 10. EFFECT OF VITAMIN E AND DPPD SUPPLEMENTS ON HATCHABILITY AND TOCOPHEROL CONTENT OF EGGS FROM BELTSVILLE SMALL WHITE TURKEYS

Group	Supplement to basal diet	% hatchability		Tocopherol[1] mcg./yolk	
		1-5 wks.	6-18 wks.	8 wks.	16 wks.
1	None	38	48	335	250
2	Vitamin E (20 mg./lb.)	81	77	910	855
3	DPPD (1/4 lb./ton)	58	68	130	160
4	Vitamin E (20 mg./lb.) + DPPD (1/4 lb./ton)	48	77	840	550

[1]Pre-experimental values, 260 mcg./yolk.

During the 10-week experimental period, the incidence of embryonic mortality was high in all groups during the 4th week of incubation. Dead embryos from all groups were characterized by edema, frequent eye disorders, an enlarged and thin-walled crop and gizzard (Figure 1), urate deposits in the kidneys and ureters and occasionally, an enlarged heart.

After 10 weeks 26 of the birds were divided into two groups (Table 9). One group was fed the basal diet plus vitamin E, and the other group received the basal diet plus condensed fish solubles and vitamin E. The hatchability of the E supplemented group and the fish solubles plus vitamin E group average 77 and 78 percent, respectively. The incidence of embryonic abnormalities was not observed during the 4-week period except for four instances; one in group 1 out of 117 fertile eggs, and three in group 2 out of 127 fertile eggs. Analyses of eggs for tocopherol content during this experiment are shown in Table 9.

Experiment 9. Beltsville Small White turkey hens, reared to maturity on wire floors, were fed the basal diet (Table 1) in which de-germed corn (63-percent) replaced ground yellow corn and ground milo (Ferguson *et al.* 1955b). Soybean oil meal was increased to 31 percent. One group received the basal diet unsupplemented and the other groups received the basal diet supplemented as indicated in Tables 10 and 11.

Hatchability was low during the first 5 weeks in all groups except the one supplemented with vitamin E (D-alpha-tocopheryl acetate) (Table 10). Some improvement was noted in hatchability when DPPD was added, but a lowered hatchability occurred when DPPD was fed with vitamin E. During the 6th through the 18th week, the hatchability improved in all groups, with those containing vitamin E giving the best hatchability. Tocopherol values of egg yolks were not increased by the addition of DPPD or dehydrated alfalfa meal (Tables 10 and 11).

TABLE 11. EFFECT OF VITAMIN E AND DEHYDRATED ALFALFA MEAL ON HATCHABILITY AND TOCOPHEROL CONTENT OF EGGS FROM BELTSVILLE SMALL WHITE TURKEYS

Group	Supplement to basal diet	% hatchability 12 wks.	Tocopherol[1] mcg./yolk 8 wks.
1	None	40	350
2	Vitamin E (20 mg./lb.)	73	715
3	Dehydrated alfalfa meal (5%)	66	260
4	Vitamin E (20 mg./lb.) + dehydrated alfalfa meal (5%)	63	805

[1]Pre-experimental values, 260 mcg./yolk.

Hatchability of eggs after 12 weeks (Table 11) was highest when vitamin E was added. An improvement in hatchability over the basal group was also obtained when dehydrated alfalfa meal was included in the diet.

Embryonic abnormalities similar to those reported in experiment 8 occurred in all groups.

Experiment 10. Whiteside *et al.* (1957) fed Broad Breasted Bronze turkey hens the basal diet (Table 1) unsupplemented or supplemented with 5 percent dried brewers yeast (Table 12). Additional groups received, in addition to the dried brewers yeast (5 percent), either 5 percent dehydrated alfalfa meal or 5 percent condensed fish solubles. One group received both dehydrated alfalfa meal (5 percent) and condensed fish solubles (5 percent) in addition to the dried brewers yeast (5 percent). Supplements were added at the expense of grain and soybean oil meal so as to maintain the same protein level among the various groups. The basal diet also contained penicillin (5 mg. per pound), aureomycin (20 mg. ped pound), and furazolidone (4.5 mg. per pound). Vitamin E was not included as a supplement to the basal diet. All unidentified factor sources increased hatchability above that obtained when the basal diet was fed.

The addition of dried brewers yeast to an all-vegetable protein basal diet, without supplemental vitamin E (D-alpha-tocopheryl acetate), improved the hatchability of eggs by 19 percent. No improvement was obtained by adding dehydrated alfalfa meal to the basal ration supplemented with dried brewers yeast. However, when dried brewers yeast was fed in combination with condensed fish solubles or with dehydrated alfalfa meal and condensed fish solubles, further increases of 6 and 8 percent, respectively, were obtained over the group fed dried brewers yeast. Hatchability records were kept only for 15 weeks. Egg production data for the 26-week experimental period, and the amount of feed required per egg are shown in Table 13.

The number of eggs produced by all groups fed dried brewers yeast was greater than the number produced by the basal group (Table 13). From 11 to 16 more eggs per hen were produced when dried brewers yeast and other unidentified factor sources were included in the diet.

The average feed intake per hen, to produce an egg, ranged from 1.12 to 1.29 pounds. A slight reduction is noted when dried brewers yeast is added. The small increases (above group 2) noted in groups 3 and 5 may be due to the lowered energy content of the feed as a result of including alfalfa in the diet.

Vitamin E and Embryonic Eye Development

A gross and microscopic study was made of the blind turkey embryos observed in experiment 5 (Ferguson *et al.* 1954). A cloudiness in the central portion of the lens was found among the embryos from hens fed the all-vegetable protein basal ration without added vitamin E. This condition was observed in both eyes of 13 out of 30 live embryos examined on the 29th day of incubation. These embryos pipped the shell but failed to hatch. Seven of these had hemorrhages in the vitreous humor. One or both of the eyes frequently had a bulging of the cornea, giving the embryo a protruding eye appearance in extreme cases (Figure 2). The cornea was found to be slightly irregular in some embryos when examined with a binocular microscope. A yellowish-white spot of material was seen in the eyes, between the lens and the cornea, in a few specimens. This condition also occurred at later stages of embryonic development. An irregular-shaped iris was often associated with the bulging cornea, which made the pupil likewise irregular. Upon

TABLE 12. EFFECT OF DRIED BREWERS YEAST, ALFALFA AND CONDENSED FISH SOLUBLES ON TURKEY HATCHABILITY FROM THE 3 TO 15-WEEK PERIOD

Group	Supplement	% hatchability, 3-15 wks.	Hatchability % increase over basal, 3-15 wks.
1	Basal	49	
2	+ DBY	68	+19
3	+ DBY + Alfalfa	69	+20
4	+ DBY + CFS	74	+25
5	+ DBY + Alfalfa + CFS	76	+27

TABLE 13. EFFECT OF DRIED BREWERS YEAST, DEHYDRATED ALFALFA MEAL AND CONDENSED FISH SOLUBLES ON THE EGG PRODUCTION OF BROAD BREASTED BRONZE TURKEY HENS FOR A PERIOD OF 26 WEEKS

Group	Supplement to basal diet	Total eggs 26 wks.	Increase over basal	Eggs/hen	Lb. feed/egg
1	Basal	918		61	1.21
2	+ DBY	1082	164	72	1.11
3	+ DBY + alfalfa	1154	236	77	1.16
4	+ DBY + CFS	1124	206	75	1.13
5	+ DBY + alfalfa + CFS	1075	157	72	1.29

Figure 2. Turkey embryos, 27 days. Embryo with bulging eyes (left) from hen on E-deficient diet. Normal embryo (right) from hen fed a diet supplemented with vitamin E.

dissection of the lens, pressure applied firmly to the outside resulted in its rupture with a splitting out of a smaller miniature lens and the loss of a watery fluid. In some instances, a still smaller lens-like structure could be forced out. The eyes of the embryos obtained from hens supplemented with vitamin E apparently were normal.

The results of a study of microscopic sections of the lens from each of the two groups of embryos are shown by camera lucida drawings in Figure 3. Figure 3A is a section near the center of the lens of a 29-day embryo from a hen that was fed added vitamin E. A portion of the iris and ciliary body are included. Figure 3B is a similar section from near the center of the lens of an embryo of the same age where the hen did not receive supplemental vitamin E. The lens is apparently the same diameter as 3A, but is flatter and has a small lens body inside the outer lens layer. Degenerative changes have occurred throughout the lens, especially in the central and posterior portions.

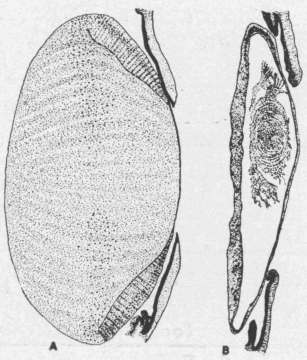

Figure 3. Drawings of microscopic sections through the eye of the turkey embryo. Normal lens is shown in A. Degenerative changes in the lens are shown in B.

Measurements of the principal skeletal parts of live embryos from the supplemented and unsupplemented groups were made. These embryos were 29 days of age, and had pipped the shell but did not hatch. The legs, wings and body length of the embryos from the groups not supplied additional D-alpha-tocopheryl acetate were less than the supplemented groups. Ten of the 30 live embryos examined from the unsupplemented groups on the 29th day of the incubation period also had an edematous area on the neck at the back of the head. This condition did not occur in the D-alpha-tocopheryl supplemented groups.

Experiment 11. A detailed pathological study of the cataract found in turkey embryos obtained from hens fed a synthetic type diet deficient in vitamin E was reported by Ferguson *et al.* (1956).

Beltsville Small White turkey hens were divided into two groups, placed in individual coops and fed the synthetic basal diet (Table 14). Vitaman E (D-alpha-tocopheryl acetate), 20 mg. per pound, was provided as a supplement to one group. After nine and one-half weeks on the test, the vitamin E (D-alpha-tocopheryl acetate) level was increased to 40 mg. per pound.

Turkey embryos from the basal group were removed daily from the 5th through the 28th day of incubation. Only living embryos were removed and preserved for histological study. Changes were noted in the external appearance, lens, lens epithelium and cornea.

These embryos showed a keratoconus-like change (a bulging of the eyes) at about 19 days of incubation, which increased as the embryo aged. Hemorrhage within the eye was observed in two of the deficient embryos. Opacities varied in size in the deficient embryos; however, the two

TABLE 14. SYNTHETIC BASAL DIET

Ingredient[1]	Percent
Sucrose	62
Drackett C-1 assay protein	24
Fresh lard	3
Salts mixture[2]	11

[1]The following ingredients were added per kg. of diet: 20 mg. thiamin, 6 mg. riboflavin, 15 mg. calcium pantothenate, 100 mg. niacin, 4 mg. pyridoxine, 20 mg. para-aminobenzoic acid, 3,800 I.C.U. vitamin D_3, 10,000 I.U. vitamin A, 33 mg. penicillin, 1,000 mg. inositol, 0.2 mg. biotin, 2 mg. folic acid, 0.5 mg. menadione, 2,000 mg. choline chloride, 7.5 gm. methionine, 4 gm. glycine, 30 mcg. vitamin B_{12}.

[2]The salts mixture supplied the following per kg. diet: 288 mg. manganese, 30 mg. iodine, 559 mg. magnesium, 246 mg. iron, 4.3 mg. copper, 6.7 mg. zinc, 0.11 mg. cobalt. The salts mixture supplied 2.5% calcium, 1.0% phosphorus, 0.5% NaCl and 0.2% potassium.

TABLE 15. INCIDENCE OF EYE DISORDERS IN BELTSVILLE SMALL WHITE TURKEY EMBRYOS FED A PURIFIED SYNTHETIC TYPE DIET

Number	Diet	Opacity in lens	Keratoconus
54	−E	21 (38.8%)	9 (17%)
55	+E	2 (3.6%)	0

eyes were usually similar in the same bird. There were opacities in one or both eyes of 21 out of 54 vitamin E deficient embryos (Table 15). Only 2 embryos with opacities were observed in 55 turkey embryos from the vitamin E supplemented group.

The most conspicuous change present in the eyes of these embryos occurred in the lens, characterized by a liquefaction of either a part or essentially all of the lens protein. The earliest changes noted were small focal areas of liquefaction located either in the center or at the periphery of the lens immediately beneath the capsule. Preliminary studies also showed degenerative changes in tissues other than the eye, which are now under investigation.

Artificial insemination was used to obtain fertile eggs. Hens were inseminated weekly with .05 ml. of pooled semen which had been diluted 1:1 or 1:2 with 0.85 percent saline solution. The average fertility of eggs as a result of this method was 90 percent over a 15-week period. The fertility of eggs from hens provided vitamin E was 95 percent, and 85 percent fertility was obtained in the group not provided vitamin E (Table 16).

Hatchability of eggs from both groups (Table 16) was determined for 15 weeks. In the group receiving the basal synthetic diet, without vitamin E, a 21 percent hatchability of fertile eggs was obtained. In the supplemented group, hatchability of eggs was 55 percent. It should be pointed out that this synthetic diet did not contain unidentified hatchability factor sources. Tocopherol values of eggs from these hens are given in Table 17.

Vitamin E Analyses of Eggs

The importance of vitamin E as a factor in increasing hatchability has been demonstrated in many of these experiments. Analyses of the eggs for their alpha-tocopherol content revealed some interesting facts.

TABLE 16. HATCHABILITY AND FERTILITY OF EGGS FROM BSW HENS FED A SYNTHETIC-TYPE DIET (15 WEEKS)

Diet	No. eggs	Fertility, %	Hatchability, %
Basal	217	85	21
Basal + Vitamin E[1]	190	95	55

[1]D-alpha-tocopheryl acetate added at level of 20 mg./lb. for 9 weeks and 40 mg./lb. thereafter.

Table 4 shows that in hens not provided supplemental vitamin E, the average tocopherol content after 9 weeks was 200 mcg. per yolk, while 838 mcg. per yolk was found in eggs from hens whose diets were supplemented with vitamin E. It was also found by these analyses that the alpha-tocopherol occurs only in the yolk and not in the egg white. A reversal of the vitamin E treatments after 9 weeks (Table 5) showed that when vitamin E was removed from the diet, the tocopherol content of the egg decreased. Tables 4 and 5 also show there was no effect on the tocopherol content of the egg due to dietary supplements of fish solubles or dried whey.

In experiment 8, the hens from groups 2, 4, 6 and 8 of experiment 5 were continued on the basal diet (Table 1) for 8 weeks. When these hens were brought back into production, it was found that the tocopherol content of the first eggs averaged 1,360 mcg. per yolk (Table 9), whereas the previous average was only 215 mcg. per yolk. As egg production progressed, the values decreased to 430 and 470 mcg. per yolk in groups not provided supplemental D-alpha-tocopheryl acetate and to 925 and 990 mcg. per yolk in the E-supplemented groups, which was consistent with earlier results. These data indicate that the hen is capable of storing this vitamin during nonlaying periods and that tocopherol storage is depleted as egg production continues. During the last few weeks of the test, when all hens were getting vitamin E, egg production declined and the tocopherol content of the eggs again increased.

Analyses of eggs from hens fed various combinations of dehydrated alfalfa meal, condensed fish solubles and dried brewers yeast (Table 18) at the end of the 17-week study showed that supplemental vitamin E maintained the tocopherol content of the egg at a high level throughout the laying period. The pre-experimental level of tocopherol was 700 mcg. per yolk for the range-reared birds. In all instances, the tocopherol was increased during the laying period. The value for group 5 is within the range of normal values obtained when supplemental D-alpha-tocopheryl acetate is added to the diet at a level of 20 mg. per pound. Values for the other groups showed that there was no consistent effect on the tocopherol content of the egg due to supplements of dehydrated alfalfa meal, condensed fish solubles or dried brewers yeast.

TABLE 17. TOCOPHEROL CONTENT OF EGG YOLKS FROM HENS FED A PURIFIED SYNTHETIC-TYPE BASAL DIET

Diet	Tocopherol per yolk (mcg.)[1]		
	Pre-Exp.	5 wks.	13 wks.
Basal	260	130	245
Basal + E[1]	260	797	1300

[1]Vitamin E added at 20 mg./lb. for 9 weeks and 40 mg./lb. thereafter.

TABLE 18. TOCOPHEROL CONTENT OF EGG YOLKS

Group no.	Supplement to basal diet	Tocopherol/ yolk mcg. 17 wks.[1]
1	None	1595
2	5% dehydrated alfalfa meal	1150
3	10% dehydrated alfalfa meal	1755
4	5% condensed fish solubles	1090
5	5% condensed fish solubles + 5% dehydrated alfalfa meal	1025
6	5% condensed fish solubles + 10% dehydrated alfalfa meal	760
7	Dried brewers yeast	1075
8	5% dried brewers yeast + 5% condensed fish solubles	1265

[1]Pre-experimental values. 700 mcg./yolk.

In experiment 9 (Tables 10 and 11), the tocopherol content of the eggs was increased only when D-alpha-tocopheryl acetate was present in the diet, and no increase was noted with the addition of dehydrated alfalfa meal or DPPD.

The tocopherol content of the egg yolks was increased in experiment 6 when vitamin E (D-alpha-tocopheryl acetate) was added to the diet (Table 6). The addition of condensed fish solubles or dehydrated alfalfa leaf meal produced, essentially, no effect on the tocopherol content of the yolks. No explanation is readily apparent for the low value obtained in group 4 (13 weeks) or the slightly increased tocopherol level in group 5 (21 weeks).

In experiment 11 the hens had been reared to maturity on wire floors and had been fed an all-vegetable protein diet without supplemental vitamin E. The experimental diet was a synthetic one which contained no added vitamin E except for one group of birds. The addition of D-alpha-tocopheryl acetate to the diet increased the tocopherol content of the egg, with higher levels of the vitamin resulting in a further increase in the amount of tocopherol in the eggs (Table 17). In this instance, fertility and hatchability percentages were both increased when vitamin E was added to the synthetic diet.

FIGURE 19. EFFECT OF DEHYDRATED ALFALFA MEAL, CONDENSED FISH SOLUBLES AND DRIED BREWERS YEAST ON EGG PRODUCTION, AVERAGE DAILY FEED CONSUMPTION AND AMOUNT OF FEED CONSUMED PER EGG PRODUCED OF BROAD BREASTED BRONZE TURKEY HENS

Group	Supplementation	Eggs/ hen/ day	Feed/ bird/ day, lb.	Feed/ egg lb.
1	None	0.34	0.44	1.36
2	5% dehydrated alfalfa meal	0.33	0.45	1.42
3	10% dehydrated alfalfa meal	0.35	0.49	1.51
4	5% condensed fish solubles	0.34	0.46	1.45
5	5% condensed fish solubles + 5% dehydrated alfalfa meal	0.40	0.53	1.41
6	5% condensed fish solubles + 10% dehydrated alfalfa meal	0.33	0.52	1.66
7	5% dried brewers yeast	0.38	0.44	1.24
8	5% dried brewers yeast + 5% condensed fish solubles	0.41	0.44	1.12

The degenerative changes in the eye of the turkey embryo, obtained from hens fed diets low in vitamin E, are of interest in view of known nutritional deficiencies and their effect on cataract formation. Rats fed a riboflavin-deficient ration develop cataracts (Day *et al.* 1931). Curtis *et al.* (1932) produced cataracts in rats by feeding a tryptophane-deficient diet. These two substances are apparently the only true primary deficiencies that were previously known to result in cataract formation. Vitamin E has been related to retrolental fibroplasia, a blindness in premature human infants (Owens and Owens, 1939). An eye lesion which suggested retrolental fibroplasia was reported in rat embryos due to a deficiency of vitamin E in the diet of the mother by Callison-and-Orent-keiles (1951).

There was no consistent effect due to unidentified hatchability factor sources or to vitamin E on egg production. However, in experiment 10 there was a marked increase in egg production when dried brewers yeast was present in the diet. In experiment 7 there was a reduction in the amount of feed required to produce an egg when supplements of dried brewers yeast were added (Table 19). The feed per egg in this experiment is in close agreement with results reported in experiment 10 (Table 13), which also contained dried brewers yeast as one of the supplements to the basal diet.

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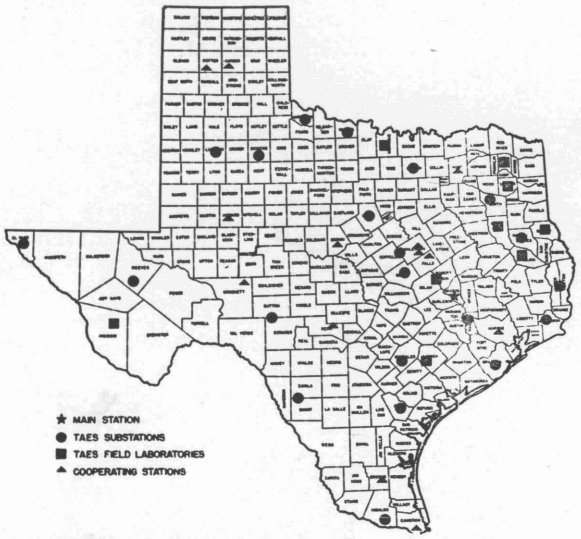

Location of field research units of the Texas Agricultural Experiment Station and cooperating agencies

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Citrus and other subtropical fruits
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Oil seed crops
Ornamental plants
Brush and weeds
Insects
Beef cattle
Dairy cattle
Sheep and goats
Swine
Chickens and turkeys
Animal diseases and parasites
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