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TEXAS AGRICULTURAL EXPERIMENT STATION

A. B. CONNER, DIRECTOR
COLLEGE STATION. BRAZOS COUNTY, TEXAS

BULLETIN NO. 493

AUGUST, 1934

DIVISION OF POULTRY HUSBANDRY

The Amount of Vitamin A Potency Required By Hens for Egg Production

Agricultural & Mechanical College of Tenas

College Station, Texas.

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[†]As of August 1, 1934

Experiments reported in Bulletin 468 showed that yellow corn does not supply enough vitamin A for maintenance and for production of eggs of proper vitamin A potency by White Leghorn pullets. The work was accordingly continued at higher levels in vitamin A potency supplied by means of heat-dried alfalfa leaf meal, which has usually been found to be quite high in vitamin A. Whereas in the former experiments vitamin A was supplied at averages of 0, 120, and 270 rat units per day, the averages supplied in the experiments here reported were 224, 336, and 444 units per day over a period of 290 days, the potency being measured biologically by use of rats.

The fowls receiving the higher amounts of vitamin A showed a larger weight, the differences appearing after the fourth month indicating that stores of the vitamin A in the bodies of the fowls fed the smaller amounts were beginning to be exhausted by that time. The mortality was greatest with the group consuming the lowest amounts of vitamin A, mostly after 8 months' feeding, indicating that this amount of vitamin. A was insufficient for egg production and maintenance of life. The fowls receiving 444 units of vitamin A laid about 15% more eggs than either of the other groups and while the vitamin A content of the eggs decreased in all groups as the laying progressed, the decline was greatest with those receiving the lower levels of the vitamin A. At the close of the experiment the eggs from the three lots contained 6, 12, and 15 units of vitamin A per gram respectively, indicating that an even greater intake than 444 units of vitamin A is desirable, since 15 units per gram in the egg is yet short of the desired amount of 20 units or more per gram. Green vegetation is recommended as the most practical means of supplying the required amount of vitamin A, although it can be done with commercial cod liver oil. Rations usually fed laying hens do not supply enough vitamin A for maintenance and high egg production unless supplemented with green feed. The hen apparently requires about four units in the feed to produce one unit of vitamin A in the eggs, in addition to the units required for maintenance.

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THE AMOUNT OF VITAMIN A POTENCY REQUIRED BY HENS FOR EGG PRODUCTION

R. M. SHERWOOD AND G. S. FRAPS

Vitamin A is essential to the life of animals, though the weight needed is very small. It is required in relatively moderate quantities for maintenance, somewhat larger quantities for growth, and in relatively very large quantities for production of eggs or milk (3), especially if the eggs or milk are to have a high potency in vitamin A (14). Vitamin A is supplied to poultry chiefly in yellow corn, alfalfa meal, alfalfa leaf meal, or in green growing grasses, clover or other grazing. The yellow coloring matter known as carotene found in such feeds may be converted in animals to vitamin A. At the present time it is not possible to say what portion of vitamin A potency is present in feeds, milk, or eggs, etc. as vitamin A and what is present as carotene. The term vitamin A is often used in this publication, for brevity, to mean vitamin A potency, that is, the biological effects due to vitamin A and/or carotene.

The fact that yellow corn alone is not sufficiently high to supply enough vitamin A to pullets for egg production has been pointed out in Bulletin 468 of this station (14). From experiments there described we estimated that the pullets required for maintenance alone about 105 units of vitamin A potency per day, or 33 units per pound per day. It was pointed out that ordinary alfalfa meal, which usually contains much more vitamin A than yellow corn, would not supply enough vitamin A to produce eggs of high potency in vitamin A even if fed as 8% of the mash. Green growing plants (7, 8) are very high in vitamin A, and laying hens apparently require much green feed to provide sufficient vitamin A for maximum egg production accompanied by high vitamin A potency of the eggs.

The experiments in Bulletin 468 (14) are the only ones reported so far attempting to ascertain the quantitative relations between the vitamin A in the feed and that in the eggs. The vitamin A potency was fed at levels of 0, of 120, and of 270 rat units per day.

It was considered desirable to continue the work referred to with larger quantities of vitamin A. Furthermore, it was desirable to have additional work to ascertain the number of units of vitamin A required for the production of one unit in the eggs. Since we have found heat-dried alfalfa leaf meal to be usually quite high in vitamin A (7), it was used for the purpose of securing a high vitamin A content of the ration.

No experiments have been reported to ascertain the digestibility of carotene or of vitamin A. The experiments here reported are based upon the total amount fed. According to Ahmad and Malik (1), animals differ in their ability to synthesize vitamin A from carotene, as judged by the vitamin A content of the liver. With the ability of rats to synthesize vitamin A rated as 100, they give chickens a rating of only 24, while

rabbits are rated at 16 and cats at 0. The vitamin A potency of feeds may thus vary for different animals according to the relative proportion of vitamin A and carotene present.

Method of Procedure

White Leghorn fowls that had been raised upon the standard ration of the Texas Agricultural Experiment Station, including free access to green feed, were divided into three groups each containing 18 pullets and 18 hens. Two cockerels were rotated from group to group for a sufficient time to secure eggs suitable for hatching. Correction was made for the food eaten by the cockerels on the assumption that they ate the same as the hens. All three lots received yellow corn as a grain feed and a mash containing 20% of yellow corn. One group received no alfalfa meal. For a second group, 4% of wheat bran was replaced in the mash given the first group by 4% of heat-dried alfalfa leaf meal and for a third group a corresponding replacement was made with 8% of the alfalfa leaf meal. These groups will be referred to as the no-alfalfa group, the 4% alfalfa group, and the 8% alfalfa group. The complete rations used are given in Table 1. The feeding here described began on the morning of November 15, 1932 and ended August 31, 1933, the duration being 290 days. The

Table 1. Ingredients of mixtures used for laying fowls

Ingredients of feed	No-alfalfa group	4% alfalfa group	8% alfalfa group
Mash	per cent	per cent	per cent
Yellow corn meal	20	20	20
Wheat gray shorts	20	20	20
Wheat bran	20	16	12 20
Alfalfa leaf meal	0	4	
Ground whole oats	20	20	8 20
Grain			
Yellow corn ad lib			

fowls were kept in pens having cement floors to prevent access to green feed. They had an abundance of sunshine, so that the equivalent of vitamin D was well supplied (11).

Vitamin A was determined in samples of the yellow corn used, the mash, the alfalfa leaf meal, and in the yolks from representative samples of the eggs by Mr. Ray Treichler of the Division of Chemistry. The modified Sherman-Munsell unit method with rats was used as we have described elsewhere in full (5, 7). The eggs were kept in cold storage. One or more eggs were boiled; the yolk was separated and weighed and weighed portions of the yolk were fed twice a week to the test rats. Eight to eighteen eggs were used for each test. The rat unit of vitamin A as here used is the amount of feed fed daily in a 6-day week which will produce a gain of approximately 24 grams in eight weeks. In making the tests, the quantity of material which was thought might have the desired gain was

fed to one group of rats, and lower and higher quantities were fed to two or more other groups. In estimating the units of vitamin A the number of rats which survived, their individual weights, and the average weight of each group, for the various amounts of the same feed, were given consideration but the most attention was paid to the quantities of feed which gave the nearest to the desired gain of 24 grams in 8 weeks. Tables 2 and 3 contain the average weights of all the rats which survived, but the weights of the individual rats which survived the entire period and those which survived six weeks were also considered in deciding on the number of units. Rats numbering 575 were used in the work here reported not including those used to check the vitamin-A-free ration. These biological methods do not possess a high degree of accuracy, which fact must be taken into consideration in connection with the results.

Table 2. Details of estimation of vitamin A potency of egg yolks

Labora- tory number	Date collected	Group	Yolk fed daily, grams	No. of rats at beginning	No. of rats at end	Average gain per rat in 8 weeks, grams	Units vitamin A to one gram yolk
37350	11-21-32	No alfalfa	.033	6	2	6	i i kara
37350	11-21-02	no anana	.050	6	3	34	25
37396	12-23-32	" "	.050	6	3	30	22
37396	"	" "	.083	6	6	51	
37397	12-23-32	4% alfalfa	.050	6	4	23	20
37397	"	" "	.083	6	5	49	
37398	12-23-32	8% alfalfa	.033	6	3 -	8	
37398	"	" "	.050	6	4	33	23
37398	"	" "	.083	6	6	51	
37452	1-17-33	No alfalfa	.067	6	1	-8	
37452	"	" "	.100	6	5	36	12
37456	1-19-33	4% alfalfa	.067	6	4	29	16
37456	"	" "	.100	6	4	31	
37453	1-17-33	8% alfalfa	.033	6	2	14	
37453	"	" "	.050	6	6	25	20
37453	"	" "	.100	6	6	72	
37524	2-16-33	No alfalfa	.067	6	5	36	17
37524	"	" "	.100	6	5	63	
37524	"	" "	.133	6	6	51	
37525	2-16-33	4% alfalfa	.067	6	6	28	16
37525	"	" "	.100	5	5	53	
37526	2-16-33	8% alfalfa	.033	6	4	19	
37526	"	" "	.050	6	6	30	22
37526	"	" "	.100	6	6	41	Section 198
37587	3-15-33	No alfalfa	.067	6	1	3	
37587	"	" "	.100	6	4	32	12
37587	"	" "	.133	6	3 3	32	S-11: 160 5/10
37588	3-15-33	4% alfalfa	.067	6	3	21	13
37588	"	" "	.100	6	6	40	12 4 12 1
37588	"	" "	.133	6	5	57	
37589	3-15-33	8% alfalfa	.050	6	3	9	
37589	"	" "	.067	6	0	0	
37589	"	" "	.100	6	5	48	13
37669	4-18-33	No alfalfa	.100	6	5	30	. 11
37669	"	" "	.150	6	5	40	C
37669	3 10 00		.200	6	5	37	
37670	4-18-33	4% alfalfa	.067	6	4	18	13
37670	"	" "	.100	6	5	69	
37670	THE REPORT OF THE PARTY OF THE		.133	6	5	43	1 9 / 8-
37671	4-18-33	8% alfalfa	.067	6	3	20	. 14 .
37671		" "	.100	6	3	34	
37671	188 " 201	." ."	.133	6	6	46	3 2 5

Table 2. Details of estimation of vitamin A potency of egg yolks-Continued

Labora- tory number	Date collected	Group	Yolk fed daily, grams	No. of rats at beginning	No. of rats at end	Average gain per rat in 8 weeks, grams	Units vitamin A to one gram yolk
37696 37696	5-19-33	No alfalfa	.100 .150	6 6	2 5	18 56	8
37696 37697 37697	5-19-33	4% alfalfa	.200 .067 .100	6 6	6 5 5	38 29 43	15
37697 37697	"	" "	.150 .200	6 6	5 6 6	47 66	
37698 37698 37698	5-19-33	8% alfalfa	.067 .100 .150	6 6	3 6 6	21 31 43	14
37698 37735	6-19-33	No alfalfa	.200 .100 .150	6 6	5	57 3 27	7
37735 37735 37736	6-19-33	" " 4% alfalfa	.200	6 6	5 5 2 5	41 27	15
37736 37736 37736	"	" "	.100 .150 .200	6 6	5 4 6	35 43 63	
37737 37737 37737	6-19-33	8% alfalfa	.067 .100 .150	6 6	6 5	31 51 35	17
37737 38159 38159	8-21-33	No alfalfa	.200 .100 .150	6 6	6	59 0 21	6
38159 38157	8-19-33	" " 4% alfalfa	.220	6 6	4 6 4	41 32	
38157 38157 38158	" " 8-19-33	" " 8% alfalfa	.100 .150 .050	6 6	6 5 3	26 28 18	12
38158 38158	"	" "	.067	6 6	6 6 0	25 49	15
37724 37724 37724	5-31-33	Green feed	.033 .050 .067	6 6	3 4	-13 36 33	22

The heat-dried alfalfa leaf meal was excellent in quality, containing about 80 units of vitamin A potency per gram when fresh. It was much richer in this respect than ordinary alfalfa leaf meal. The yellow corn was of good quality, as it contained about 7 units of vitamin A per gram at the beginning of the experiment. The determination of vitamin A in the mash containing 20% yellow corn, and no alfalfa meal, showed that the yellow corn accounted for all of the vitamin A in this mash, showing that the other ingredients of this mash contained practically no vitamin A.

It has been shown by Fraps and Treichler (9) that the vitamin A potency of corn, alfalfa, and other feeds decreases slowly while they are stored. In order to allow for this decrease, it was assumed that the rate of decrease at the end of a month is approximately 7% of that at the beginning of the month, which is the rate found in previous work. The figures used in the calculations for the feeds are therefore corrected for the estimated loss of vitamin A. This correction is of course only approximate, a sufficient number of estimations not having been made to show exactly the rate of loss of vitamin A under the different conditions

which affect the loss. It is believed, however, that it is more accurate to make the corrections than to ignore the losses during storage.

Table 3. Details of estimation of vitamin A potency of feeds

Labora- tory number		Date rat feeding begun	Feed fed daily, grams	Number of rats at beginning	Number of rats at end	Average gain per rat in 8 weeks, grams	
37360	Mash for no alfalfa group	12-12-32	.600	6	1	4	
37360	" " " " "	12-14-32	.800	6	1 3	25	11/4
37361	Yellow corn	12- 9-32	.150	6	4	25	61/2
37361	" "	12- 7-32	.200	6	5	36	
37362	Heat-dried alfalfa leaf meal	12-16-32	.010	6	4	8	80
37362	" " " " "	12-19-32	.020	6	4 6 5	71	
37362	" " " " "	12-23-32	.030	6	5	45	
37362	" " " " "	4-12-33	.017	6	1	15	
37595	Yellow corn	3-31-33	.150	6	1 3 5 5	4	
37595	" "	4- 3-33	.200	6	5	29	5
37595	" "	4- 3-33	.300	6	5	35	
37596	Heat-dried alfalfa leaf meal	4- 3-33	.013	6 6	0		
37596	" " " " "	4- 7-33	.017		1	26	55
37596	" " " " "	4- 7-33	.020	6	5	31	
37695	Yellow corn	6-16-33	.200	6	1 5 3 5	27	5
37695	" "	6-19-33	.300	6		36	
37695	" "	6-16-33	.150	6	3	8	
37733	Yellow corn	7-21-33	.150	6	3 3 2 5	16	
37733	" "	7-24-33	.200	6 [2	20	4
37733	" "	7-26-33	.300	6	5	38	

The average amounts of vitamin A fed during the period of 290 days was 224 units per day per fowl for the no alfalfa group, 336 units for the 4% alfalfa group, and 444 units for the 8% alfalfa group.

Effect of Quantity of Vitamin A in the Food on Mortality and Weights of the Fowls

In the experiments previously reported (14) when no source of vitamin A was used, the pullets fed on white corn lived an average of 135 days; there was a wide variation with the individual pullets since they lived from 34 to more than 199 days. Therefore, in the experiment here reported it is probable that an equally wide variation occurred in the amounts of vitamin A stored by the fowls.

The mortality of the fowls in the present experiment is shown in Table 4. The table shows that fowls in all the groups began to die in May, towards the end of the laying season, and that about twice as many died in the no-alfalfa group as in either of the groups receiving alfalfa meal. This would indicate that the fowls of the no-alfalfa group were not receiving enough vitamin A in the yellow corn for proper maintenance under the strain of egg production. These fowls received an average of 224 units of vitamin A per day. The results are somewhat different from those presented in Bulletin 468, since in this work the mortality was low for the two groups of pullets receiving 120 and 270 units of vitamin A per day, being fed yellow corn, but this difference can be accounted for by the fact that

the previous experiment was discontinued the first of May, because almost all the pullets in the white-corn group had died by that time, and the few remaining in this group were in bad condition. In the experiment here

Table 4. Number of deaths of fowls with day of month on which each occurred

	No-alfal	fa group	4% alfa	lfa group	8% alfalfa group		
Month	Number	Day of month	Number	Day of month	Number	Day of month	
November	0		0	with integ	0		
December	0	_	0	_	0	_	
January	0	-	0	_	2	8,8	
February	1	3	0	~	0	_	
March	0	-	0	-	0	-	
April	0	-	0	-	0	_	
May	1	18	1	10	2	11,26	
June	2	1,21	3	7,12,14	1	12	
July	5	1,2,4,6,15	0		1	5	
August	2	14,18	1	3	0	-	
Total	11		5		6		

reported, there was a rapid increase in the death rate of the fowls in the no-alfalfa group (which had yellow corn, however) just after having been on the experiment for the same length of time as in the previous experiment. Experiments such as these should be continued for several years.

Payne and Hughes (13) have also shown that hens fed a feed containing 65% of yellow corn have a higher mortality than those fed a similar feed containing 4% of alfalfa leaf meal.

The average weights of the living fowls during the progress of the experiment are shown in Table 5. The 4% alfalfa group averaged slightly more than the other two groups to begin with, but after four months

Table 5. Average of weights of fowls in pounds

	No-alfalfa group	4% alfalfa group	8% alfalfa group
November 16, 1932	2.97	3.07	3.00
December 16	3.06	3.20	3.16
January 16, 1933	3.39	3.56	3.44
February 16	3.41	3.59	3.52
March 16	3.24	3.38	3.45
April 16	3.12	3.28	3.28
May 16	2.87	2.98	3.08
June 16	2.89	2.98	3.11
July 16	2.88	2.87	2.99
August 16	2.71	2.87	2.94
Average	3.05	3.18	3.20

(February) the average weights were almost all closely related to the quantities of vitamin A potency fed. The pullets fed the most vitamin A were heaviest. Similar results were obtained with pullets fed the lower quantities of vitamin A already referred to (14).

Effect of Quantity of Vitamin A on Number of Eggs Produced

In the previous experiment it was shown that the number of eggs laid per pullet was closely related to the quantity of vitamin A fed. The pullets on the white corn averaged 55 eggs per pullet for the period of the experiment, those on the mixed corn 66, and those on the yellow corn 80. The number of eggs produced on the higher levels of vitamin A potency fed in the present experiment is given in Table 6. The fowls in the noalfalfa group and those in the 4% alfalfa group produced practically the

8% alfalfa Month No-alfalfa 4% alfalfa group group group November* 1.34 2.06 1.70 5.62 5.59 4.81 December 12.14 10.31 12.88 January February 14.75 14.20 14.67 18.70 March 18.94 19.37 17.13 18.31 19.10 April May 14.56 14.15 June 11.37 10.41 14.33 12.40 8.44 July 9.43 7.37

4.87 107.69

124.59

Table 6. Average number of eggs per fowl

4.40

August

same number of eggs, but the number of eggs produced by the fowls receiving 8% of alfalfa leaf meal was appreciably greater. The higher level of vitamin A potency apparently effected a greater production of eggs.

Payne and Hughes (13) have also shown that hens receiving 5% of alfalfa leaf meal in an all-mash feed containing 65% of yellow corn produced more eggs than hens receiving the yellow corn as a sole source of vitamin A.

Effect of Vitamin A on Percentage of Eggs That Hatched

In the previous experiment it was shown that the percentage of eggs hatched increased with the increase in the amount of vitamin A fed the pullets.

The results secured in the present experiment at higher levels are given in Table 7. Here, also, the percentage which hatched was directly related to the vitamin A potency of the feed, being highest for the eggs from the 8% alfalfa group.

Effect of Quantity of Vitamin A on the Vitamin A Content of Eggs

From the results of our previous work we anticipated that alfalfa leaf meal would not furnish sufficient vitamin A to produce eggs of high potency even when fed in quantities much higher than is the ordinary practice. The vitamin A potency of the egg yolks is given in Table 8. It is seen that with the fowls fed no alfalfa the vitamin A in the eggs

^{109.03} *These records represent production during last half of November only.

decreases during the laying period, as was also found in the previous experiment (14). The decrease is smaller in the eggs from the fowls in the 4% or 8% alfalfa group and the minimum content of vitamin A

Table 7. Relation of percentages of eggs which hatched to vitamin A in feed of hens and in corresponding egg yolks

Date hatched	r group vitamin er day, hatched	a group vtamin er day hatched	a group vitamin er day	Estimated units per gram of egg yolk				
	No-alfalfa g 224 units vit A fed per o per cent hat	4% alfalfa g 336 units vts A fed per o	8% alfalfa g 444 units vit A fed per	No-alfalfa group, units	4% alfalfa group, units	8% alfalfa group, units		
May 1, 1933	13.8	31.3	33.3	11	13	14		
June 9, 1933	47.0	50.2	. 60.2	8	15	14		
Average	38.4	46.0	52.4	9.5	14	14		

occurs near the point of maximum egg production, after which the units are almost constant, and even show a tendency to increase with the diminished egg production. But these eggs at the end of the experiment have appreciably less vitamin A potency than the eggs at the beginning. None

Table 8. Units of vitamin A per gram of yolk of eggs collected at different times

Date collected	No-alfalfa group	4% alfalfa group	8% alfalfa group
November 21, 1932	25		
December 23, 1932	22	20	23
January 17, 1933	12	16	20
February 16, 1933	17	16	22 *
March 15, 1933	12	18	13
April 18, 1933	11	13	14
May 19, 1933	8	15	14
June 19, 1933	7	15	17
July (Assumed)	6.5	13.5	16
August 19, 1933	6	12	15

of the rations furnished enough vitamin A to maintain the vitamin A potency of the eggs at the high level found at the beginning of the experiment.

After 9½ months, the eggs from the no-alfalfa group contained 6 units per gram of egg yolk, those from the 4% alfalfa group contained 12 units per gram, and those from the 8% alfalfa group contained 15 units per gram.

Bethke, Kennard and Sassaman (2) tested the eggs of White Leghorn hens after they had been fed 9 months on various feeds. The results are not expressed in units, but from the diagrams given it may be estimated that the hens receiving an all-mash ration containing 30% of yellow corn produced eggs containing about 4 units of vitamin A per gram

of egg yolk. Those receiving the all-mash ration and also having access to alfalfa hay, produced eggs containing 5 units per gram of yolk. The hens which had access to a blue-grass pasture produced eggs containing 18

Table 9. Pounds of grain and mash eaten per month per fowl

	No-alfal	fa group	4% alfa	lfa group	8% alfalfa group		
Month	Grain	Mash	Grain	Mash	Grain	Mash	
November*	1.23	1.36	1.34	1.37	1.31	1.17	
December	2.42	3.42	2.70	2.84	2.75	2.59	
January	2.86	2.32	2.81	3.20	2.88	2.73	
February	2.04	3.34	2.50	3.32	2.31	3.64	
March	1.72	3.90	2.02	3.65	2.18	3.15	
April	3.01	3.13	2.91	3.55	2.88	3.53	
May	2.40	2.61	2.39	2.70	2.39	3.12	
June	2.29	2.06	1.95	1.87	1.89	2.98	
July	1.92	2.27	2.51	2.05	2.44	2.62	
August	1.59	1.53	1.58	1.57	1.67	2.26	
Total	21.48	25.94	22.71	26.12	22.70	27.79	

^{*}These records represent feed consumed during last half of November only.

units per gram of yolk, while those receiving a similar mash containing 2% of medicinal cod liver oil produced eggs containing 14 units of vitamin A per gram of yolk. These results, as recalculated, are in full accord with those presented in this and the previous bulletin. Similar results have been found with cows, as the vitamin A in the butter depends upon the feed (3) as well as on the stage of lactation.

The total output of vitamin A in the eggs is given in Table 11. The average weight of the yolks in all the eggs was 16.6 grams and this figure

Table 10. Total units of vitamin A fed per hen per month in both grain and mash

Month	No-alfalfa group	4% alfalfa group	8% alfalfa group
November*	4432	6753	7956
December	9160	13770	17169
January	9206	13855	16825
February	7008	12348	16984
March	6243	11109	14339
April	8584	12092	15517
May	6633	9151	12621
June	5765	6555	10392
fuly	4634	7301	9878
August	3443	4580	7137
Total	65108	97514	128818
Average per day	224	336	444

^{*}These records represent units of vitamin A for last half of November only.

was used in the calculations. The output in the eggs is directly related to the quantity of vitamin A fed the fowls. The eggs of the no-alfalfa group contained 20,697 rat units per hen, those of the 4% alfalfa group contained 26,338 units, and those of the 8% alfalfa group contained 34,141 units. The average daily output of vitamin A in the eggs was 72

units for the no-alfalfa group, 91 for the 4% alfalfa group, and 118 for the 8% alfalfa group.

Vitamin A potency is stored in the body of the animal either as vitamin A as such or as carotene and part of this reserve store is placed in the eggs when the quantity in the food is not sufficient, so that the decrease in vitamin A potency of the eggs is gradual (Table 8). The question arises whether pullets can secure enough vitamin A even from food very high in

Table	11.	Units	of	vitamin	A	per	fowl	in	eggs	laid	during	each	month	and
				apparent	pe	rcen	tage	rece	overed	in	eggs			

Month	No- alfalfa group	4% alfalfa group	8% alfalfa group	Apparent percentage recovery in eggs		
				No-alfalfa group	4% alfalfa group	8% alfalfa group
November*	556	855	706	12.6	12.7	8.9
December	2052	1856	1837	22.4	13.5	10.7
January	2418	2738	4276	26.3	19.8	25.4
February	4162	3772	5357	59.4	30.5	31.5
March	3725	4087	4180	59.7	36.8	29.2
April	3128	3951	4439	36.4	32.7	28.6
May	1879	3626	4174	28.3	39.6	33.1
June	1321	2592	4044	22.9	39.5	38.9
July	1018	1891	3293	22.0	25.9	33.3
August	438	970	1835	12.7	21.2	25.7
Total	20697	26338	34141		ALL TO	
Average				30.2	27.2	26.5
Average per day	72	91	118	100		

^{*}Only the last half of November figures in these computations.

vitamin A potency to produce eggs high in vitamin A to the end of the laying season. That is to say, the requirements of a laying hen are very high and it might not be possible for her to utilize the vitamin A potency in the food rapidly enough to serve as a source of all that is needed. If this is the case the vitamin A potency of the eggs must fall off towards the end of the laying period regardless of the quantity fed. In order to ascertain if this occurs, eggs were tested from near the end of the laying period from pullets which had been receiving green feed. The results are given in Table 2 for sample No. 37724. It was found that these eggs contained 22 units per gram of yolk, nearly as high as the 25 units of vitamin A secured at the beginning of this and the previous experiment. That is to say, if the hens are supplied with sufficient amounts of vitamin A in the food it appears probable that they can maintain the vitamin A potency of the eggs.

Quantities of Vitamin A Required for the Vitamin A in the Eggs

In the experiment in Bulletin 468, previously referred to, it was calculated that one unit of vitamin A in the egg requires 6.3 units in the feed. The requirements for the eggs were calculated for this experiment in the same way as for the previous one. The weights of grain and mash

consumed are given in Table 9, and other details necessary for these calculations are given in Tables 3, 6, and 8. The number of units of vitamin A consumed monthly in the feed is given in Table 10. The number of units in the eggs divided by the number of units fed gives the apparent recovery and, expressed in per cent, is from 8.9 to 59.7, with an average of 30.2, 27.2, and 26.5 respectively for the three groups (Table 11). Apparently the percentage of vitamin recovered in the eggs is greatest at the lowest levels, and least at the highest level of feeding. This apparent percentage recovery is not correct because the vitamin A stored by the hen is not taken into account and some of the vitamin A or carotene fed is used for maintenance, while some of it is probably not digested.

With the no-alfalfa group, the apparent percentage recovered is low at the beginning and at the end of the laying period, with a maximum in March. This maximum coincides with the greatest production of eggs. The apparent recovery of vitamin A decreases from March because the number of eggs and their vitamin A potency both decrease to a greater extent than the consumption of feed.

The apparent utilization of vitamin A by the other two groups of fowls is about the same at the beginning of the laying period, rises to a maximum in May and June with the 4% alfalfa group and in June with the 8% alfalfa group, and then decreases, but not so much or so rapidly as that of the no-alfalfa group. The maximum apparent utilization is due to a lowered food intake in May and June, as can be seen in Table 10. The maximum production of eggs comes earlier.

The number of units required for one unit in the egg for the entire period is calculated in Table 12. The fowls of the 4% alfalfa group consumed 32,406 units more vitamin A than those which did not receive the alfalfa and they produced 5,641 units more vitamin A in the eggs. If we assume that none of the supplementary vitamin A was used for maintenance, one unit of vitamin A in the egg requires 5.7 units in the feed. By a similar calculation, using the results from the 4% alfalfa group and those of the 8% alfalfa group, we find that one unit of vitamin A in the eggs requires 4.0 units in the feed. In another calculation, using the data from the no-alfalfa group and the 8% alfalfa group, we find that one unit of vitamin A potency in the eggs require 4.7 units in the feed, which is the average of the other two groups. These figures represent the average for the entire period.

The units of vitamin A required in the feed for one unit in the eggs decrease as the average number of units fed increases. One unit in the eggs requires 6.3 units in the feed at 270 units (14), 5.7 units at 336 units, and 4.0 at 444 units in the feed.

In the previous work at lower levels (14), we found that 6.3 units in the feed were required for one unit in the egg. In the work here reported 5.7 units were required when the results with the no-alfalfa and the 4% alfalfa group are used for the calculation. However, at the higher level of 8% alfalfa, compared with 4% alfalfa, only 4 units in the feed are required for one unit in the eggs. This difference may be due to some of

the vitamin A fed the 4% alfalfa group being used for maintenance as well as for producing the eggs. That this explanation may be correct is suggested by the high mortality of the no-alfalfa group towards the end of the laying period (Table 4), which indicates that these fowls were not receiving sufficient vitamin A for maintenance, so that at the 4% alfalfa level, some of the vitamin A would be used for maintenance. When the maintenance requirements are more nearly satisfied, the vitamin A is apparently more efficiently utilized for the eggs. There is also some evidence for the correctness of this explanation in the fact that the

Table 12. Units of vitamin A potency required for the units of vitamin A in eggs

n eggs, 4% alfalfa group	26338 20697
Difference in eggs due to 4% alfalfa n feed, 4% alfalfa group n feed, no-alfalfa group	5641 97514 65108
Difference in feed due to 4% alfalfa. Units in feed required for one unit in eggs, no-alfalfa versus 4% alfalfa	32406 5.7
In eggs, 8% alfalfa group In eggs, 4% alfalfa group	34141 26338
Difference in eggs due to 4% alfalfa	7803 128818
Difference in feed due to 4% alfalfa	$\frac{97514}{31304}$
Units in feed required for one unit in eggs, 4% alfalfa versus 8% alfalfa	4.0
n eggs, 8% alfalfa group n eggs, no-alfalfa group	34141 20697
Difference in eggs, due to 8% alfalfa feed, 8% alfalfa group feed, no-alfalfa group	13444 128818 65108
Difference in feed due to 8% alfalfa	63710

units of vitamin A required in the feed for one unit in the egg decrease regularly from 6.3, as shown in Bulletin 468, to 5.7 to 4.7 to 4.0, with the increase of the vitamin A fed, as pointed out above. A portion of these differences, however, may be due to the fact that the estimation of vitamin A is not very accurate, and also to differences in the quantities of vitamin A stored in the bodies of individual fowls. It is possible that at still higher levels the efficiency of utilization may not be so good as here given.

If we assume for the purpose of this discussion that the estimate of 4 units of vitamin A potency for one unit of vitamin A in the eggs is correct, then 25% of the vitamin A is utilized for the eggs, and any apparent excess in the eggs over this percentage comes from the body of the fowl. On the other hand, when less than 25% of the vitamin A fed appears in the eggs, the difference may be used for maintenance or may be partly stored in the body of the fowl. The apparent percentage re-

covery of vitamin A as given in Table 11 may be studied on the basis of this assumption. The fowls of the no-alfalfa group could have been gaining vitamin A or using some of that fed for maintenance, in November, December, June, July, and August. In January, February, March, April, and May, they were transferring vitamin A from their bodies to the eggs. The fowls of the 4% alfalfa group could have gained vitamin A in November, December, January, and August. In the other months, they should have been losing from their bodies. Those of the 8% alfalfa group were apparently losing every month except November and December, although very slightly in January and August. Thus the hens fed the smallest amounts of vitamin A lost vitamin A during 5 months of the experiment, those fed the next amounts lost during 6 months, and those fed the most vitamin A lost during 8 months of the 91/2 months of the experiment. It does not seem plausible that the fowls receiving the higher amounts of vitamin A drew on their bodies more continuously than those receiving lower amounts. It is more probable that the fowls utilized for the eggs more than 25% of the vitamin A fed in excess of maintenance.

The highest apparent average percentage recovery was 47% for four months with the no-alfalfa group, 37% for 5 months with the 4% alfalfa group, and 32% for 6 months with the 8% alfalfa group. Although it seems probable that the fowls will draw upon the vitamin A stored in the bodies as long as the eggs are below normal in vitamin A, these figures also indicate the possibility that the fowls during the laying period can place in the eggs more than 25% of the vitamin A fed in excess of maintenance, possibly nearly 30%. That is to say, if the fowls receive just enough vitamin A for maintenance and for the highest vitamin A potency of the eggs that they are capable of producing, they may require a little less than 4 units in the feed above maintenance for one unit in the eggs. Below this point, the fowls draw on the vitamin A stored in the body; above it, the efficiency would be less, since the maximum limit of utilization has already been reached. The amount of vitamin A required in the feed would depend upon the number of eggs being laid.

Quantities of Vitamin A Required in the Food for Maintenance and Egg Production

The work here reported does not permit an estimation of the maintenance requirements of the fowls, since there is no way to allow for the vitamin A stored in their bodies at the beginning of this experiment. The previous estimate (14) of 105 units a day in the feed for maintenance of the laying pullet is probably too low, since some of the vitamin A assigned to the production of eggs in these tests must have gone to maintenance, and some of the units used for maintenance must have come from the body of the fowl. The previous estimate (14) of the requirement of 6.3 units in the feed to a unit in the eggs was a little high, since, as shown here, some of the vitamin A assigned to the eggs went to maintenance purposes. If the maintenance requirement given in Bulletin 468 is re-

calculated, using 4 units of vitamin A in the feed for one unit in the egg, the vitamin A in the feed used for maintenance of the mixed-corn lot is raised from the 105 units previously given, to 110 units. The difference is small.

Estimates of the quantity of vitamin A required for maintenance and egg production were given in Bulletin 468. For White Leghorn eggs with a yolk weighing 15 grams and the vitamin A content of 20 units to the gram it was estimated that pullets producing 10 eggs a month would require 1410 units in the feed per day. While the work here presented indicates that somewhat smaller amounts of vitamin A might be required than was previously calculated, the requirement still remains very high. A mash containing 8% of heat-dried alfalfa meal, high in vitamin A, together with 20% of yellow corn, with a scratch grain consisting entirely of yellow corn, fell below furnishing the amount required for producing eggs of high potency. In ordinary practice, lower percentages of alfalfa leaf meal are used and less yellow corn. Ordinary alfalfa meal is much lower in vitamin A than the heat-dried alfalfa meal used in this work. This confirms our previous statement that it is very difficult to supply laying hens with sufficient amounts of vitamin A to produce eggs of maximum vitamin A content without feeding green feed.

As shown in Table 11, the output of vitamin A in the eggs varies considerably. During the heavy laying period, the requirements are heavy, but while the hens are producing few or no eggs, the total requirements for eggs are low. During the rest period, in addition to satisfying maintenance requirements, the animal may store vitamin A sufficient in addition to maintenance requirements to produce eggs of high vitamin A potency at the beginning of the next laying period and to serve in maintaining the life of the animal.

The hen requires high quantities of vitamin A to produce eggs of high potency in vitamin A. At a somewhat lower level of vitamin A feeding, it is possible the hen may be able to maintain her output of eggs and survive for the usual period of life, even though the eggs are a little low in vitamin A potency. At a somewhat lower level, the output of eggs decreases and the life of the animal is shortened. The requirements for each level would depend to some extent upon the total output of eggs during the year. Experiments lasting over the entire life of the animal would be necessary to ascertain the units of vitamin A potency required at these various levels of feeding.

Supplying Vitamin A to Laying Hens

The work here presented shows that when hens are fed a scratch grain composed entirely of yellow corn and mash containing 20% of yellow corn, the eggs decrease in vitamin A with the advance of the laying season and the fowls suffer a high mortality towards the end of the season. Such a feed apparently does not furnish sufficient vitamin A for egg production and for maintenance.

Hens fed yellow corn ad lib and a mash containing 20% ground yellow corn and either 4% or 8% of alfalfa leaf meal of high vitamin A potency, also produced eggs which were deficient in vitamin A and the deficiency was more apparent as the laying season progressed. Further work is needed to show whether or not such rations contain sufficient vitamin A for maintenance through a period of several years.

Fresh growing grasses, alfalfa, clover, mustard, spinach, turnip greens, or similar green feeds are necessary if laying hens are to secure sufficient vitamin A to produce eggs of maximum potency. Sprouted oats, cabbage, lettuce, or similar light colored green feeds are not sufficiently rich in vitamin A for this purpose.

Cod liver oil is high in vitamin A (7) and can be used for the purpose of supplying it (2, 10). According to Marcus (12), 85% of the vitamin A is destroyed in 10 days when the oil is mixed with a basal diet, and it is destroyed when mixed with other powdered materials. If this occurs, cod liver oil would not be a satisfactory carrier of vitamin A for commercial mixed feed, and if used to supply vitamin A, should be mixed with the feed not more than 2 or 3 days before it is fed.

Heywang (10) found sun-cured alfalfa meal to be a poor substitute for fresh alfalfa, while sun-cured alfalfa leaf meal was more satisfactory than the sun-cured alfalfa meal. Whole alfalfa hays were not reliable substitutes for fresh alfalfa. Fresh alfalfa fed ad libitum and cod-liver oil at the rate of 1.5% of the feed intake were superior, he says, to all the other supplements.

Payne and Hughes (13) state that alfalfa meal fed as 10% of the total ration supplied adequate amounts of vitamin A, basing this statement upon the eggs laid and the mortality of the hens. Five per cent of the total feed intake of alfalfa leaf meal proved adequate when 65% of the ration consisted of yellow corn, and 10% was sufficient when white corn composed 65% of the ration. However, the vitamin A content of neither the yellow corn, the alfalfa meal, nor the eggs was given. If we assume the yellow corn to average 5 units of vitamin A per gram and the alfalfa leaf meal to average 30 units, this mash containing 65% of yellow corn and 5% of alfalfa meal would average about 4.8 units of vitamin A per gram. One square inch of green sprouted oats, they say, did not supply sufficient vitamin A.

Green growing plants appear to be very high in vitamin A. Spinach may contain as much as 952 units (6) per gram of dry matter. There are considerable losses in drying (6) or curing (4, 15) green feeds. Besides green feed, yellow corn and alfalfa products are the chief sources of vitamin A in chicken feed. Fresh yellow corn may contain 7 units of vitamin A to a gram, while the stored corn may contain less than 3 units, since the vitamin A content decreases in storage (9). Alfalfa meal (7), which is the ground alfalfa hay, may contain 3 to 20 units per gram, though Colorado alfalfa hay (4), cured in diffused light, was claimed to contain 9 to 64 units per gram. Heat-dried alfalfa may contain as much as 100 units (7) to a gram. Commercial alfalfa leaf meal may contain 7 to 20 units to a

gram (or perhaps more), while heat-dried alfalfa leaf meal (4, 7) may contain 50 to 80 units, or more. The content would depend upon the method of preparing the hay and the time it has been stored. According to Smith and Briggs (15), alfalfa hay cured in the dark contains about 200 units a gram, while that exposed in the field contains about 120 units with a field exposure of 2¾ hours, decreasing to about 7 units with field exposure of a week. In any event, alfalfa products are quite variable in their vitamin A potency.

In the work here reported, the yellow corn contained 6.5 units of vitamin A per gram at the beginning of the experiment and about 4 units at the end, while the heat-dried alfalfa leaf meal contained about 80 units at the beginning and 37 units per gram at the end. Since the grain fed was composed exclusively of yellow corn, it furnished from 6.5 to 4 units per gram. The mash containing 8% of alfalfa leaf meal and 20% of yellow corn, furnished from about 7.2 to 4.4 units of vitamin A per gram; that with the yellow corn alone contained 1.3 to 0.8 units per gram; and that with 4% alfalfa leaf meal in the mash, from 4.5 to 2.6 units per gram. Since the mash and grain were eaten in approximately equal proportions, the noalfalfa lot received about 3.9 to 2.4 units per gram, and this amount was certainly inadequate. The 4% alfalfa lot received 5.8 to 3.5 units per gram and the 8% alfalfa lot received from 6.8 to 4.2 units per gram, but none of these provided enough vitamin A for full potency of the eggs.

Since the ordinary scratch grains and mashes contain usually less yellow corn and alfalfa leaf meal than was fed in this experiment, and since the commercial alfalfa meal or alfalfa leaf meal used usually has a much lower content in vitamin A, it appears probable that hens as ordinarily fed, unless they have access to suitable green feeds, do not receive sufficient vitamin A and produce fewer eggs and have shorter lives, than they would have if they received adequate supplies of vitamin A.

SUMMARY

Three groups of White Leghorn fowls consumed in their feed daily, on an average, 224, 336, and 444 units of vitamin A potency. The group receiving the smallest amount of this vitamin was fed a mash containing 20% of ground yellow corn and yellow corn alone as the grain feed. The other groups received the same grain, and mash in which 4% and 8% of heat-dried alfalfa leaf meal replaced an equal amount of wheat bran. The vitamin A potency of the feed and that in the eggs were estimated by biological methods by means of rats.

The mortality was highest in the group receiving the smallest amount of vitamin A. The 224 units of vitamin A eaten daily by the hens on this ration was slightly less than that eaten by the hens on a similar ration the previous year. The mortality was no higher this year for the same length of time than in the experiment of last year. After that time a rather large number (nearly 30%) of the fowls of the no-alfalfa group died. This ration was evidently very deficient in vitamin A for a long-time feeding.

The fowls receiving on an average 444 units of vitamin A in their feed daily averaged heaviest in weight, followed by those receiving 336 units. Those receiving 224 units were lightest. These differences in weight appeared after the fourth month of the experiment.

The fowls fed the two lower amounts of vitamin A laid approximately the same number of eggs. Those receiving the largest amount of vitamin A laid nearly 15% more eggs than did the other groups. The vitamin A content of the eggs from all of the lots decreased as the laying period progressed. The decline was greatest for the lot receiving the highest amount of vitamin A potency. Egg yolks of the different lots at the close of the period of 9½ months contained 6, 12, and 15 units of vitamin A per gram, respectively.

In two hatching tests the percentage of eggs hatched increased with the increase in the amounts of vitamin A in the feed, but the percentage hatched was not closely related to the amount of vitamin A in the eggs.

The units of vitamin A potency required in the feed in addition to maintenence required for one unit in the eggs decreased as the average number of units fed increased. One unit in the eggs required 6.3 units in the feed when 270 units a day were fed, 5.7 units when 336 units were fed, and 4.0 units when 444 units were fed daily. The fowls apparently used for maintenance larger proportions of the vitamin A fed at the lower levels of feeding than at the higher levels. Since none of the lots was fed a ration free from vitamin A, it was not possible to estimate maintenance requirements in this experiment. Laying hens have high requirements for vitamin A, especially if eggs high in vitamin A potency are desired.

The results of this study as well as those of last year point out that rations usually fed laying hens apparently do not supply enough vitamin A for maintenance and high egg production unless the hens have access to green grass or similar green feed. It seems probable that laying fowls which do not have access to green feed and fed many of the ordinary laying feeds are likely to break down from deficiency of vitamin A during the second and third year, or possibly sooner.

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