SOME FACTORS INFLUENCING THE STORAGE OF VITAMIN A IN FATTENING LAMBS

SOME FACTORS INFLUENCING THE STORAGE OF VITAMIN A IN FATTENING LAMBS

By

JAMES O. TUCKER

Bachelor of Science

Oklahoma Agricultural and Mechanical College

Stillwater, Oklahoma

1947

Submitted to the Department of Animal Husbandry

Oklahoma Agricultural and Mechanical College

In Partial Fulfillment of the Requirements

for a Degree of

MASTER OF SCIENCE

1948

APPROVED BY:

H. M. Briggs
Chairman, Thesis Committee

A. E. Darlow
Head of the Department

D. C. McIntosh
Dean of the Graduate School

ACKNOWLEDGEMENT

The sincere thanks of the author are extended to Dr. H. M. Briggs of the Animal Husbandry Department of Oklahoma Agricultural and Mechanical College for his time and assistance in making this study possible.

The author is also indebted to Dr. W. D. Gallup of the Department of Agricultural Chemistry of Oklahoma Agricultural and Mechanical College for guidance in performing various chemical analyses and for other valuable assistance.

TABLE OF CONTENTS

INTRODUCTION

Investigators are studying various nitrogen concentrates as protein supplements in the rations of cattle and sheep. The question has arisen as to the influence some of these nitrogenous compounds, particularly urea, might have when used to replace common forms of preformed protein. This station has been interested in knowing if the addition of urea would alter the vitamin A level of blood plasma and the storage of vitamin A in the liver.

Most roughages commonly fed sheep contain appreciable amounts of vitamin A or its precursor, carotene. Furthermore it is hard to deplete the vitamin A storage from the livers of sheep. Hauge et. al. (1940) reported that soybean meal seems to interfere with the utilization of vitamin A in dairy cattle. It was considered possible that soybean meal might also influence the utilization of vitamin A in sheep. It was thought urea might have some effect on the utilization and storage of vitamin A in rumminants, since the utilization of non-protein nitrogen is apparently directly associated with the flora of the digestive tract of such animals. An alteration of the flora might change the efficiency with which the animal uses various nutrients.

REVIEW OF LITERATURE

Vitamin A or carotene, its precursor, has been demonstrated by many workers to be necessary for the growth and development of most animals. Moore (1929) states that there are at least four carotenes containing vitamin A activity. These are the alpha, beta, gamma and hydroxy-beta-carotenes; the beta form is twice as effective as the other three forms in supplying the requirements of the animal body of this essential vitamin.

Jones et al. (1943), Maynard (1947), Erb et al. (1947) and others, working with cattle, and Hart (1940), working with sheep and cattle, point out many symptoms of vitamin A deficiency. Night blindness is the first symptom usually observed, while watering of the eyes, nasal discharge, sluggishness, and even total blindness may follow.

The minimum requirements of carotene and vitamin A for growth and the prevention of the various vitamin A deficiency symptoms have been estimated by various workers. Ward and co-workers (1940) found that eleven micrograms of carotene per day per pound of body weight was sufficient for growing calves. Lewis and Wilson (1945) reported 32 U.S.P. Units of vitamin A per kilogram body weight were sufficient for growth, but not for liver storage in the calf. Kuhlman and Gallup (1942) found that from forty to forty-five micrograms of carotene per pound of body weight per day were required by dairy cows for normal reproduction. Boyer et al. (1942) obtained some growth even after the animal was depleted to a stage where it showed common symptoms of avitaminosis A. Guilbert et al. (1937)(1940) estimated the minimum vitamin A requirement of cattle at 5.1-6.4

micrograms per kilogram of body weight and for sheep 4.3 to 6.3 micrograms per kilogram of body weight. Peirce (1945) reported that the minimum estimated by Guilbert, et al. brought about recovery in only one of three instances of night-blindness in sheep.

Varied lengths of time have been necessary to deplete the animal of vitamin A storage. Guilbert, Miller and Hughes (1937) reported that under the conditions of their experiment it took twenty-two to twenty-three months to deplete four seven-year-old ewes of vitamin A stores to a point where they showed symptoms of night-blindness. Riggs (1940) worked with range cattle and found that young cattle were more easily depleted of their vitamin A storage than were older animals. He compared four different age groups and found that sixteen-month-old steers required 109 to 268 days, six- to eight-month-old steer calves required 75 to 208 days, four- to six-month-old heifer calves required 75 to 128 days, and three- to five-month-old steer calves required 45 to 74 days for the depletion of their vitamin A storage. Jones et al. (1943) reported that the depletion time for range cattle varying in age from three to sixteen months was 45 to 268 days. Guilbert and Hart (1934), Riggs (1940), Jones (1943), and many others agree that younger animals are more easily and completely depleted than are older animals.

Hoefer and Gallup (1947) reported that some sources of vitamin A were superior to others when fed to lambs. Alfalfa meal was found to be superior to a carotene concentrate from carrots, and fish liver oil superior to alfalfa meal in supplying vitamin A to lambs. They also confirmed the work of Hibbs and Krauss (1946) in showing there is little direct relationship between values of blood plasma vitamin A and liver vitamin A when the blood plasma values are above twenty micrograms percent. Investigations by Ward and co-workers (1940) and Lewis and Wilson (1945) have shown that additional vitamin A is without effect on the gaining ability of sheep that have appreciable body reserves of this vitamin. These conclusions were confirmed by Hoefer and Gallup (1947).

Glover et al. (1946) reported that in rats the vitamin A levels of the blood plasma are proportional to the free vitamin A in the liver and not to the total liver storage. They advanced the theory that some mechanism, as yet obscure, tends to maintain the vitamin A level in the blood by keeping the free vitamin A in the liver constant. Their work indicated that the percentage of total storage existing as free vitamin A was much less when storage was large than when it was negligible. When the total storage was 13,500 I.U. per gram percent there was five percent free vitamin A, but when the total storage was 4 I.U. per gram percent there was sixty-five percent free vitamin A.

Draper and Evans (1944) reported that soybean oil meal alone had a higher gross value, when fed as a protein supplement to chicks, than any combination of soybean meal and cottonseed meal. When cottonseed meal was used as the sole protein supplement, growth was very slow and the chicks appeared listless. Philips and co-workers (1920) had earlier reported that chicks made comparatively better gains on soybean meal than did mammals.

Mitchell and Smuts (1932) found soybeans were too deficient in cystine to promote satisfactory growth in rats. However, Hayward, Steenbock and Bohstedt (1936) showed that although cystine might be a limiting amino acid in raw soybeans, this was not due to an actual deficiency but rather to a lack of availability of cystine or its equivalent. Heating the beans seemed to correct the cystine deficiency. Hayward et al. (1936) found that raw soybeans and those heated at low temperatures had less nutritional value for rats than those meals prepared commercially at high temperatures. Almquist et al. (1942) found no increase in rate of gain in baby chicks when l-cystine was added to the ration, but did find methionine to be the principal growth--limiting deficiency in raw soybean protein. Csonka and Jones (1934) reported that the percentage of cystine, tryptophane, and tyrosine varied in different varieties of soybeans.

Draper and Evans (1944) reported that different commercial soybean oil meal samples varied in nutritive value, perhaps due to differences in heat treatment. Johnson et al. (1939) suggested that soybeans contain some sulphur and nitrogen complex which can be absorbed but cannot be used for building tissue. They also reported that heat treatment of soybeans corrected this condition. Wilgus, Norris and Hensen (1936) indicated that high quality protein for feeding poultry could be produced from soybeans by the expeller, hydraulic or solvent processes. Bethke and Sweet (1939) reported similar results from feeding chickens expeller and toasted solvent-extracted soybean oil meals.

Hauge, Hilton and Wilbur (1940) demonstrated that a vitamin A supressing factor (thermo-stable), found in soybeans and soybean oil, interfered with the transference of vitamin A activity of feed to the butter fat secreted by dairy cows. They reported that this factor could be removed from the soybean oil by activated carbon.

Armsby (1911) suggested that micro-organisms found in the rumen of polygastric animals were able to synthesize protein from non-protein sources. Flingerling et al. (1937) demonstrated that urea could be used as a non-protein nitrogen supplement. Goss (1943) suggested that bacteria and protozoa may synthesize useful protein from non-protein sources such as urea and ammonium salts. Pearson and Smith (1943) found that the first step in protein synthesis of urea was the formation of NH_3, and the synthesis of such protein was reduced when the concentration of NH_3 became too high. They reported that several small feedings of urea per day were more completely utilized than one large feeding. Work at the Oklahoma Station by Darlow et al. (1946) indicated big steers did not make effecient use of urea when it was fed on alternate days under range conditions. Briggs et al. (1946) obtained more complete utilization of urea nitrogen by beef cattle, when fed twice daily than when the same total amount was fed on alternate days. Frequency of feeding did not appear to influence the utilization of the nitrogen from cottonseed meal.

Wegner and co-workers (1941) (1943) found that the protein content of the rumen ingesta showed a decided increase when the level of protein in the concentrate fed was increased to twenty-four percent. However, the rate of conversion of urea nitrogen to protein in the rumen decreased as the level of protein in the rumen ingesta became greater than twelve percent.

Hart et al. (1939) used urea in feeding growing dairy calves. They reported that when urea nitrogen furnished forty-three percent of the required nitrogen for growth the gains were only slightly less than gains made when casein protein supplied sixty-six percent of the required nitrogen.

Owen, Smith, and Wright (1943) conducted an experiment with dairy cattle to study the utilization of urea in milk production. Each animal received a ration for four weeks in which blood meal supplied one-third of the required nitrogen. The blood meal was then replaced by its nitrogen equivalent of urea plus its energy equivalent of pure starch. They found that the milk yield was maintained when urea replaced blood meal. Rupel, Bohstedt, and Hart (1943) compared urea and linseed meal as a source of supplemental nitrogen for dairy cattle and found no significant difference in the two for milk production. Willet et al. (1946) found dairy cows were able to utilize nitrogen from urea in the production of milk, but apparently not as efficiently as the nitrogen derived entirely from plant sources. When urea was used as the supplement and fed at daily intakes of either 0.48 pounds or 0.24 pounds milk yields were decreased. These amounts provided thirty-six percent and nineteen percent, respectively, of the total crude protein equivalent intake.

Cullison (1944) compared two types of silage for wintering cattle. He filled one silo in the regular manner and in the other urea was added, as the silo was being filled, at the rate of ten pounds per ton of silage. The animals which received treated silage maintained their weight through the winter satisfactorily, while those which received untreated silage lost an average of forty-seven pounds per cow during the winter period.

Harris and Mitchell (1941) reported that the addition of urea to a low nitrogen ration, that in itself was unable to support appreciable growth in lambs or consistently maintain nitrogen equilibrum, converted it into a ration capable of promoting a normal or nearly normal rate of growth. Loosli and Harris (1945) reported that lambs stored significantly more nitrogen on diets which contained urea plus methionine than on diets which contained only urea. Lofgreen and associates (1947) confirmed this report.

Briggs and co-workers (1947) reported that pellets containing approximately 25 percent of their nitrogen as urea have given as satisfactory performance as cottonseed meal when fed to yearling heifers on the range. Further work by Briggs and associates (1948) showed that the same type of pelleted feeds permitted about the same storage of nitrogen as cottonseed meal when fed to lambs. They also found that the addition of urea alone, as "Two-Sixty-Two", to a basal ration of low protein prairie hay increased the apparent digestibility of hay nutrients and changed nitrogen balances from slightly negative values to slightly positive values.

Axelsson (1942) reported that urea fed alone had a toxic effect and suggested that it was better to feed it in the presence of some carbonaceous material such as molasses or beet pulp. Mills et al. (1944) obtained better utilization of urea in heifer calves by the addition of molasses and starch. They reported no toxic effects from urea. Briggs et al. (1947) reported that no results indicating toxic effects of urea were obtained in studies with cattle although urea was fed as a source of nitrogen at relatively high levels. Dinning (1948) reported results which indicated large concentrations of urea, when given to cattle, are toxic. However the toxic effects obtained were brought about by administrating urea as a drench. When urea was added to the ration in as large amounts as the animal would consume it exhibited no toxic effects.

OBJECTIVES OF THE EXPERIMENT

This experiment was designed to study: (1) the influence of the level of vitamin A intake upon its concentration in the blood and storage in the liver of lambs, (2) the influence of soybean meal as compared to cottonseed meal in the effeciency of the utilization of vitamin A, (3) the influence of dietary urea on the utilization of vitamin A, (4) the influence of source of supplemental nitrogen and vitamin A level on the rate and efficiency of gain of fattening lambs.

EXPERIMENTAL PROCEDURE

(a) Depletion Period

Forty-four head of mixed, native, feeder lambs were secured on January 21, 1947, and placed on a vitamin A deficient ration of fifty pounds cottonseed hulls, thirty pounds oats, and twenty pounds cottonseed meal. The chemical compositions of these feeds are shown in Table 1. The lambs were "worked" to full feed in about two weeks. A mineral mix of equal parts of salt, ground limestone and bone meal was kept before the lambs at all times during the pre-experimental and the experimental periods. Some lambs were docked soon after arrival and all lambs were treated with phenathiozine at the beginning of the experiment. After February 22, each lamb was fed individually and weighed every two weeks. Weight and feed consumption records were kept throughout the experiment.

All lambs were depleted of vitamin A for a period of 115 days. The lambs were kept in dry lot and did not have access to green material of any kind. One lamb died, of undetermined cause, and three ewe lambs were removed from the experiment because of pregnancy. Eight lambs were sacrificed at the beginning of the experiment and the plasma vitamin A and liver storage of these lambs are shown in Table 2. The average daily gain for the last days of the depletion period was 0.25 pounds per lamb.

(b) Experimental

The design of this experiment was the 2x2x2 factorial as shown in Table 3. The lambs were alloted on May 15, 1947, after a consideration of their previous weight gains, body weight, and plasma vitamin A levels. The ration fed each lot is shown in Table 4. The lambs were individually fed.

The vitamin A was administered at two different levels, these being $1\frac{1}{4}$x and 10x the estimated minimum requirement. The minimum requirement was estimated from the work of Guilbert et al. (1937) (1940), and 5.7 micrograms per pound body weight per day was used as the minimum. The oil used was Borden's vitamin A oil concentrate with a potency of 40,000 U.S.P. units per gram. The oil was fed in capsules twice weekly with two weeks supply (four capsules) being made up each time. The weight of the animal at each weighing determined the amount of vitamin A that lamb would receive the following two weeks. The average vitamin A intake per day is shown in Table 5.

The blood was analysed for vitamin A three times during the experiment, at the beginning, after fifty days, and at the end of the experiment. To facilitate these blood collections the wool was sheared from the neck of each lamb to expose the jugular vein. The vein was blocked by the use of a short piece of 10 mm. rubber tubing. The blood was collected in citrated tubes and analysed for vitamin A by a modification of the Kimble (1939) procedure.

The lambs were taken to the packing house and slaughtered on August 20. Approximately one-half of the lambs broke at the break joint. The livers were collected at this time and cooled. A representative sample was then taken and wrapped in celophane and waxed paper. Due to the lack of satisfactory chloroform at this time the samples were kept frozen until the vitamin A analysis was made on October 28, 1947. The method used for this analysis of liver for vitamin A was that of Gallup and Hoefer (1946).

TABLE 1.

THE CHEMICAL COMPOSITION OF FEEDS USED IN FEEDING LAMBS

Description	Date	H_2O	Ash	Protein	Fat	Fiber	N.F.E.
		%	%	%	%	%	%
Dupont's "262"	4-23-47		2.65	262.50			
Cottonseed Meal	4-23-47	7.85	6.92	37.50	7.53	10.15	30.05
Soybean Meal*	5-15-47	9.85	5.41	43.50	4.75	4.69	31.80
Cottonseed Hulls	4-23-47	12.41	2.24	3.25	1.41	36.08	44.61
Oats	4-23-47	9.23	3.86	11.15	5.07	12.25	58.44

* The soybean meal had a very low urease activity.

TABLE 2.

PLASMA VITAMIN A LEVELS AND VITAMIN A LIVER STORAGE OF PRE-EXPERIMENTAL LAMBS

Lamb No.	Ration	Plasma Vit. A. ug/100 ml.	Liver Vit. A. ug/100 gm.	Wt. of Liver In gms.	Total Liver Storage
1	Basal	22.9	4,320	482	20,822
2	"	23.5	13,560	454	61,562
15	"	22.3	5,887	454	26,727
16	"	24.0	5,438	397	21,589
20	"	24.3	4,950	539	26,681
30	"	24.0	9.045	397	35,909
40	"	24.2	5,295	482	25,522
42	"	24.3	1,320	482	6,362
Average		23.7	6,227	461	28,147

TABLE 3.

TREATMENT OF LAMBS IN THE EXPERIMENT TO DETERMINE THE INFLUENCE OF THE SOURCE OF PROTEIN ON VITAMIN A OF BLOOD PLASMA AND VITAMIN A STORAGE OF THE LIVER

	Plant Protein Group		Urea Group	
Vitamin Level	Cottonseed Meal Ration I	Soybean Meal Ration II	Cottonseed Meal and Urea Ration III	Soybean Meal and Urea Ration IV
	LOT 1 Lamb No.	LOT 3 Lamb No.	LOT 5 Lamb No.	LOT 7 Lamb No.
1½x minimum requirements of vitamin A	14 19 24 25	6 10 13 41	8 17 23 38	9 21 37 44
	LOT 2 Lamb No.	LOT 4 Lamb No.	LOT 6 Lamb No.	LOT 8 Lamb No.
10x minimum requirements of vitamin A	3 11 35 39	7 28 29 33	4 22 26 27	5 18 36 43

TABLE 4.

THE RATIONS FED TO THE VARIOUS LOTS OF EXPERIMENTAL LAMBS

Feeds	Ration Number I lbs.	II lbs.	III lbs.	IV lbs.
Cottonseed Hulls	48	52	50	49
Oats	30	30	38.5	39.5
Cottonseed Meal	22	—	10	—
Soybean Meal	—	18	—	8
Urea (Dupont's "Two-Sixty-Two")	—	—	1.5	1.5

EXPERIMENTAL RESULTS AND DISCUSSION

After all lambs had been on a vitamin A deficient ration for 115 days they were bled and vitamin A content of the blood plasma determined by a modification of the Kimble (1939) procedure. The results of these analyses are shown in Table 5. The amount of vitamin A present in the plasma of the lambs at the end of the depletion period varied from 8.1 micrograms per 100 milliliters to 34.6 micrograms per 100 milliliter of plasma. The average was 23.9 micrograms per 100 milliliter of plasma. These analyses, previous gain, and the weight of the lambs were considered in alloting the lambs into the experimental groups for further feeding.

Eight lambs were sacrificed immediately after the initial bleeding while they were still being fed the basal ration. The average plasma vitamin A level of these eight lambs was 23.7 micrograms per 100 milliliters of plasma. The individual plasma vitamin A levels of the lambs sacrificed are given in Table 2. Analyses were made of the livers of these lambs to determine the storage of vitamin A, and they were found to have an average of 6,227 micrograms per 100 grams of liver.

All of the lambs in the experiment were again bled on July 3. The blood plasma levels at this bleeding showed a rise in vitamin A levels as compared to the initial bleeding. The lambs receiving vitamin A at 1½x the minimum requirement showed only slight increases while those receiving 10x the minimum, in most cases, showed marked increases. This would tend to indicate that the liver stores of vitamin A were near the minimum when the initial bleeding was made. Braun (1945) found in cattle that the blood plasma levels remained about normal until the liver stores of the vitamin were reduced to a minimum.

A third bleeding was made at the close of the experiment. The average of two analyses of blood drawn on successive days was taken for the reading of the final bleeding. These bleedings on August 18 and 20 showed a decided drop in the amount of vitamin A in the plasma of the various lambs. The plasma vitamin A levels of all lots except 2 and 4 dropped below the initial bleeding. These comparatively low levels of blood plasma vitamin A may be attributed to the very hot weather that prevailed throughout the first half of August. Low vitamin A levels of plasma in hot weather may be caused by the increased activity of the thyroid gland. During hot weather the basal metabolic rate is increased. Maynard (1947), McCollum (1947) and others have reported that the thyroid hormone, thyroxine, was responsible for the control of the basal metabolic rate in most animals and an increased basal metabolic rate was associated with hyper-thyroidism. Wendt (1936), Abelin (1936) and Sure and Buchanan (1937) reported thyroxin and vitamin A to be antagonistic. They reported abnormally low blood concentrations of vitamin A in cases of exopthalmic goiter, a condition of hyper-thyroid activity. When parts of the thyroid gland were removed the blood vitamin A returned to normal. Johnson and Bauman (1947) reported a somewhat different situation in rats. They found that within the dosage limits employed, the ability to store preformed vitamin A was approximately the same in hypo-throid or the hyper-throid rats as in normal rats.

It is quite evident from the data given in Table 5 that the liver stores in the lambs which received 10x the minimum daily requirement

TABLE 5.

SUMMARY OF VITAMIN A INTAKE, PLASMA LEVELS OF VITAMIN A AND THE VITAMIN A STORED IN THE LIVERS OF THE EXPERIMENTAL LAMBS

Lot	Lamb No.	Ration	Vit. A Intake	Average Daily Vit. A Intake µg	Blood Plasma Levels µ/100 ml.			Liver Storage µg/100 gm.	Liver Weight gm.	Total Storage
					May 15 Initial	July 3	Aug. 18-20 Final			
1	14	C.S.M.	1¼ X	561.9	8.1	23.2	14.0	218	453.6	988.8
	19		Min.	588.1	34.3	30.1	24.7	5,729	453.6	25,986.7
	24			771.8	24.7	25.8	18.0	4,586	567.0	26,002.6
	25			742.5	25.0	25.2	23.5	6,787	567.0	38,482.3
Lot Average				666.1	23.0	26.1	20.1	4,330	510.3	22,865.1
2	3	C.S.M.	10 X	5,097.4	23.2	30.7	29.5	14,787	680.4	100,610.7
	11		Min.	5,895.4	11.2	30.7	16.2	14,301	623.7	89,195.3
	35			5,724.4	34.6	34.1	27.3	25,875	623.7	161,382.4
	39			4,902.0	25.0	41.4	31.2	19,281	538.7	103,866.7
Lot Average				5,404.8	23.5	34.2	26.1	18,561	616.6	113,763.7
3	6	S.B.M.	1¼ X	756.7	23.1	20.8	7.8	1,148	567.0	6,509.2
	10		Min.	772.9	12.3	27.8	19.4	560	680.4	3,810.2
	13			639.0	29.3	33.5	23.4	1,447	567.0	8,204.5
	41			590.3	24.6	26.0	15.0	2,000	453.6	9,072.0
Lot Average				689.7	22.3	27.0	16.4	1,289	567.0	6,898.9
4	7	S.B.M.	10 X	6,392.1	23.6	32.0	27.7	16,323	567.0	92,551.4
	28		Min.	4,812.4	13.8	38.7	35.5	15,201	538.7	81,887.8
	29			6,123.4	32.0	38.7	22.0	24,150	680.4	164,316.6
	33			6,139.7	26.7	39.3	21.5	17,373	680.4	118,205.9
Lot Average				5,849.1	24.0	37.2	26.7	18,262	616.6	114,240.4

TABLE 5 (CONT'D).

Lot	Lamb No.	Ration	Vit. A Intake	Daily Vit. A Intake µg	Blood Plasma Levels µ/100 ml: May 15 Initial	July 3	Aug. 18-20 Final	Liver Storage µg /100 gm.	Liver Weight gm.	Total Storage
5	8	C.S.M.	1¼ X	562.9	14.4	20.0	12.5	831	453.6	3,769.4
	17	+	Min.	653.2	26.5	24.6	19.8	3,266	567.0	18,516.2
	23	Urea		704.9	23.1	24.9	17.4	1,360	538.7	7,326.3
	38			852.0	33.0	28.4	19.3	11,304	623.7	70,503.0
Lot Average				693.3	24.2	24.5	17.3	4,190	545.7	25,028.7
6	4	C.S.M.	10 X	5,352.0	26.6	25.3	19.6	19,431	567.0	110,173.8
	22	+		4,885.7	16.1	32.0	20.0	21,753	425.3	92,515.5
	26	Urea		5,398.7	29.8	27.8	24.0	25,875	425.3	110,046.4
	27			5,830.3	23.3	29.5	29.7	24,150	595.4	143,789.1
Lot Average				5,366.7	24.0	28.7	23.3	22,802	503.2	114,131.2
7	9	S.B.M.	1¼ X	590.3	17.8	21.6	14.5	1,345	396.9	5,338.3
	21	+	Min.	781.0	28.2	33.8	17.3	5,400	652.1	35,213.4
	37	Urea		669.4	28.1	32.6	27.3	1,492	567.0	8,459.6
	44			680.7	22.6	30.1	18.2	4,109	510.3	20,968.2
Lot Average				680.3	24.2	29.5	19.3	3,087	531.5	17,494.7
8	5	S.B.M.	10 X	6,384.0	18.1	22.7	17.9	15,648	708.8	110,913.0
	18	+	Min.	5,341.7	19.5	29.8	18.6	19,995	482.0	96,375.9
	36	Urea		4,975.3	29.8	28.7	25.0	18,345	510.3	93,614.5
	43			5,211.4	29.9	36.4	18.6	28,983	425.3	123,264.7
Lot Average				5,478.1	24.3	29.4	20.0	20,743	531.6	106,042.0

of vitamin A were much higher than the stores of the lambs which received $1\frac{1}{4}$x the minimum requirement. These differences were highly significant. Differences of 5,650 and 7,609 were required for significance at the 5 percent and 1 percent respectively when the data was analysed by the method suggested by Snedecor (1946). These results are in agreement with those of Hoefer and Gallup (1947), with regard to the storage of vitamin A in the liver of lambs at high and low levels of vitamin A or carotene intake.

The average vitamin A stored in the livers of the eight lambs sacrificed at the beginning of the experiment, after having been on a vitamin A free ration for 115 days, was 6,227 micrograms per 100 grams of liver. This amount was higher than the average for any of the lots receiving $1\frac{1}{4}$x the minimum requirement of vitamin A. This would tend to indicate, as has previously been suggested by Peirce (1945) and Hoefer and Gallup (1947), that the minimum requirement of vitamin A for lambs, estimated by Guilbert et al. (1937), is too low.

When vitamin A was fed at the 10x the minimum requirement level there were no marked differences between cottonseed meal and soybean meal in their effect on storage of vitamin A in the livers of lambs under the conditions of this experiment. When vitamin A was given at $1\frac{1}{4}$x the minimum requirement there appeared to be some retarded storage of the vitamin in the lambs receiving soybean meal; however, the differences in storage between these lambs and the ones receiving cottonseed meal were not significant.

Urea was substituted for both the cottonseed meal and soybean meal by replacing one-half the nitrogen furnished by the meals. This required approximately 1.5 pounds of urea per 100 pounds of feed.

Vitamin A was administered to the urea lots at two different levels as was the case in the non-urea lots. There were no differences manifested in liver storage of vitamin A when the cottonseed meal plus urea and soybean meal plus urea lots were compared.

Under the conditions of this experiment urea appeared to have no influence on the storage of vitamin A in the livers of lambs. In most instances there was more storage when urea was added to the ration than when only a vegetable protein was fed. These differences were not significant.

As is shown in Table 6, the lots of lambs that did not receive urea made more gains than did the urea fed lots; however, this difference was not significant except between lots 4 and 8. Differences in daily gains of 0.06 pound and 0.08 pounds were required at the 5 percent and 1 percent points, respectively. Assuming that all lambs were receiving adequate amounts of vitamin A for growth, lot 2, which received cottonseed meal, made significantly greater gains than did the cottonseed meal plus urea group in lot 5.

Without an exception the urea lots required more feed per pound of gain than did the non-urea lots. This was true in both the cottonseed meal and the soybean meal groups. Willit et al. (1946) obtained less milk production from dairy cows when urea was fed, whether the daily intake was 0.48 pounds or 0.24 pounds. These amounts provided 36 percent and 19 percent, respectively, of the total crude protein equivalent intake. Briggs et al. (1948) reported that pellets containing approximately 25 percent of their nitrogen as urea and the remainder almost entirely from cottonseed meal permitted about the same storage

TABLE 6.

THE AVERAGE GAINS AND FEED CONSUMPTIONS DURING THE EXPERIMENT

Lot	Ration	Lamb No.	Daily Gain lbs.	Lbs. Feed Per lb. Gain	Lot	Ration	Lamb No.	Daily Gain lbs.	Lbs. Feed Per lb. Gain
1	C.S.M.	14	.09	26.3	3	S.B.M.	6	.20	14.9
	1¼ X	19	.10	23.6		1¼ X	10	.32	10.0
	Min.	24	.21	14.2		Min.	13	.21	13.1
	Vita. A.	25	.21	15.1		Vita. A	41	.09	26.3
Average			.16	19.8	Average			.21	16.1
2	C.S.M.	3	.17	18.6	4	S.B.M.	7	.25	13.4
	10 X	11	.29	11.8		10 X	28	.21	12.6
	Min.	35	.19	16.5		Min.	29	.22	13.5
	Vita. A	39	.14	20.8		Vita. A	33	.30	10.6
Average			.20	16.9	Average			.25	12.5
5	C.S.M.	8	.08	28.1	7	S.B.M.	9	.09	29.8
	+ Urea	17	.11	22.5		+ Urea	21	.20	15.4
	1¼ X Min.	23	.20	15.3		1¼ X Min.	37	.15	20.2
	Vita. A	38	.17	16.7		Vita. A	44	.25	11.4
Average			.14	21.2	Average			.18	19.2
6	C.S.M.	4	.18	16.3	8	S.B.M.	5	.17	17.5
	+ Urea	22	.08	31.8		+ Urea	18	.17	17.4
	10 X Min.	26	.13	20.8		10 X Min.	36	.13	19.6
	Vita. A	27	.19	16.6		Vita. A	43	.18	14.6
Average			.15	21.4				.17	17.3

of nitrogen as straight cottonseed meal when fed to lambs at the level to provide equivalent nitrogen intake.

The results obtained in this experiment indicate that when urea was added to a ration fed to lambs, at the rate of 1.5 pounds per 100 pounds of ration, neither the amount of gain nor the economy of gain was as good as when only protein from plant sources were fed.

All lots which received soybean meal appear to have made more gains than those lots which received cottonseed meal. Although these differences were not significant, the difference between lots 1 and 3 and between 2 and 4 approached significance. The effeciency of gain was slightly better in the soybean meal lots.

There were no toxic effects manifested from the feeding of urea at any time during this experiment. This is in agreement with Mills et al. (1944), who fed heifer calves, and with Briggs, et al. (1947), and Dinning (1948), who fed yearling steers. In their experiments urea was safe for feeding these larger ruminants when fed at recommended amounts. When offered orally in water urea was shown by Dinning (1948) to be toxic to steers.

SUMMARY

A factorial design experiment was conducted with thirty-two fattening lambs that had previously been depleted of vitamin A reserves. Four lambs were used in each of eight experimental treatments. The lambs were fed two levels of vitamin A, two kinds of vegetable protein, and a combination of vegetable protein and urea nitrogen to study the influence of these variations in nutrient composition on the vitamin A level of blood plasma and the storage of vitamin A in the liver. Data were secured on the rate and economy of gain obtained from feeding cottonseed meal, soybean meal, and a combination of vegetable protein and urea nitrogen.

Vitamin A was fed to lambs at levels of 1¼x the minimum and 10x the minimum estimated requirement and no significant differences in blood plasma levels of vitamin A were obtained between the two levels of vitamin A intake. However, average liver stores up to 22,802 micrograms per 100 grams of liver per lot were obtained when lambs were fed 10x the minimum requirement, while the highest average for any lot receiving 1¼x the minimum requirement was 4,330 micrograms per 100 grams of liver. Added vitamin A appeared to have no influence on the rate and economy of gain.

Two of the lots received cottonseed meal and two received soybean meal as the source of protein in the ration. The source of plant protein had no effect on the vitamin A plasma level or storage of the vitamin in the liver. Lambs receiving soybean meal appeared to make larger gains per unit of feed consumed than did the lambs receiving cottonseed meal.

In two lots urea nitrogen replaced approximately one-half of the

nitrogen supplied by cottonseed meal and in two other lots a similar amount of the nitrogen of soybean meal was replaced by urea nitrogen. The addition of urea to the ration did not influence the vitamin A level of the plasma nor the storage of vitamin A in the liver. The rate and economy of gain were lowered when urea nitrogen was substituted for the plant sources of protein.

BIBLIOGRAPHY

Abelin, I. 1936. Vitamin A Therapy in Hyperthyroidism. Schweiz. Med. Wochenchr. 66:1106-1107.

Almquist, H. J., E. Mecchi, F. H. Kratzer and C. R. Grau. 1942. Soybean Protein as a Source of Amino Acids for the Chick. Jour. Nutr. 24:385-392.

Armsby, H. P. 1911. The Nutritive Value of the Non-protein of Feeding Stuffs. U. S. Dept. Agr., Bur. An. Ind. Bul. 139.

Axelsson, J. 1942. Urinamnets Värde Och Användbaret för Husdjurens Näring. Nutr. Abs. and Rev. 12:687, No. 3883.

Bethke, R. M. and M. C. Sweet. 1939. The Comparative Value of Expeller and Toasted Solvent Soybean Oil Meals for Chicks. Ohio Agri. Exp. Sta. Bimonthly Bulletin 199:122-125.

Boyer, P. D., P. H. Phillips, W. S. Lundquist, C. W. Jensen and I. W. Rupel. 1942. Vitamin A and Carotene Requirements for the Maintenance of Adequate Blood Plasma Vitamin A in the Dairy Calf. Jour. Dairy Sci. 25:433-440.

Braun, Werner. 1945. Studies of the Carotenoid and Vitamin A Levels in Cattle. Part II. Carotenoids and Vitamin A in the Liver, Their Ration and Their Relationship to Blood Levels. Jour. Nutr. 29:73-79.

Briggs, H. M., W. D. Gallup, A. E. Darlow, D. F. Stephens, and C. Kinney. 1947. Urea as an Extender of Protein When Fed to Cattle. Jour. An. Sci. 6:445-460.

Briggs, H. M., W. D. Gallup, V. G. Heller, and A. E. Darlow. 1948. Urea as an Extender of Protein When Fed to Lambs. Jour. An. Sci. 7:35-40.

Csonka, Frank A., and D. Breese Johes. 1934. The Cystine, Tryptophane, and Tyrosine Content of the Soybean. Jour. Agri. Research. 49:279-282.

Cullison, A. E. 1944. The Use of Urea in Making Silage from Sweet Sorghum. Jour. Ani. Sci. 3:59-62.

Darlow, A. E., V. G. Heller, W. D. Campbell, J. C. Hillier, and J. A. Hoefer. 1946. Protein and Mineral Supplements for Wintering Two Year Old Steers on Grass. Okla. Exp. Sta. Bul. 296:8-11.

Dinning, James S. 1948. The Addition of Urea to Rations for Cattle and Sheep. A Thesis, Submitted to the Department of Animal Husbandry, Oklahoma Agricultural and Mechanical College, in Partial Fulfillment of the Requirements for the Degree of Doctor of Philosophy.

Draper, C. I., Robert John Evans. 1944. Cross Values of Combinations of Cottonseed Meal, Soybean Oil Meal, and Herring Fish Meal. Poultry Sci. 23:189-192.

Draper, C. I., and Robert John Evans. 1944. Plant Protein Concentrates in the Chick Ration. Poultry Sci. 23:507-509.

Erb, R. E., F. N. Andrews, S. M. Hauge and W. A. King. 1947. Observations on Vitamin A Deficiency in Young Dairy Bulls. Jour. Dairy Sci. 30:687-702.

Fingerling, G., G. Herntzsch, H. Kinze, and K. Reifgerst. 1937. Ersatz des Nahrungseiweisses durch Harnstoff bein Wachsenden Rinde, Landw. Vers. Sta. 128:221-235.

Gallup, W. D., and J. A. Hoefer. 1946. Determination of Vitamin A in Liver. Indus. and Eng. Chem. (Anal. Ed.). 18:288.

Goss, Harold. 1943. Some Peculiarities of Ruminant Nutrition. Nutr. Abs. and Rev. (A Review). 12:531-538.

Guilbert, H. R. and G. H. Hart. 1934. Storage of Vitamin A in Cattle. Jour. Nutr. 8:25-44.

Guilbert, H. R., R. F. Miller, and E. H. Hughes. 1937. The Minimum Vitamin A and Carotene Requirements of Cattle, Sheep and Swine. Jour. Nutr. 13:543-564.

Guilbert, H. R., C. E. Howell, and G. H. Hart. 1940. Minimum Vitamin A and Carotene Requirements of Mammalian Species. Jour. Nutr. 19:91-103.

Glover, J., T. W. Goodwin and R. A. Morton. 1946. The Relationship Between Blood Vitamin A Levels and Liver Stores in Rats. Biochem. Jour. 40:lvii.

Hart, E. B., G. Bohstedt, H. J. Deobald, and M. I. Wegner. 1939. The Utilization of Simple Nitrogenous Compounds Such as Urea and Ammonium Bicarbonate by Growing Calves. Jour. Dairy Sci. 22:785-798.

Hart, George H. 1940. Vitamin A Deficiency and Requirements of Farm Mammals. Nutr. Abs. and Rev. 10:261-272.

Harris, Lorin E. and H. H. Mitchell. 1941. The Value of Urea in the Synthesis of Protein in the Paunch of the Ruminant. II. In Growth. Jour. Nutr. 22:183-196.

Hague, S. M., J. H. Hilton, and J. W. Wilbur. 1940. The Adsorption of the Vitamin A Suppressing Factor from Soybean Oil. Jour. Dairy Sci. 23:719-723.

Hayward, J., H. Steenbock, and G. Bohstedt. 1936. The Effect of Cystine and Casein Supplements upon the Nutritive Value of the Protein of Raw and Heated Soy Beans. Jour. Nutr. 12:275-283.

Hibbs, J. W. and W. K. Krauss. 1946. The Effect of Feeding Vitamin A on the Blood Picture and on Liver Storage in Calves. Jour. Dairy Sci. 29:519-520.

Hoefer, J. A. and Willis D. Gallup. 1947. The Comparative Value of a Carotene Concentrate, Alfalfa Meal, and a Fish Liver Oil in Maintaining the Vitamin A Content of the Blood and Liver of Fattening Lambs. Jour. An. Sci. 6:325-333.

Johnson, Margaret L., Helen T. Parsons, and Harry Steenbock. 1939. The Effect of Heat and Solvents on the Nutritive Value of Soybean Protein. Jour. Nutr. 18:423-434.

Johnson, R. M. and C. A. Baumam. 1947. The Effect of Thyroid on the Conversion of Carotene into Vitamin A. Jour. Biol. Chem. 171:513-521.

Jones, J. H., H. Schmidt, R. E. Dickson, G. S. Fraps, J. M. Jones, J. K. Riggs, A. R. Kemmerer, P. E. Howe, W. H. Black, N. R. Ellis, and Paul T. Marion. 1943. Vitamin A Studies in Fattening Feeder Calves and Yearlings. Texas Agri. Exp. Sta. Bul. 630.

Kimble, M. S. 1939. The Photo-colorimetric Determination of Vitamin A and Carotene in Human Plasma. Jour. Lab. and Clin. Med. 24:1055.

Kuhlman, A. H. and W. D. Gallup. 1942. Carotene (Provitamin A) Requirements of Dairy Cattle for Conception. Jour. Dairy Sci. 25:688-689.

Lewis, J. M. and Logan T. Wilson. 1945. Vitamin A Requirements in Calves. Jour. Nutr. 30:467-475.

Lofgreen, G. P., J. K. Loosli, and L. A. Maynard. 1947. The Influence of Protein Source upon Nitrogen Retention by Sheep. Jour. An. Sci. 6:343-347.

Loosli, J. K. and L. E. Harris. 1945. Methionine Increases the Value of Urea for Lambs. Jour. An. Sci. 4:435-437.

Maynard, Leonard A. 1947. Animal Nutrition. McGraw- Hill Book Company, Inc. 180-193.

McCollum, E. V., Elsa Orent-Keiles and Harry G. Day. 1947. The Newer Knowledge of Nutrition. The MacMillan Company.

Mills, R. C., C. C. Lardinois, I. W. Rupel, and E. B. Hart. 1944. Utilization of Urea and Growth of Heifer Calves with Corn Molasses or Cane Molasses as the Only Readily Available Carbohydrate in the Ration. Jour. Dairy Sci. 27:571-578.

Mitchell, H. H. and D. B. Smuts. 1932. The Amino Acid Deficiencies of Beef, Wheat, Corn, Oats, and Soy Beans for Growth in the White Rat. Jour. Biol. Chem. 95:263-281.

Moore, Thomas. 1929. Vitamin A and Carotene. Biochem. Jour. 23:803-811.

Owen, E. C., J. A. B. Smith, and N. C. Wright. 1943. Urea as a Partial Protein Substitute in the Feeding of Dairy Cattle. Biochem. Jour. 37:44-53.

Pearson, R. M. and J. A. B. Smith. 1943. The Utilization of Urea in the Bovine Rumen. Biochem. Jour. 37:142-148.

Peirce, A. W. 1945. The Effect of Intake of Carotene on the General Health and on the Concentration of Carotene and of Vitamin A in the Blood and Liver of Sleep. Aust. Jour. Exp'l Biol. and Med. Sci. 23:295.

Philips, A. C., R. H. Carr, and D. C. Kennard. 1920. Meat Scraps Versus Soybean Proteins as a Supplement to Corn for Growing Chicks. Jour. Agri. Res. 18:391-398.

Riggs, J. K. 1940. The Length of Time Required for Depletion of Vitamin A Reserves in Range Cattle. Jour. Nutr. 20:491-500.

Rupel, I. W., G. Bohstedt, and E. B. Hart. 1943. The Comparative Value of Urea and Linseed Meal for Milk Production. Jour. Dairy Sci. 26:647-664.

Snedecor, George Waddel. 1946. Statistical Methods Applied to Experiments in Agriculture and Biology. Ames, Ia., The Collegiate Press, Inc.

Sure, B. and K. S. Buchanan. 1937. Influence of Hyperthyroidism on Vitamin A Reserves of the Albino Rat. Jour. Nutr. 13:521-524.

Ward, R. E., S. I. Bechdel, and N. B. Guerrant. 1940. Carotene and Vitamin A in the Nutrition of Growing Dairy Cattle. Jour. Dairy Sci. 23:115-123.

Wegner, M. I., A. N. Booth, G. Bohstedt, and E. B. Hart. 1941. The Utilization of Urea by Ruminents as Influenced by the Level of Protein in the Ration. Jour. Dairy Sci. 24:835-844.

Wegner, M. I., A. N. Booth, G. Bohstedt, and E. B. Hart. 1943. The "In Vitro" Conversion of Inorganic Nitrogen to Protein by Micro-organisms from the Cows Rumen. Jour. Dairy Sci. 23:1123-1129.

Wendt, H. 1936. Thyroid and Vitamin A. Med. Klin. 32:27-29.

Wilgus, H. S. Jr., L. C. Norris, and G. F. Heuser. 1936. Effect of Heat on Nutritive Value of Soy-Bean Oil Meal. Ind. and Engr. Chem. (Indus. Ed.). 28:586-588.

Willett, E. L., L. A. Henke, and C. Maruyama. 1946. The Use of Urea in Rations for Dairy Cows Under Hawaiian Conditions. Jour. Dairy Sci. 29:629-638.

TYPED BY: FLOREINE E. ADAMS