TEXAS AGRICULTURAL EXPERIMENT STATION
A. B. CONNER, DIRECTOR
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DIVISION OF CHEMISTRY

THE VITAMIN A REQUIREMENTS OF DAIRY COWS

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Dairy cows must have feed high in vitamin A potency in order to continue to produce butter high in vitamin A potency. Silage and ordinary hays and fodders apparently will not supply enough vitamin A potency to maintain a high content of vitamin A in the butter fat. Green growing pasture grasses appear to be needed to maintain the production of butter fat high in vitamin A potency.

The vitamin A potency of butter fat of cows fed insufficient amounts of vitamin A decreased regularly during the period of lactation of the cow, from about 33 to 38 biological units per gram at the beginning to about 16 to 20 units in 4 weeks and to 5 to 12 units at the end of 5 months, the vitamin A potency of the butter depending to some extent on the vitamin A potency of the feed. The decrease during lactation is probably due to depletion of the vitamin A stored at the beginning of the lactation period.

At the beginning of the lactation period, the cows drew heavily upon the vitamin A and carotene stored in their bodies. Cows receiving little or no vitamin A in the feed continued to produce butter containing vitamin A for several months. Feeding 116,000 units of vitamin A potency per day in about 7 pounds of yellow corn and 6 pounds of heat-dried alfalfa meal was not sufficient to maintain the vitamin A potency of the butter fat. This number of units is probably greater than that ordinarily supplied in hays and fodders, since these are usually much lower in vitamin A potency than the heat-dried alfalfa meal used in the experiments. When cows producing butter containing only 1 and 5 units respectively of vitamin A potency per gram had been placed in pasture for two weeks, the vitamin A potency of the butter increased to 25 and 32 units respectively.

On an average one unit of vitamin A potency in the butter required approximately 11 units in the feed over maintenance. This is much higher than the 4 to 6 units over maintenance required by hens for the production of one unit in eggs. The cow seems to use vitamin A potency less efficiently than the hen. Poor quality hay or fodder low in vitamin A may cause losses of cows or calves.

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THE VITAMIN A REQUIREMENTS OF DAIRY COWS

G. S. Fraps, O. C. Copeland, and Ray Treichler

It has been known for some time that the vitamin A potency of milk depends upon the diet (14, 16) and that the vitamin A potency of butter varies from time to time (12). Cows receiving feed which did not contain sufficient amounts of vitamin A produced butter low in vitamin A potency (5). Seasonal changes of vitamin A potency in butter are known to occur which parallel the consumption of feed containing carotene (1, 2, 5). According to Moore (16) feeding of carotene increased both the carotene and the vitamin A of butter fat. Baumann and Steenbock (1) have recently claimed that only about 15 per cent of the vitamin A potency of butter is due to carotene, the remainder probably being due to vitamin A. Shrewsbury and Kraybill (22), however, state that the carotene of butter fat may account for an appreciable amount of its vitamin potency. McCosh and others (15) claim that the addition of 15 gm. of cod liver oil daily to an abundant and well chosen diet did not increase the vitamin A content of human milk. Baumann, Steenbock, Beeson, and Rupel (2) state that definite differences due to the breed were found in the butter fat by spectroscopic estimations of carotene and vitamin A.

Vitamin A potency may be due either to vitamin A itself, or to carotene, which can be converted into vitamin A by the animal or may be used for the same purpose as vitamin A. Carotene is known to occur in at least three different forms, alpha, beta, and gamma carotene, which may have different values for vitamin A potency (3). Different species or breeds of animals and individual animals of the same breed may differ in their ability to utilize carotene (2, 11, 23).

No attempt is made in this work here reported to distinguish between vitamin A potency due to vitamin A itself, and that due to carotene. The term vitamin A may be used, for brevity, to indicate vitamin A potency.

No studies have hitherto been made to ascertain the vitamin A potency of milk or butter fat at various stages of lactation. It has been shown by Sherwood and Fraps (20, 21) that the vitamin A potency of eggs decreases during the laying period even when the hens are fed liberal amounts of vitamin A, though the decrease was less with liberal feeding than with less abundant feeding.

Little work has been done to ascertain the quantities of vitamin A required by animals. Sherwood and Fraps (20) estimated that White Leghorn pullets while laying require about 33 units a pound for maintenance and 6.3 units for each unit in the egg yolk and that rations usually fed laying hens do not supply enough vitamin A for production of eggs rich in vitamin A, unless they have access to green grass or similar green feed. Later work by Sherwood and Fraps (21) showed that hens fed at higher levels of vitamin A required approximately four units of vitamin A in the feed for one unit in the eggs, and that the larger

amounts required at lower levels of feeding vitamin A were probably due to some of the vitamin A being required for maintenance. Fraps and Treichler (8) estimated that rats require 4 units for maintenance per day per pound, and 6 units for growth, and assumed that humans require 1,000 units per day per person. Copeland and Fraps (5) found that dairy cows fed for a long period of time with cottonseed meal and hulls or cottonseed meal, hulls, and sorghum silage produced butter fat very low in vitamin A. As the silage, though low in vitamin A, was fed in large quantities and supplied about 106,000 units of vitamin A per day, it appeared that the requirements of dairy cows for vitamin A were much higher than had hitherto been supposed. Baumann et al (2) calculated that 1.1 per cent of the carotene ingested in a low carotene ration was secreted into the milk; on a high carotene ration only 0.4 per cent was secreted. According to Converse and Meigs (4) there are indications that five to six months on low-quality hay rendered the milk nutritionally unsafe for calves, and that cows getting over-ripe timothy hay as a sole roughage consistently failed to have normal calves, probably due chiefly to deficiency of vitamin A.

The objects of the work here discussed were to ascertain the relation of the vitamin A content of the butter fat at various stages of lactation of the cow to the vitamin A content of the feed, and also to secure information regarding the number of units of vitamin A required for maintenance of the cow and for the secretion of vitamin A in the butter, work similar to that with hens previously reported from this Experiment Station (20, 21).

Method of Procedure

The work includes two separate experiments. In the first experiment, two cows were used. No. 59 received little or no vitamin A in her food while No. 61 received 6 to 7 pounds a day of yellow corn as a source of vitamin A. As will be seen, the yellow corn had practically no effect upon the vitamin A of the butter. In the second experiment, three cows were used, two of which were fed much larger quantities of vitamin A than in the first experiment. Heat-dried alfalfa meal, which is usually high in vitamin A activity (8), and yellow corn were the sources of vitamin A. Cow No. 196 received vitamin A in yellow corn only. No. 322 received 3 pounds of heat-dried alfalfa meal and yellow corn and No. 329 received yellow corn and 6 pounds of the alfalfa meal, which was about as much as the cow could eat. In each case, the feed and the milk were weighed daily. The butter fat in the milk was determined by the Babcock test. According to Palmer, the yellow color of corn is chiefly due to xanthophyll (18), which is not at present believed to be a precursor of vitamin A. The vitamin A activity of corn is, however, associated with the yellow color, since yellow corn has vitamin A potency and white corn has little or none. It may be present as vitamin A or as the small amount of carotene in the yellow corn.

The vitamin A potency of the feed was determined by means of rats by the modified Sherman-Munsell procedure already described in detail (7, 21). The results are expressed as rat units. (See Table 4.) The yellow corn in the first experiment contained about 6 units per gram, while that used in the second experiment contained only 2.5 to 3 units per gram. Butter was prepared from the milk at regular intervals during the period of experiment. The butter was melted, the fat separated, and kept in an electric refrigerator; the vitamin A was determined by the method referred to above.

Details of the Experiment with Low Amounts of Vitamin A

Two high-grade Jersey cows were selected which had just freshened and had been on good green pasture for some time. Cow No. 59 was 4 years old, weighed 709 pounds at the beginning of the experiment, and freshened April 2, 1932. Cow No. 61 was 3½ years old, weighed 730 pounds, and freshened March 9, 1932. Cow No. 59 was fed a mixture of 60 parts white corn, 36 parts cottonseed meal, 3 parts limestone, and 1 part salt at the rate of approximately 1 pound to 2½ pounds of milk. Cow No. 61 was fed in the same way, excepting that yellow corn was used in place of white corn. Both cows also received all the cottonseed hulls and beet pulp they desired to eat. Table 1 shows the quantities of yellow corn eaten

Table 1. Yellow corn eaten and milk and fat produced per day, at low levels of vitamin A

	Cow No. 61 yellow corn pounds	Cow. No. 59, white corn			Cow No. 61, yellow corn		
		Milk pounds	Fat per cent	Fat pounds	Milk pounds	Fat per cent	Fat pounds
April 5-11 inc., 1932	6.0	25.9	4.6	1.19	22.7	5.2	1.18
April 12-18 inc.	6.4	27.8	4.6	1.28	23.4	5.2	1.22
April 19-25 inc.	4.9	25.9	4.6	1.19	19.4	5.2	1.01
April 26-May 2 inc.	6.9	25.0	5.3	1.33	21.9	5.5	1.20
May 3-16 inc.	6.9	26.7	6.0	1.60	23.5	5.8	1.36
May 17-30 inc.	6.9	25.7	6.0	1.54	23.3	5.8	1.35
May 31-June 27 inc.	6.9	26.1	4.1	1.07	24.1	5.2	1.25
June 28-July 25 inc.	6.5	24.2	4.9	1.19	21.8	5.8	1.26
July 26-August 22 inc.	6.0	18.7	5.1	.95	17.7	6.5	1.15
August 23-September 19 inc.	6.0	16.9	5.7	.96	17.2	6.4	1.10
September 20-October 17 inc.	6.0	14.2	5.7	.81	14.3	6.6	.94

by Cow No. 61 and the quantity of milk, percentage of fat, and pounds of fat produced by both cows.

The experiment began April 5, 1932. Samples of butter were collected at the end of the first, second, third, and fourth week, then every 2 weeks for the second 4 weeks, and then every 4 weeks. The total time was equivalent to 7 periods of 28 days each. Details of the estimation of vitamin A in the butter fat are given in Table 5.

Details of the Experiment with High Amounts of Vitamin A

Three pure-bred Jersey cows were selected which had been on good green pasture. Cow No. 196 was nearly 7 years old, weighed about 1000 pounds

at the beginning of the experiment, and freshened February 28. Cow No. 322 was 4 years and ten months old, weighed about 900 pounds, and freshened April 18. Cow No. 329 was 4 years and 8 months old, weighed about 950 pounds, and freshened March 9. The experiment began April 22, 1933.

The grain mixture fed consisted of 60 parts yellow corn, 27 parts cottonseed meal, 10 parts wheat bran, 2 parts ground limestone, and 1 part salt. All three cows were fed all the cottonseed hulls they would eat. They were also fed about 20 pounds of wet beet pulp daily. Cow No. 322 received 3 pounds of heat-dried alfalfa meal daily and cow No. 329 was fed 6 pounds. Considerable difficulty was encountered in getting the cows to eat the alfalfa meal. It was thoroughly mixed with the beet pulp and the grain was spread over the mixture in small quantities at a time

Table 2. Yellow corn and alfalfa eaten at high levels of feeding—pounds per day

Period	Cow No. 196		Cow No. 322		Cow No. 329	
	Corn	Alfalfa	Corn	Alfalfa	Corn	Alfalfa
April 22-May 19 inc.	7.20	.00	7.09	2.77	7.16	5.82
May 20-June 16 inc.	7.20	.00	7.11	2.71	7.20	6.00
June 17-July 14 inc.	7.20	.00	7.20	3.00	7.20	6.00
July 15-Aug. 11 inc.	7.20	.00	7.14	2.89	7.14	6.00
Aug. 12-Sept. 8 inc.	6.60	.00	7.20	3.00	7.20	6.00

in order to get the cows to eat it. The grain mixture was fed at the rate of approximately one pound to 2½ pounds of milk. All three cows were kept in a lot which was kept free from all vegetation and in which water, block salt, and shelter were available.

The average quantities of corn and alfalfa meal eaten are given in Table 2. Table 3 gives the average production of milk and fat for each

Table 3. Production of milk and fat by cows fed yellow corn and alfalfa meal—average per day

	Cow No. 196 Yellow corn			Cow No. 322 Alfalfa, 3 pounds			Cow No. 329 Alfalfa, 6 pounds		
	Milk pounds	Fat per cent	Fat pounds	Milk pounds	Fat per cent	Fat pounds	Milk pounds	Fat per cent	Fat pounds
April 22-May 19 inc.	21.48	5.30	1.14	40.96	3.65	1.50	34.10	5.10	1.74
May 20-June 16 inc.	20.83	5.05	1.05	38.50	4.30	1.66	33.76	5.10	1.72
June 17-July 14 inc.	20.29	5.60	1.14	34.33	5.10	1.75	30.74	5.55	1.71
July 15-Aug. 11 inc.	17.45	5.25	.92	33.23	5.20	1.73	29.96	5.80	1.74
Aug. 12-Sept. 8 inc.	11.34	5.20	.59	30.99	5.05	1.57	27.40	5.75	1.58

cow. Table 4 shows the details of the estimation of the vitamin A in the feed.

Samples of the butter were collected when the period of feeding began and at intervals of 28 days. The details of the estimation of vitamin A in the butter fat are given in Table 8.

Relation of Vitamin A Activity of the Butter to the Feed and to the Stage of Lactation of the Cow

The units of vitamin A in the butter fat are shown in Table 5 and 6. Details of the estimation of the vitamin A in the butter fat are given in Tables 7 and 8.

With all five of the cows, the butter was high in vitamin A when the cows were fresh, containing 25 to 70 units per gram. It decreased in vitamin A with the length of the lactation period, or the period of time that the cows had been on the feed being tested. This is brought out in Figure 1.

As shown in Table 5, at the end of the first and third weeks the butter fat from the cows on the low levels of vitamin A contained practically the

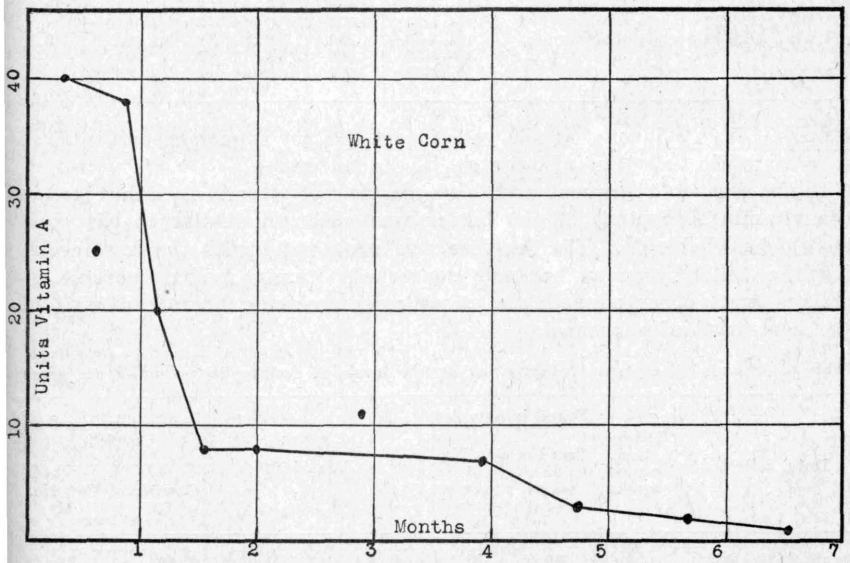

Fig. 1. The number of units of vitamin A potency in the butter fat of cow No. 59 fed white corn decreased with the length of the period of feeding. The dots show the amounts estimated in the samples collected.

normal amount of 38 units to the gram. For some reason the vitamin content of the butter from both cows decreased during the second week. At the end of the fourth week the butter fat contained 15 to 20 units per gram, which was a decided drop in the quantities of vitamin A. Another decided drop occurred at the end of 6 weeks with cow No. 59 (white corn) to 8 units and at the end of 8 weeks with cow No. 61 (yellow corn) to 7

units. After this the vitamin A decreased more slowly until at the end of 196 days the butter fat of the cow receiving no vitamin A contained only one unit, and the butter fat of the cow receiving yellow corn contained

Table 4. Details of estimation of vitamin A potency of feeds

Laboratory number	Feed tested	Date rat feeding begun	Grams fed per day	Number rats at beginning	Number rats at end	Average gain per rat in 8 weeks	Units vitamin A to one gram
36375	Ground yellow corn	5-27-32	.150	6	2	28	
"	" " "	6- 1-32	.200	6	2	20	6
"	" " "	6- 6-32	.250	6	4	37	
37675	Heat-dried alfalfa meal	6-23-33	.010	6	0	0	
"	" " " "	6-30-33	.013	6	0	0	
"	" " " "	6-30-33	.020	6	4	17	40
"	" " " "	10-16-33	.040	6	2	51	
37676	Yellow corn	6-19-33	.150	6	1	—3	
"	" "	6-23-33	.200	6	2	—10	
"	" "	6-23-33	.300	6	4	14	2.5
38509	Alfalfa meal	10- 4-33	.020	6	1	32	
"	" "	10- 4-33	.030	6	2	9	
"	" "	10- 2-33	.040	6	2	30	
"	" "	12- 8-33	.050	6	3	22	25
"	" "	12- 6-33	.070	6	3	51	
38510	Whole yellow corn	10- 4-33	.300	6	2	24	3
"	" " "	10- 6-33	.400	6	3	31	
"	" " "	10- 9-33	.500	6	4	46	

5 units. It is evident that the yellow corn did not supply enough vitamin A to maintain the vitamin A content of the butter.

In the second experiment with high amounts of vitamin A, a decrease in the vitamin A content of the butter was observed similar to that just mentioned (Table 6). The decrease was most rapid with the cow receiving the yellow corn as her sole source of vitamin A, but marked decreases were observed with the other cows receiving liberal amounts of the heat-dried alfalfa meal.

Table 5. Vitamin A content of butter fat at low levels of feeding in rat units per gram

Date collected	Depletion period weeks	Found in samples		Period	Assumed average for period	
		Cow No. 59 white corn	Cow No. 61 yellow corn		Cow No. 59	Cow No. 61
April 11-12, 1932	1	40	38	April 5-11	40	38
April 18-19, 1932	2	25	17	April 12-18	32½	27½
April 25-26, 1932	3	38	38	April 19-25	31½	27½
May 2-3, 1932	4	20	15	April 26-May 2	29	26½
May 16-17, 1932	6	8	12	May 3-16	14	13½
May 30-31, 1932	8	8	7	May 17-30	8	9½
June 27-28, 1932	12	11	6	May 31-June 27	9½	6½
July 25-26, 1932	16	7	5	June 28-July 25	9	5½
August 22-23, 1932	20	3	2	July 26-August 22	5	3½
Sept. 19-20, 1932	24	2	4	August 23-Sept. 19	2½	3
October 17-18, 1932	28	1	5	Sept. 20-October 17	1½	4½
November 2-3, 1932 (pasture)		32	25			

The butter from cow No. 322 fed 3 pounds of alfalfa meal daily was unusually high in vitamin A at the beginning of the test. This is perhaps due to the early stage of lactation at which the experiment was begun with this cow. This cow took the place of one originally selected, but which could not be used. The other two cows had been milking over a month when this cow calved. There was not much difference between the vitamin A of the butter fat of the other two cows at the beginning of the experiment, but none of the cows received sufficient quantities of vitamin A to maintain the vitamin A in the butter fat at its original content. That is, an average of about 116,000 units per day per cow was not sufficient for maintenance of the cows and for the production of butter high in vitamin A. The vitamin A content of butter depends both upon the vitamin A potency of the feed and the length of time the cow has been fed upon it. (See Table 6 and Figure 1.)

Where insufficient vitamin A is fed, the vitamin A in the butter fat decreases with the length of time the cow has been on the feed. The effect of the feed cannot be judged by tests covering short periods of time.

In the test comparing the yellow and white corn (Table 6), the cow on the white corn gave butter containing more vitamin A than the cow on the yellow corn in all the samples except the last three. A probable

Table 6. Vitamin A content of butter fat and of feed—units per gram. Second experiment

	Butter fat cow No. 196 yellow corn	Butter fat cow No. 322 alfalfa 3 pounds	Butter fat cow No. 329 alfalfa 6 pounds	Alfalfa meal	Corn
Found in samples indicated					
April 22,1933	25	70	33	—	—
May 20	8	14	20	—	—
June 17	8	14	12	40	2.5
August 12	4	10	11	—	—
September 9	3	7	10	25	3
November 4	—	10	10	—	—
Assumed average for each period					
April 27-May 19 inc.	17	42	27	46	3.1
May 20-June 16 inc.	8	14	16	43	2.8
June 17-July 14 inc.	7	13	12	40	2.5
July 15-August 11 inc.	6	11	12	37	2.4
August 12-Sept. 8 inc.	4	9	10	33	2.3

explanation of this is that the cow on the white corn had more stored vitamin A at the beginning of the experiment than the cow on the yellow corn. Great differences have been found in the storage of vitamin A by pullets (20) as measured by the time of their survival without vitamin A, and similar differences perhaps occur with cows.

In the first experiment, the effect of the vitamin A in the yellow corn on the vitamin A in the butter, as shown by the difference in vitamin A content of the butter from the two cows, was not apparent until the

Table 7. Details of estimation of vitamin A potency of butter fat from cows fed white corn or yellow corn

Laboratory number	Date collected	Cow number	Weeks after feeding begun	Butter fat fed per day grams	Number of rats at beginning	Number of rats at end	Average gain per rat in 8 weeks grams	Units vitamin A to one gram fat
36292	April 11-12, 1932	59	1	.027	6	4	32	40
				.053	6	6	52	
36293	April 11-12, 1932	61	1	.027	6	4	27	38
				.053	6	5	44	
				.053	6	6	41	
36302	April 18-19, 1932	59	2	.027	6	5	9	
				.040	6	5	27	25
				.053	6	5	49	
36301	April 18-19, 1932	61	2	.027	6	2	10	
				.040	6	4	16	
				.053	6	4	16	
				.066	6	3	34	17
36342	April 25-26, 1932	59	3	.027	6	4	29	38
				.053	6	6	47	
36343	April 25-26, 1932	61	3	.027	6	4	30	38
				.053	6	5	51	
36350	May 2-3, 1932	59	4	.027	6	0	—	
				.053	6	4	31	20
36349	May 2-3, 1932	61	4	.027	6	1	—5	
				.053	5	1	25	
				.070	6	5	29	15
36383	May 16-17, 1932	59	6	.027	6	2	—8	
				.053	6	3	18	
				.070	6	2	17	8
				.100	6	2	18	
38382	May 16-17, 1932	61	6	.027	6	1	—9	
				.053	6	3	16	
				.070	6	2	16	12
				.200	6	5	91	
36454	May 30-31, 1932	59	8	.027	6	2	—32	
				.053	6	3	—14	
				.100	6	2	28	
				.133	6	3	32	8
				.200	6	5	77	
36455	May 30-31, 1932	61	8	.027	6	1	—26	
				.053	6	2	—7	
				.100	6	3	22	7
				.133	6	0	0	
				.200	6	4	43	
36775	June 27-28, 1932	59	12	.100	6	4	31	11
				.200	6	6	50	
36776	June 27-28, 1932	61	12	.100	6	2	37	6
				.200	6	5	31	
36908	July 25-26, 1932	59	16	.100	6	1	19	7
				.200	6	4	44	
36909	July 25-26, 1932	61	16	.100	6	3	5	
				.200	6	3	26	5
37143	Aug. 22-23, 1932	59	20	.133	6	2	—3	
				.200	6	1	11	3
37142	Aug. 22-23, 1932	61	20	.133	6	0	—	2
				.200	6	0	—	
				.400	6	0	—	
37275	Sept. 19-20, 1932	59	24	.200	6	0	—	
				.250	6	1	9	2
				.400	6	3	10	
37276	Sept. 19-20, 1932	61	24	.200	6	3	42	
				.250	6	3	29	4
37330	Oct. 17-18, 1932	59	28	.200	6	2	—25	
				.400	6	1	—11	1
37331	Oct. 17-18, 1932	61	28	.200	6	4	29	5
				.400	6	5	59	
37363	Nov. 2-3, 1932	59	pasture 2 weeks	.033	6	4	30	32
				.050	6	6	47	
37364	Nov. 2-3, 1932	61	pasture 2 weeks	.033	6	3	14	25
				.050	6	4	39	

end of the sixth period of 28 days. During both the sixth and seventh periods the cow receiving yellow corn produced butter fat containing about twice as many units of vitamin A per gram as the cow receiving white corn. The number of units in the butter in both cases was, however, very low. The results here reported for butter are similar to those reported for eggs (20, 21), since the vitamin A content of the eggs decreased with the period of feeding whether or not yellow corn was fed, and the yellow corn had little effect upon the vitamin A in the eggs.

The cow on white corn was very weak at the end of the experiment but after the experiment was completed, when both cows were placed on green pasture she regained her strength rapidly. The large amounts of vitamin A consumed in the pasture plants affected the butter quickly (Table 7), as was shown in the increase from 1 up to 32 units per gram of butter fat of the cow previously fed on white corn, and from 5 up to 25 units per gram of butter fat of the cow previously fed the yellow corn. From these results it also appeared possible that the cow on white corn was superior to the cow fed on yellow corn in her ability to utilize vitamin A or carotene.

The vitamin A in the butter fat from the cow fed yellow corn decreased more rapidly in the second experiment than that in the butter fat from the other cows, and from the end of the first month on was about half as much as that in the butter fat from the other two cows. At the end of this experiment, the butter from the cow fed yellow corn alone contained 3 units a gram, somewhat less than the 5 units a gram found in the previous experiment with yellow corn, but the yellow corn fed was also poorer in vitamin A than that fed in the first experiment. This cow also suffered from night blindness which indicates that the yellow corn used did not supply enough vitamin A to maintain a lactating cow.

The experiment in which white corn alone was fed showed that vitamin A was at first supplied to the butter in large quantity by the reserves stored in the cow. As the reserves became used up, the vitamin A in the butter decreased. When the cow is on a feed supplying insufficient quantities of vitamin A, the vitamin A in the butter fat decreases with the period of time the cow has been on the feed or the stage of lactation, on account of the depletion of the reserve of vitamin A stored by the cow at the beginning of lactation. A herd of cows contains animals at various stages of lactation, so that the mixed butter of the herd would be more nearly constant in vitamin A content than that of a single cow. The high vitamin A content of the butter from the cows at an early stage of lactation could offset the low content of the butter fat from the cows at a late stage of lactation. In order to ascertain the effect of the feed, the breed of cows, or other factors upon the vitamin A content of the butter, it is necessary to examine the butter from cows at similar stages of lactation. The use of the mixed butter from cows at different stages of lactation introduces complications, as shown above.

Experiments covering short periods of a month or even more will not show the full effect of insufficient vitamin A on the vitamin A content of

Table 8. Details of estimation of vitamin A potency of butter fat from cows fed yellow corn and alfalfa meal—Second experiment

Laboratory number	Date collected	Cow number	Butter fat fed per day grams	Number of rats at beginning	Number of rats at end	Average gain of rats in 8 weeks, grams	Units vitamin A to one gram fat
37677	April 22, 1933	196	.027	6	4	11	
			.040	6	6	24	25
37678	April 22, 1933	322	.017	6	6	33	70
			.020	6	5	37	
			.027	6	6	40	
			.040	6	6	66	
37679	April 22, 1933	329	.027	6	5	21	33
			.040	6	4	37	
37699	May 20, 1933	329	.050	6	5	23	20
			.070	6	5	41	
37700	May 20, 1933	196	.040	6	1	0	
			.050	6	0	0	
			.070	6	4	11	
			.100	6	4	13	
			.133	6	3	21	8
37701	May 20, 1933	322	.030	6	0	0	
			.040	6	3	11	
			.050	6	4	13	
			.070	7	6	25	14
			.100	5	3	45	
37738	June 17, 1933	196	.070	6	0	0	
			.100	6	2	12	8
			.200	6	3	54	
37739	June 17, 1933	322	.050	6	4	—4	
			.070	6	5	21	14
			.100	6	2	15	
			.150	6	4	44	
37740	June 17, 1933	329	.050	6	1	24	
			.067	6	3	14	
			.100	6	6	30	12
			.150	6	6	44	
38160	August 12, 1933	196	.100	6	0	0	
			.200	6	2	10	4
			.400	6	4	35	
38161	August 12, 1933	322	.070	6	4	17	
			.100	6	3	26	10
38162	August 12, 1933	329	.050	6	1	0	
			.070	6	2	16	
			.100	6	3	29	11
38428	September 9, 1933	196	.100	6	0	0	
			.200	6	0	—3	
			.400	6	3	28	3
38429	September 9, 1933	322	.070	6	2	12	
			.100	6	2	32	
			.133	6	5	20	7
38430	September 9, 1933	329	.050	6	1	—13	
			.070	6	1	4	
			.100	6	2	23	10
			.133	6	2	31	
39075	November 4, 1933	322	.100	6	3	21	10
			.150	6	3	21	
39076	November 4, 1933	329	.100	6	5	24	10
			.150	6	4	11	

the butter, since the vitamin A of the butter may continue to decrease for several months.

Relation of Quantity of Vitamin A in the Feed to Quantities of Vitamin A in the Butter Fat

The quantities of vitamin A fed were calculated from the data already given. The determination of vitamin A in the yellow corn used in the

first experiment was made near the middle of the experiment, and for this reason the yellow corn used was assumed to average the 6 units per gram found. In the second experiment, allowance of 7 per cent a month (10, 21) was made for the loss of vitamin A due to storage of the yellow corn and heat-dried alfalfa meal. The estimated content used in the calculations is given in Table 6. For the butter fat, the quantities of vitamin A in the first experiment were those given in Table 6. Since in the second experiment the samples were taken at the beginning and end of each period, the units used in the calculation of the quantities in the butter fat were the mean of these two samples as shown in Tables 5 and 6.

In Table 9, the number of units of vitamin A fed per day in the first experiment is compared with the number of units in the butter. The

Table 9. Total units vitamin A in feed and in butter per cow per day—low levels of feeding

Period	Days	In feed		In butter		Percentage in butter of that in feed	
		Cow No. 59 white corn	Cow No. 61 Yellow corn	Cow No. 59 white corn	Cow No. 61 Yellow corn	Cow No. 59	Cow No. 61
April 5-11	7	0	16330	21587	20335	——	124.5
April 12-18	7	0	17418	18866	15215	——	87.4
April 19-25	7	0	13336	17000	12596	——	94.5
April 26-May 2	7	0	18779	17492	14424	——	76.8
May 3-16	14	0	18779	10158	8326	——	44.3
May 17-30	14	0	18779	5587	5816	——	31.0
May 31-June 27	28	0	18779	4610	3685	——	19.6
June 28-July 25	28	0	17690	4858	3143	——	17.8
July 26-August 22	28	0	16330	2154	1825	——	11.8
Aug. 23-September 19	28	0	16330	1088	1497	——	9.2
Sept. 20-October 19	28	0	16330	551	1919	——	11.8

percentage that the vitamin A in the butter is of that in the feed is also given. It varies from 9.2 to 124.5 per cent. This might be taken for the apparent recovery, except for the fact that cow No. 59, which received practically no vitamin A in her feed, produced for five months butter containing more vitamin A than cow No. 61, which received yellow corn as a source of vitamin A. This means vitamin A was put in the butter from that stored in the body of the cow and that the apparent recovery shown is not the real recovery. As shown elsewhere (21), hens also put vitamin A in the eggs from the vitamin A stored in their bodies.

The total number of units eaten per day in the second experiment at high levels of vitamin A are given in Table 10. On an average, cow No. 196 consumed 8432 units of vitamin A per day, cow No. 322, 60189 units per day, and cow No. 329, 116,101 units per day.

In Table 11 are given the units of vitamin A in the feed eaten and in the butter produced, as well as the percentage in the butter of that in the feed. This percentage varies from 16 to 87 per cent with cow No. 196, fed yellow corn alone, 12 to 42 per cent for the cow fed yellow corn

and 3 pounds alfalfa meal daily, and 7 to 16 per cent with the cow fed yellow corn and 6 pounds of alfalfa meal daily. The percentages of the vitamin A fed which we found in the butter are much higher than those reported for carotene by Baumann et al (2). Their calculation from data secured with the spectroscope indicated that the vitamin A and carotene

Table 10. Total units of vitamin A eaten per day—high levels of feeding

Period	Cow No. 196	Cow No. 322			Cow No. 329		
	In corn	In corn	In alfalfa	Total eaten	In corn	In alfalfa	Total eaten
April 22-May 19	10124	9970	57798	67768	10068	121438	131506
May 20-June 16	9145	9030	52858	61888	9145	117029	126164
June 17-July 14	8165	8165	54432	62597	8165	108864	117029
July 15-August 11	7838	7773	48503	56276	7773	100699	108472
August 12-Sept. 8	6886	7512	44906	52418	7512	89813	97325
Average per day	8432	8490	51699	60189	8533	107569	116101

in the butter were only 3.3 per cent of the carotene fed on a low carotene ration and 1.3 per cent of the carotene fed on a high carotene ration. Our percentage are also higher than those found by Russell (19), who states that less than 5 per cent of the vitamin A value in the amount of dried

Table 11. Comparison of vitamin A in feed with that in butter—high levels of feeding

	Units eaten per day			Units in butter per day			Percentage in butter of that in feed		
	Cow No. 196	Cow No. 322	Cow No. 329	Cow No. 196	Cow No. 322	Cow No. 329	Cow No. 196	Cow No. 322	Cow No. 329
April 22-May 19	10124	67768	131506	8791	28577	21310	87	42	16
May 20-June 16	9145	61888	126164	3810	10542	12483	42	17	10
June 17-July 14	8165	62597	117029	3620	10319	9308	44	16	8
July 15-August 11	7838	56276	108472	2504	8632	9471	32	15	9
August 12-Sept. 8	6886	52418	97325	1071	6409	7167	16	12	7
Calculated for period	.....	.....	.....	.....	.....	.....	47	21	10
Average	8432	60189	116101	3959	12896	11948			

alfalfa and corn silage usually fed appears in the milk of dairy cattle. The previous feed of the animals may have had something to do with these differences.

As with hens, we found the apparent percentage recovery to be highest with feed lowest in vitamin A. The apparent recovery decreases as the period of lactation advances. Since, as shown above, some of the vitamin A comes from the body of the cow, the recovery is only apparent, and is not the actual recovery.

Quantities of Vitamin A Required in the Feed over Maintenance Requirements for a Unit of Vitamin A in the Butter

No previous work has shown the units of vitamin A required in feed for a unit in the butter in addition to maintenance requirements. In experiments with hens, Sherwood and Fraps (20, 21) have shown that the vitamin A in the feed required for one unit in the eggs, in addition to the maintenance requirements, appeared to depend to some extent upon the quantity fed. One unit in the eggs required 6.3 units in the feed when 270 units a day were fed, 5.5 units when 336 units were fed, and 4.0 units when 444 units were fed daily. A probable explanation of the high requirements at the lower levels of feeding is that some of the vitamin A was used for maintenance.

The amount of vitamin A in the feed needed over maintenance requirements for the vitamin A in butter could not be calculated from the first experiment, since the cow fed yellow corn produced less vitamin A than the cow fed white corn. It was calculated from the second experiment, in which alfalfa meal was fed, with the results given in Table 12. The results are calculated for each period of 28 days. If cows No. 196 and No. 322 are compared, it is found that cow No. 322 on an average consumed daily

Table 12. Units of vitamin A potency required for units of vitamin A in butter fat

	Apr. 22 May 11	May 20 June 16	June 17 July 14	July 15 Aug. 11	Aug. 12 Sept. 8	Average
Found in butter fat, Cow 322	28577	10542	10319	8632	6409	12896
Found in butter fat, Cow No. 196	8791	3810	3620	2504	1071	3959
Excess in butter fat over Cow 196	19786	6732	6699	6128	5338	8937
Total fed, Cow 322	67768	61888	62597	56276	52418	60189
Fed in corn, Cow 196	10124	9145	8165	7838	6886	8432
Excess in feed over Cow 196	57644	52743	54432	48438	45532	51758
Units in feed for one unit in fat	3	8	8	8	9	6
Found in butter fat, Cow 329	21310	12483	9308	9471	7167	11948
Found in butter fat, Cow 196	8791	3810	3620	2504	1071	3959
Excess in fat of Cow 329 over Cow 196	12519	8673	5688	6967	6096	7989
Total fed, Cow 329	131506	126164	117029	108472	97325	116101
Fed in corn, Cow 196	10124	9145	8165	7838	6886	8432
Excess in feed Cow 329 over Cow 196	121382	117019	108864	100634	90439	107669
Units in feed for one unit in fat	10	13	19	14	15	13
Excess in fat of Cow 329 over Cow 322	—7267	1941	—1011	839	758	—948
Excess in feed of Cow 329 over Cow 322	63738	64276	54432	52196	44907	55910
Units in feed for one unit in fat	.....	33	.....	62	59	.....

51,758 more units of vitamin A and produced daily 8937 more units of vitamin A in the butter, so that one unit of vitamin A required on an average 6 units in the feed. If cows No. 196 and 329 are compared, it is found that Cow No. 329 consumed daily 107,669 more units of vitamin A and produced 7,989 more units in the butter, so that one unit of vitamin A in the butter required 13 units in the feed. If cows No. 322 and 329 are compared, it is seen that while cow No. 329 consumed more vitamin A in the feed, she produced on an average less vitamin A in the butter. This excess was produced during the first and third periods, but in the other

two periods, her production of vitamin A was out of proportion to that of the others. This might be explained on the supposition that cow No. 322 had stored more vitamin A than the other two cows but the fact that this cow was placed on experiment at an earlier stage of lactation than the other cows, may have something to do with it. This cow may also have greater ability to utilize vitamin A than the other cows.

If the individual periods are examined, we find a remarkable uniformity in the results for four of the five months with the first comparison, and three of the five months with the second comparison. The average of the four uniform months in the first comparison would be 8 units in the feed over maintenance for one unit in the butter, whic his much higher than the average for the entire period on account of the low figure secured for the first month. This average of 8 is probably more nearly correct than the average of 6 secured from all the periods. The average of the three uniform months in the second experiment is 14 units in the feed over maintenance for one unit in the butter, which is only a little higher than the 13 units which is the average for the entire period.

On account of the probable variations in the vitamin A stored in the bodies of the different cows, the differences in the stage of lactation at which the cows were placed on experiment, and the other causes of variation, some uncertainty is attached to the figure for the number of units of vitamin A required for a unit in the butter, after allowing for maintenance. From the consideration of all the details discussed above, it appears that on an average of the two comparisons, one unit in the butter requires approximately eleven in the feed.

Since one unit of vitamin A in the eggs requires 4 to 6 units over maintenance in the feed for hens, while one unit in the butter of cows requires approximately 11 units in the feed over maintenance, it is evident that the cow utilizes vitamin A much less efficiently than the hen.

The feed o fthe cow must be high in vitamin A in order for the animal to continue to produce butter fat high in vitamin A. The butter fat may be high in vitamin A at the beginning of the lactation period, but unless the feed is very high in vitamin A, the butter will decrease in vitamin A potency during the course of the lactation period. Silage and ordinary hays and fodders apparently will not supply sufficient quantities of vitamin A to maintain the vitamin A content of the milk. Green and growing pasture grasses furnish the abundant supply of vitamin A that is needed.

Vitamin A Requirements for Butter Fat High in Vitamin A

It is shown in the experiments here reported that 6 to 7 pounds of yellow corn, containing 6 to 2.5 units of vitamin A per gram and 6 pounds of heat-dried alfalfa, containing about 25 to 40 biological units per gram, did not supply sufficient vitamin A to maintain a high vitamin A potency of butter. The quantities of vitamin A in various hays and fodders have not been estimated, but ordinary alfalfa meal may contain 7 to 20 units per gram (7). A sample of sorghum silage

contained only 5 units per gram (5). The vitamin A content of grasses seems to decrease considerably during the process of curing. It is improbable that other hays would contain more vitamin A than alfalfa hay while fodders such as corn fodder or various straws, would probably be much lower in vitamin A than alfalfa. Since commercial alfalfa hay ordinarily will probably contain much less vitamin A than the heat-dried alfalfa meal used in these experiments, and since both the alfalfa meal and the yellow corn were fed in liberal amounts, it is safe to say that the rations ordinarily fed dairy cows would not contain enough vitamin A to maintain the vitamin A potency of the butter, unless supplemented by green growing grasses or good pasture.

SUMMARY

The vitamin A potency of butter fat decreased during the experimental period. With a cow receiving about 17,000 units in 6 to 7 pounds daily of yellow corn, the vitamin A in the butter decreased from about 38 rat units per gram at the beginning of the test to about 16 units in 4 weeks and 5 units at the end of 5 months. With a cow receiving about 7 pounds of corn and 6 pounds of heat-dried alfalfa meal, supplying 116,000 units daily, it decreased from 33 units per gram at the beginning of the test to 20 units in 4 weeks, and 12 units in 8 weeks, after which it remained at about 11 to 12 units for about 5 months.

When a cow is on a feed containing insufficient amounts of vitamin A, the vitamin A potency of the butter will depend upon the length of time the ration has been fed as well as the vitamin A potency of the ration.

Feeding 116,000 units of vitamin A per day to the cow was not sufficient to maintain the vitamin content of butter fat. This number of units is probably greater than that ordinarily supplied in hays and fodders, since they are usually much lower in vitamin A potency than the heat-dried alfalfa meal used in these experiments.

When two cows producing butter fat containing 1 and 5 units of vitamin A per gram, respectively, were placed on pasture, the vitamin A content of the butter increased to 35 units and 25 units, respectively, within two weeks. The cows were able to utilize the vitamin A and carotene of the pasture grass sufficiently to produce butter fat high in vitamin A activity even towards the end of the lactation period.

The average apparent percentage of the vitamin A in the feed recovered in the butter fat was 47 per cent for a cow receiving daily 8432 units of vitamin A in about 7 pounds of yellow corn, 21 per cent for a cow receiving 60,189 units of vitamin A in about 7 pounds of yellow corn and 3 pounds of alfalfa, and 10 per cent for a cow receiving 116,101 units of vitamin A daily in about 7 pounds of yellow corn and 6 pounds of heat-dried alfalfa. The apparent percentage recovered is not the actual recovery, since some of the vitamin A in the butter comes from that previously stored in the body of the cow.

A cow fed several months on a ration practically free from vitamin A stored more vitamin A in her butter fat than a cow receiving about 17000 units daily from 6 to 7 pounds of yellow corn; a probable explanation of this fact is that the vitamin A stored in the body of the first cow was higher than that stored by the second. Consumption of 17,000 units of vitamin A daily was not sufficient to maintain the health of the cow.

In a comparison of two cows in five periods of 28 days each, 8 units in the feed over maintenance requirements were required for one unit in the butter, while in another similar comparison, 14 units in the feed were required for one unit in the butter. In a third comparison in two of the five periods, the cow receiving more vitamin A produced less vitamin A in the butter fat than the cow receiving the smaller amount of vitamin A.

On account of the small numbers of cows used in these experiments and individual variations in the amounts of vitamin A stored in the body of these cows, the estimation of the units of vitamin A required for one unit in the butter was somewhat uncertain. Differences in the stage of lactation and probable differences in the powers of individual animals to utilize carotene and vitamin A also affected the results. On an average, one unit in the butter required approximately 11 units in the feed over maintenance. This is much higher than the 4 to 6 units over maintenance required by hens for the production of one unit in the eggs. The cow seems to use vitamin A or carotene less efficiently than the hen.

In order to ascertain the effect of the feed, the breed of cow, or similar factors upon the vitamin A content of the butter, it will be necessary to examine the butter from cows at similar stages of lactation, so as to allow for the effect of the stage of lactation of the cow. The difference in the vitamin A potency of butter at different stages of lactation is probably due to the depletion of the vitamin A stored in the body of the cow at the beginning of lactation.

The feed of cows must be high in vitamin A potency in order for the animal to continue to produce butter high in vitamin A potency. Silage and ordinary hays and fodders apparently will not supply enough vitamin A potency to maintain a high content of the butter fat. Green growing pasture grasses appear to be needed to maintain the production of butter fat high in vitamin A.

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