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Vitamin a content of the hen's egg yolk as influenced by ingestion of Cod-Liver oil

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THE VITAMIN A CONTENT OF THE HEN'S EGG YOLK AS INFLUENCED BY INGESTION OF COD-LIVER OIL

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CHAPTER I

INTRODUCTION

It has been known for some time that the amount of vitamin A in the egg of the chicken (Gallus domesticus) depends upon the amount of that factor in the chicken's diet. In 1935 DeVaney, Titus, and Nestler discovered that with graded percentages of cod-liver oil in the ration, graded amounts of the vitamin A factor were found in the yolk of the egg. In the experiments referred to above, the chickens were confined indoors and were not exposed to any direct sunlight. These men had for their primary objective the study of the interrelationships of the vitamins A and D.

The result stated above indicated that graded amounts of cod-liver oil in the hen's ration had a graded effect on the amount of vitamin A content of the egg yolk. Therefore, it seemed that a similar experiment should be carried out with the chickens under ordinary living conditions. For this reason, the feed already being used was taken as the basal diet, with an increase in the percentage (in weight) content of cod-liver oil in the different rations.

The specific objective of this study was to determine the percentage influences of different amounts of cod-liver oil ingested. The experiment was conducted with the chickens under conditions to which they were accustomed in the ordinary feeding processes. The study was expected to reveal some definite information as to the relative amount of vitamin A in the feed which was already being used. Cod-liver oil was added in known weight percentages with definite U. S. P. units of vitamin A. To check the egg yolk, albino rats were used, as they have been found to respond readily

to the presence or absence of the vitamin A factor in the diet (Sherman-Munsell, 1925).

CHAPTER II

REVIEW OF LITERATURE

Proof that eggs are a good source of vitamin A was first brought to light through experiments by Osborne and Mendel, and McCollum and Davis in 1913. In these experiments concerned with the "Necessity of Certain Lipins in the Diet During Growth," (1913) they used chemically pure rations containing purified casein, carbohydrates and various salt mixtures. Growth was found to continue for periods varying from 70 to 120 days. After that time no growth could be induced with the use of the components of that ration, though they were mixed in varied proportions. When the rats were fed naturally occurring food substances, extracted from eggs with ether, growth was immediately resumed. This test was made by feeding rats these various food mixtures. When pure lard, olive oil and almond oil were substituted for the egg extract, growth was not resumed. This indicated that there was some organic substance lacking in the chemically pure food that was present in the ether extract from eggs. In 1920 this substance received the name of vitamin A. In 1931 Sherman and Smith confirmed this work in their book, "The Vitamins." They reported that the vitamin was contained principally in the yolk of the egg, and that the concentration depends to an extent upon the food of the hen. To what extent it depended upon the ingested vitamin factor had not been determined at that time. The egg as a whole may be expected to contain about 15 to 20 units of vitamin A per gram, and the yolk usually contains a concentration of about three times that amount.

Bethke, Kennard, and Sasserman in 1927 reported that hens fed cod-liver oil lay eggs containing five times as much vitamin A in the

yolks as hens fed the basal diet containing yellow corn and alfalfa hay as the sole source of vitamin A. In these experiments the hens were fed a basal diet for a period of nine months containing: yellow corn, 30 parts; ground wheat, 20 parts; ground oats, 20 parts; wheat middlings, 10 parts; meat scraps, 10 parts; and oyster shell and water. These hens were kept in pens; pen 12 was fed the basal diet plus 2% of cod-liver oil and kept indoors; pen 13 was fed only the basal diet and kept indoors; pen 14 was fed the basal diet and allowed to run on the range with access to blue-grass; and pen 15 was fed the basal diet plus chopped alfalfa meal. The only sunlight received by the hens in pens 12, 13, and 15 was that which filtered through windows from a southern exposure. The vitamin A test was run according to the Sherman-Munsell method (1925).

It was found that the vitamin A content of the egg yolks from hens which received 2% of cod-liver oil added to the basal diet increased five times. The same was found to be true with those which were allowed to run on the range. The yolks from those fed the basal diet plus chopped alfalfa meal showed an increase, but notso much as was shown in the other two. All of these were checked against pen 13, which received only the basal diet. It served as a control.

Holmes, Doolittle, and Moore reported in 1927 in their work on the vitamin A potency of cod-liver oil that when it was added to the deficient diet of the chickens, there was improvement in egg production, hatchability, weight of eggs, fertility, and the viability of the chicks hatched.

In 1932 Sherwood and Fraps, while studying the quantities of vitamin A required by pullets for maintenance and for egg production, reported that yellow corn may serve as a source of vitamin A for maintenance of the animal body but not for the egg. The eggs tested at the

beginning of the experiment contained about 20 Sherman-Munsell units. When the hens were kept on a diet with yellow corn as the sole source of vitamin A, the content of vitamin A decreased from 20 units, and ranged from 5 to 8 units.

In 1933, Ellis, Miller, Titus and Beverly reported that color pigment was not an indication of the amount of vitamin A content of the egg yolk, especially when cod-liver oil is added to the basal diet. Their basal diet was made up of the following: yellow corn, 500 parts; wheat bran, 245 parts; oat meal 150 parts; and alfalfa meal, 55 parts. The diets for two groups of hens were prepared by adding other ingredients, one of them called the normal diet and the other the deficient diet. The normal diet was prepared by adding the following to 80 parts of the basal diet: fish meal, 7 parts; meat meal, 8 parts; and dried butter milk, 5 parts. The deficient diet was prepared by adding to 80 parts of the basal diet 20 parts of a protein mixture low in vitamin B in which vitamin A was increased by the addition of cod-liver oil. The normal diet was rich in vitamin B but poor in vitamin A. The egg yolks from hens fed the normal diet were normal in pigmentation, but were low in vitamin A content. The yolks from hens fed the deficient diet were poor in pigmentation, but had a high vitamin A content. The vitamin A test was run by the Sherman-Munsell method (1925).

In 1934 Bisbey and his co-workers reported that yellow corn and cod-liver oil had a decided effect on the vitamin A content of the egg yolk. They ran the vitamin A test according to the Sherman-Munsell method (1925). Their report revealed the following findings:

1. When 65% yellow corn and 10% alfalfa were added to the basal diet as the sole source of vitamin A, the yolks yielded 28 Sherman-Munsell units of vitamin A per gram.

2. When 65% yellow corn was added to the basal diet as the only source of vitamin A, the yolks yielded 9 Sherman-Munsell units of vitamin A per gram.

3. When 35% yellow corn was added to the basal diet as the only source of vitamin A, the yolk yielded 7 Sherman-Munsell units of vitamin A per gram.

4. When 25% yellow corn plus 0.5% cod-liver oil were added to the basal diet as the sole source of vitamin A, the yolk yielded 28 Sherman-Munsell units of vitamin A per gram.

The data above point out the fact that cod-liver oil added to the diet of the hen does have a decided effect upon the amount of the factor contained in the egg yolk.

In 1934, Wilcke, Nelson and Henderson reported that hens fed mash as a basal diet, supplemented with 2% cod-liver oil, produced eggs the yolk of which contained 20 Sherman-Munsell units of vitamin A per gram. A group fed yellow corn and allowed to run on blue-grass produced eggs the yolks of which contained 34 Sherman-Munsell units per gram. This further points to the fact that cod-liver oil in the diet influences the vitamin A content of the egg yolk, but not so much as the blue-grass.

Russell reported in 1934 that when hens were fed a mash containing 32% yellow corn and 1% cod-liver oil, and a scratch feed containing equal parts of cracked corn (yellow) and wheat, the eggs contained 260 vitamin A units. This number represented 47% of the vitamin A in the diet. When white corn was substituted for yellow corn in the mash and in the scratch feed, the eggs contained 200 vitamin A units which represented 48% of the vitamin A units contained in the diet. This points to the fact that nearly half of the ingested vitamin A by hens is contained in the yolk of the egg.

In 1935, DeVaney, Titus, and Nestler reported in their study to determine the interrelationships of vitamins A and D, that eggs laid by pullets fed different amounts of cod-liver oil, in connection with the

basal diet, contained different amount of the vitamin A factor, corresponding to the amount of the cod-liver oil in the diet. In these experiments pairs were used. Each pen contained 18 hens and 1 cockerel. The hens were confined indoors and received no direct sunlight.

The basal diet used was composed of the following: yellow corn meal, 52.63%; wheat bran, 25.79%; rolled oats, 15.79%; and alfalfa leaf meal, 5.79%. To the basal diet of the various pairs of pens was added: Pair I--Pen 1, 1% cod-liver oil; Pen 2, 1% cod-liver oil plus 0.5% viosterol 160D; Pair II--Pen 1, 2% cod-liver oil; Pen 2, 2% cod-liver oil plus 0.5% viosterol 160D; Pair III--Pen 1, 8% cod-liver oil; Pen 2, 8% cod-liver oil plus 0.5% viosterol 160D.

The addition of viosterol to the diet did not have any apparent effect on the transfer of the vitamin A to the egg. The hens that were fed the ration containing 8% cod-liver oil laid eggs the yolk of which contained about 80 units of vitamin A per gram. The hens fed the diets containing 1% and 2% of cod-liver oil laid eggs the yolks of which contained about 40 units of vitamin A per gram. Those hens which were used as controls and received no supplement by way of cod-liver oil, laid eggs the yolks of which contained about 20 units per gram. The Sherman-Munsell method (1925) was used in the vitamin A assay.

Koenig, Kramer, and Payne reported in 1935 in their experiment on "The Vitamin A Content of Eggs as Related to Rate of Production" that after four months of this experiment, the high and low producers' eggs were similar in vitamin A content. They contained approximately 25 vitamin A units per gram. Near the close of the first year of the experiment the low producers' eggs contained 33 units per gram, and the high producers' eggs contained 20 units per gram. When pullets were fed 1% cod-liver oil as the only source of vitamin A, they laid eggs with a

pale yolk, but relatively rich in vitamin A.

The findings above indicate definitely that there is a relationship between the amount of vitamin A ingested by the hen and the amount of the factor contained in the egg yolk. Cod-liver oil seems to be a rich source of the vitamin A factor that may be easily transferred to the yolk of the egg.

CHAPTER III

MATERIALS AND METHOD

Three groups (A, B, and C) of white (Leghorn) hens owned by Spelman College were used in this experiment. Two of the groups (A and B) contained five hens each while group C contained the remainder of approximately three hundred. There was no cockerel present in any group. The three groups were kept in standard chicken houses, with only a partition of wire between them. This was done in order to get approximately uniform living conditions as far as light and temperature were concerned. All houses were opened from the south side, providing natural sunlight each day. The feeding procedure was as usual, the only difference in the food content being an increase in the weight content of cod-liver oil in the laying mash for groups A and B. This mash was available at all times as usual, and the scratch feed was fed once each day as usual. A description of the basal diet and the vitamin A supplement was:

Group C: The basal diet described below was fed this group.

Full-O-Pep Laying Mash--This was prepared by the Quaker Oats Company. It had the following guaranteed analysis:

Crude Protein, not less than	20.00 per cent
Crude Fat, not less than	4.5 per cent
Crude Fiber, not less than	8.00 per cent
Carbohydrate, Nitrogen-free Extract	45.00 per cent

The ingredients as listed by the company, but not in percentage contents, were: Oat meal, hominy feed, yellow hominy feed, wheat bran, wheat standard middlings, barley feed, corn gluten meal, fortified sardine oil, dried butter-milk, cane molasses, dehydrated alfalfa, dehydrated cereal grass (from wheat, oats, rye, and barley), and three-fourths of one per cent salt. The scratch feed was a mixture of wheat, yellow corn,

maize, and rye. It was not possible to ascertain the per cent of vitamin A present in the feed above.

The vitamin A supplements in the feed for groups A and B in the form of cod-liver oil containing 1000 U. S. P. units were as follows:

Group A: To the basal diet 4% in weight of cod-liver oil was added.

Group B: To the basal diet 9% in weight of cod-liver oil was added.

The mash as prepared above was stored in separate cans in a relatively dark place to prevent any loss of vitamin A potency from radiation or oxidation (Dunn-1924, Evers-1929). Twenty-five pounds were mixed at the time, which lasted for approximately one month.

The hens were put on the diet above October 29, 1940. The first sample of eggs for analysis was taken January 9, 1941. The eggs from each group of hens were taken at random.

The vitamin A test was run according to the Sherman-Munsell method (1925), described as follows: Young albino rats were used--ages from 21 to 29 days, weight 35 to 50 grams. The nutritional history of the rats was known. The rats were placed in a raised screen floor cage to prevent access to excreta, which may contain vitamin A. They were fed in non-scatter diet cups with openings the size of which would permit only the head to enter. This was done to prevent the feet from getting into the food. The water was provided through a self-flow water bottle (Thorpe-type), which prevented contamination. All utensils were sterilized each week. The rats were placed on the vitamin A free diet, complete, however, in every other way. They were kept on this diet until they ceased to gain in weight, and developed the characteristic ophthalmic trouble. The latter is characterized by swelling of the eye lids and accumulation of exudate in the corners of the eyes (Sherman-Munsell-1925).

Weight records were taken at least once each week until the cessation of growth was expected. The rats were then weighed every other day until growth had ceased; then each day for at least two or three days, when complete cessation of growth was definitely ascertained. At this time, called the end of the fore period, the animals were placed in separate cages, in groups or singly. The cages were of the type mentioned above. One group was continued on the basal diet (vitamin A-free) as a negative control, while the other groups received the vitamin A free basal diet, with the systematic addition of graded amounts of the test food.

The test was carried on for a period of eight weeks (Sherman & Smith-1931). Any gain in weight over the control group was attributed to the vitamin A received from the test food. This is measured in units, called Sherman-Munsell units. One unit of vitamin A is reckoned as that amount of vitamin A which when fed daily for a period of 4 to 8 weeks just suffices to support a rate of gain in weight of 3 grams per week in a standard test animal (Sherman-1937).

In this experiment all of the steps above were observed except the time and known nutritional history. Instead of continuing the study for a period of 8 weeks as prescribed by Sherman-Munsell-1925, the test was continued for a period of 5 weeks (Coward-1933). The basal diet for the rats was the same as that used by Sherman-Munsell-1925. It was prepared by the General Biological Supply House 1937, and contained the following ingredients:

Vitamin A free casein (purified)	18 per cent
Dried Brewer's yeast	5 " "
Salt mixture (Osborne-Mendel)	4 " "
Sodium Chloride	1 " "
Corn starch	67 " "
Biosterol	.5 per cent

In this experiment 4 young rats were used (A1, B1, C1 and D1), the ages agreeing with that mentioned above. The rats were put on the deficient diet when they were 27 days old. They showed cessation of growth at about the twenty-seventh day after being put on the deficient diet. The test period began on the twenty-ninth day after beginning the deficient diet.

Three eggs were used in making the egg sample for the test food. They were boiled not over 10 minutes and the yolk carefully removed and well mixed. Each rat of the three groups on the test food was fed .05 gram of the yolk mixture per day. The animals were fed twice per week. Between the feedings, each mixture of yolk which was not used at one feeding was stored in a sterilized container in the frigidaire at 10 degrees F. The yolk was weighed at each feeding on an analytical balance. This food was fed as a supplement to the basal diet, to group A1, B1 and C1. As a negative control, group D1 was kept on the basal diet until death.

CHAPTER IV

DATA AND RESULTS

It was first pointed out in 1913 by Osborne and Mendel, and McCullum and Davis, that growth would cease in young albino rats fed a chemically pure diet, devoid of certain lipins (later classified as vitamin A). Due to a store of this factor already in the body of the rat, growth was found to continue for periods varying from 70 to 120 days. At the end of the growth period at which time the animal was said to be depleted, growth was found to be immediately resumed on feeding as a supplement to the depletion diet an ether extract from egg yolk. Sherman and Smith (1931) pointed out that the hen's egg yolk was a sufficient source of the vitamin A factor and that the concentration depended to a great extent upon the food ingested by the hen. In 1935 DeVaney, Titus and Nestler discovered that hens ingesting different amounts of vitamin A in cod-liver oil laid eggs containing different percentages of the vitamin A factor.

Drummond and Coward (1920) introduced a quantitative method which was perfected in 1925 by Sherman and Munsel. As pointed out earlier in this paper, young rats were to be used as they were more sensitive to growth promoting food factors than older ones.

On March 20 the test animals were put on the deficient diet as described in an earlier chapter on material and methods. All animals at this time were apparently in good health and very active. Their weights at the beginning of the depletion period were:

Rat AI	2 cuts on the right ear	49.4 grams
Rat BI	1 cut " " left "	47.5 "
Rat CI	1 " " " right "	48.1 "
Rat DI	no " " either ear	48.4 "

Throughout the depletion period a steady gain in weight was maintained by all of the test animals. They showed no apparent effects until about the 26th day. At this time the animals were noticeably inactive; they showed inflamed eyes, and on the 27th day no weight gain was shown. All maintained a fair appetite, excepting AI, which did not have as good appetite at the close of the depletion period as at the beginning. AI and CI were the only animals which showed any type of health deformity at the close of the depletion period. AI apparently lost all of the color in one eye. AI and CI suffered a small degree of respiratory trouble. Their weights at the close of the depletion period were:

Rat AI. . . . . .149.9 grams
Rat BI. . . . . .134.7 "
Rat CI. . . . . .104.14 "
Rat DI. . . . . .177.7 "

This points out the fact that Rat DI made a gain greater than any other animal on the test. This happened though they all weighed within two grams of the same weight at the beginning of the depletion period. As the nutritional history of the test animals was not known, this weight gain might have been due to the fact that this animal, as well as those others that made rapid growth over CI, had had access to a greater amount of the vitamin A factor and had stored in the body a sufficient amount to continue to put on weight even though they did not have access to that factor during the depletion period, as they all ate from the same containers (Osborne and Mendel-1921).

On April 18 the animals were put on the test food. The test food was fed as a supplement to the basal diet as pointed out earlier, in the chapter on materials and methods. The details of the test feeding results were as shown below.

Rat AI weighed at the beginning of the test period 149.9 grams. The hens which laid the eggs, the yolks of which was fed to this animal,

were fed a supplement of 4% in weight of cod-liver oil. The fur of this animal was somewhat ruffled and, as stated above, the pink color of one eye was lost and little activity was shown. After the first week on the test food decided changes had taken place in these conditions. The weight had increased to 154 grams. By May 13 the color of the eye was again pink and other outward appearances were as before the depletion was effected. Weight was gained each week and on the final day of the test, May 22, the weight was 169.21 grams. This showed an increase of 20.12 grams over a period of 5 weeks, and an average gain per week of 4.025 grams. A unit of vitamin A is reckoned as that amount of the factor which when fed to a test animal over a period of from four to eight weeks is just sufficient to effect an average gain of 3 grams per week (Sherman-1937). The average here was 4.025. The test animal was fed .05 gram of the test food per day. The average gain shows that this amount of the test food contained approximately 1 1/3d units of the vitamin A factor. To ascertain the number of units per gram of the test food, 1 1/3 was divided by .05, the amount of the food fed per day. This gave approximately 26 units of vitamin A per gram (General Biological Supply House Inc.-1937).

Rat BI weighed at the beginning of the test period 134.7 grams. The hens which laid the eggs, the yolks of which was fed to this animal, were fed a supplement of 9% in weight of cod-liver oil. The general features of appearance of this animal were similar to that of AI except that there was no loss of eye color. After oneweek on the test food a decided change had taken place in the general conditions. The eyes had cleared of any exudate, the fur had again become smooth, and activity had been resumed. The weight had increased to 142.6 grams. By May 13 the outward appearances were as they were before the animal was depleted. On the final day, May 22, the animal weighed 169 grams. This was an increase of 35.7

grams over a period of 5 weeks, an average of 7.1 grams per week. Using the method used above, .05 grams of the test food in this case yielded approximately 2.3 units of the vitamin A factor. Two and three-tenths divided by .05 gives 46 units of the factor in each gram.

Rat CI weighed 104.1 grams when placed on the test food. The hens which laid the eggs the yolks of which was fed to this animal received no supplement of cod-liver oil in their ration. The physical conditions were similar to those of AI with the exception that there was no loss of eye color. After the first week on the test food the physical appearance had improved considerably. The weight had increased to 112.14 grams. The conditions as well as the weight continued to improve until the final date, May 22, when the weight had reached 120.12 grams. This was an increase of 16.02 grams for a period of 5 weeks and an average of 3.2 grams per week. Receiving as the animals above did, .05 grams of the test food per day, it is found that this food contained approximately 20 units of vitamin A per gram.

Rat DI was the control animal which weighed 177.7 grams at the end of the depletion period. This animal was suffering similar to AI, BI and CI. The control animal was kept on the deficient diet until death. The animal continued to be increasingly inactive, suffered increased respiratory trouble and rapid loss of weight until May 14 when death occurred. At the time of death the weight was 134.3 grams.

CHAPTER V

DISCUSSION

Osborne and Mendel, and McCullum and Davis (1913) first pointed out that young albino rats depleted of vitamin A showed symptoms of sore eyes containing exudate, ruffled fur, loss of weight and general health weakness. They also pointed out that these conditions could be eliminated immediately in such animals by feeding them ether extract of egg yolk. This indicated that egg yolk was a good source of that fat soluble substance which is essential to animal growth and maintenance of general healthful conditions. The fat soluble substance later became known as vitamin A. In 1931 Sherman and Smith confirmed this work and further pointed out that the amount of the factor contained in the yolk of the hen's egg depended upon the food ingested by the hen.

Later, Bethke, Kennard and Sasserman (1927); Holmes, Doolittle and Moore (1927); Ellis, Miller, Titus and Beverly (1933); Bisbey and his co-workers (1934); Wilcke, Nelson and Henderson (1934); and Russell (1934) pointed out that the addition of cod-liver oil to the diet of the hen, even as low as a 2% increase in weight, increased the amount of vitamin A in the egg yolk.

In the above-mentioned works consideration was given primarily to the fact that the addition of cod-liver oil to the hen's ration increased the amount of the vitamin A factor in the yolk of the egg. In 1935, DeVaney, Titus and Nestler discovered that when different groups of hens were fed different amounts of cod-liver oil in their ration, the yolks of the eggs laid by them contained correspondingly different amounts of the vitamin A factor. This information discovered by DeVaney and his co-workers was

ascertained while working with another experiment in which graded amounts of cod-liver oil were fed to different groups of hens. However, it bore strong implications on which the present study was based. Thus far no other work pointing out the relationship between the amount of cod-liver oil ingested by hens and the vitamin A content of the egg laid by them has been reported.

As pointed out earlier in these findings, hens fed a supplement of 4% in weight of cod-liver oil in their ration laid eggs containing approximately 46 units of vitamin A per gram of yolk. Those hens that were fed a supplement of 9% in weight of cod-liver oil laid eggs containing approximately 26 units of vitamin A per gram of yolk. Those hens fed no supplement of cod-liver oil in their ration laid eggs containing approximately 20 units of vitamin A per gram of yolk.

DeVaney et al. pointed out, in their findings in 1935, that hens fed a supplement of 1 to 2% in weight of cod-liver oil in their ration laid eggs containing approximately 40 units of vitamin A per gram of yolk. Those hens fed a supplement of 8% in weight of cod-liver oil in their ration laid eggs containing approximately 80 units of vitamin A per gram of yolk. Those hens which were kept on the basic ration with no supplement of cod-liver oil laid eggs containing approximately 20 units of vitamin A per gram of yolk.

There is a wide variation in the findings of these two studies with the exception of the results from the basic ration. In this ration the hens of both studies apparently produced the same amount of the vitamin A factor in their eggs. Therefore we may conclude that the basic rations were equal in vitamin A potency. The variation arose in the rations in which the supplement was added. As pointed out above, DeVaney et al. found that an 8% supplement produced eggs with 80 units of the factor and

1 to 2% produced eggs with 40 units of the factor. On the other hand, this study reports that 9% of the supplement produced eggs with 46 units of the factor and a 4% supplement produced eggs with 26 units of the factor.

The variation, as pointed out above, may be explained in the following way. The hens in the study by DeVaney et al. were fed for a period of 153 days before the first eggs were taken for a test as compared with a period of 71 days in the present study. It may be suggested that the length of time that an animal is fed a diet containing vitamin A has a definite influence on the amount of the factor which the animal may store (Sherman & Smith-1931). It is therefore altogether possible as well as probable that the 82-day advantage of the hens fed by DeVaney et al. accounted for the wide variation in the vitamin A content of the eggs in the two studies.

The age of the test animals (rats) has an influence on the response which may be received from the test food. It may be noted that the animals used by DeVaney et al. were put on the deficient diet at the age of 21 days, whereas the animals for this study were put on the deficient diet at the age of 27 days. This gave DeVaney et al.'s animals 6 days' advantage on those of this study. It has been pointed out that the younger the animal, the more ready the response to the test food (Sherman & Munsel-1925). Therefore it is possible that this factor played a great part in the variation in the vitamin A content of the eggs of the two studies.

It may also be pointed out that there is a possibility that the age of the hens used played a part in the content. As pointed out above, the younger the animal, the more ready the response to the test food (Sherman & Munsel-1925). The hens used by DeVaney and his co-workers (1935) were young pullets, laying for the first time the fall in which he

used them. In this study, the hens used were at least two years of age at the beginning of the feeding period. Again, there was an advantage in the possibility of storage by the animals in the former study.

It may be stated that the above-mentioned factors do not alter the fundamental facts involved. It was the objective in this study to show the relationship, if any, between the ingestion of graded amounts of cod-liver oil by hens and the content of the vitamin A factor in the egg yolk of the hens. The findings above do point out that there is a definite relationship which exists between these two factors. Hens fed a supplement of 9% in cod-liver oil laid eggs containing approximately twice the amount of vitamin A that was contained in eggs laid by hens fed a 4% supplement. Therefore, with the facts in mind concerning the feeding period of the chicks, the age of the rats, as well as the age of the hens, the findings in this study may be considered to have revealed some significant information concerning the amount of cod-liver oil ingested by the hen and the amount of vitamin A contained in the egg yolk. Not only that, it also re-emphasizes the fact that age is a factor in response and storage, as well as the fact that length of the feeding period is a factor in storage.

The study points out substantially that there is a definite relationship between the amount of cod-liver oil ingested by the hen and the vitamin A content of the egg yolk. This does not indicate, however, that it is safe to increase the vitamin A content of the hen's ration in order to produce eggs with a higher vitamin A potency. It has been pointed out (Mohler-1934) that any appreciable increase of vitamin A in the hen's ration above 2% for a long period of time decreases the laying capacity of the hens as well as the hatchability of the eggs.

CHAPTER VI

SUMMARY

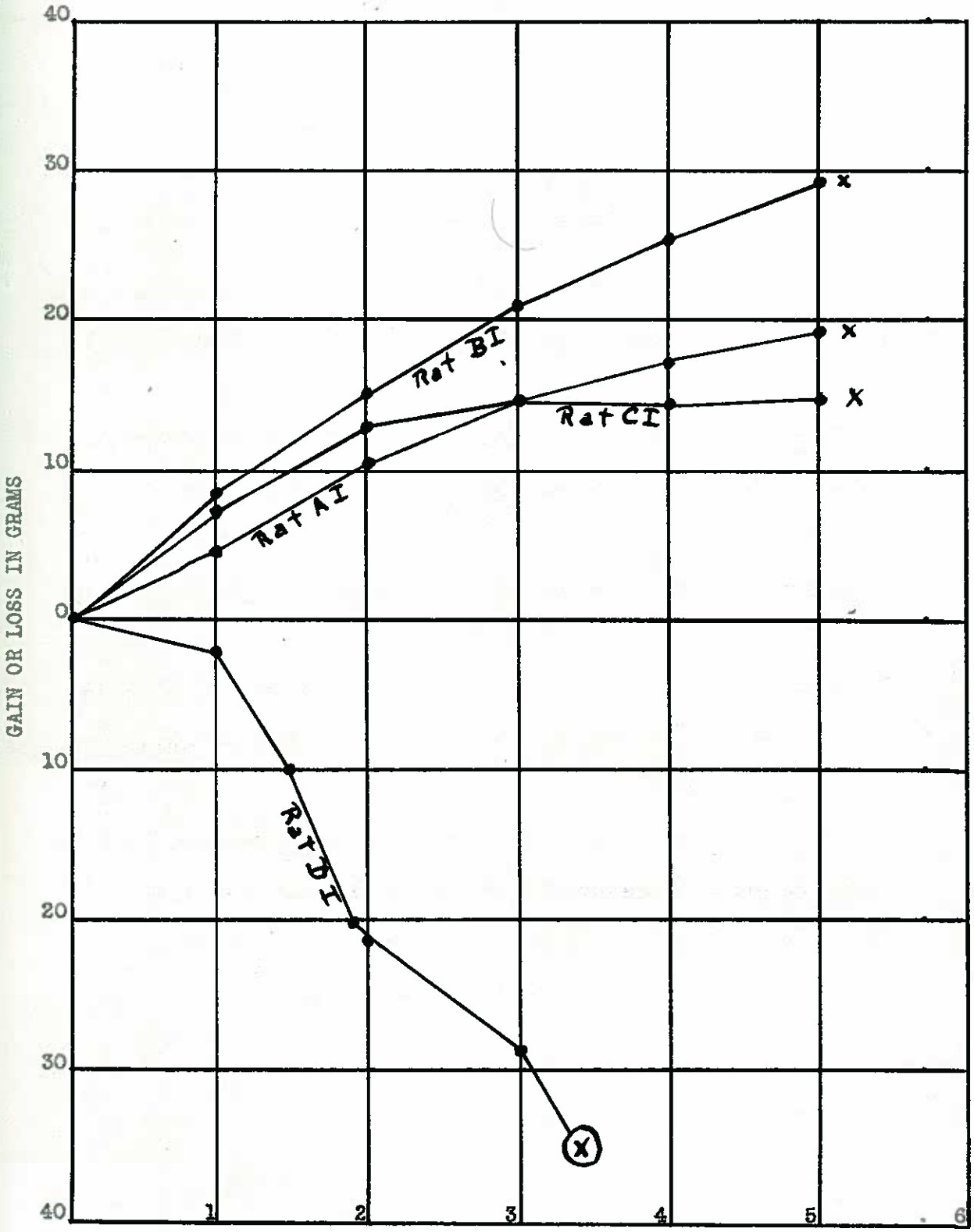
When young albino rats, between the ages of 21 and 29 days are fed a diet deficient in vitamin A, they show cessation of growth, inflammation of the eyes, ruffled fur, and general health weakness.

Animals depleted of the vitamin A factor decrease in weight rapidly. If they are continued on the deficient diet, they not only lose weight but will become increasingly inactive, develop respiratory trouble, and die within 35 days after being depleted.

Hens fed a mash with a supplement of cod-liver oil of at least 4%, over a period of at least 71 days, lay eggs the yolk of which is richer in vitamin A than eggs laid by hens fed the same mash containing no cod-liver oil supplement. If the cod-liver oil supplement is as much as 9%, the vitamin A content of the egg yolks is approximately twice the amount contained in eggs laid by hens fed the same mash with no cod-liver oil supplement.

It is not concluded that the hen's egg can be profitably increased in vitamin A by a supplement of cod-liver oil above 2%.

GROWTH CHART OF FEEDING TEST
AFTER THE DEPLETION PERIOD



Explanation of Chart

The chart covers the weight gain or loss after the end of the depletion period. The weight is reckoned in grams or fractional parts of a gram.

1. All animals' weights were checked at zero. The weight which was found at the end of the depletion period was reckoned as the zero weight.
2. Beginning at 0, the distance from one horizontal line to the other represents 10 grams in weight.
3. Beginning from the left-hand side of the page and counting the vertical lines to the right, the distance between each of them represents seven days or one week.
4. The weight was checked at intervals of a week, except in one case. That case was on the death of the control animal. Each week's weight is indicated by a heavy dark circle. The point of death of an animal is indicated by a circle inclosing an X.

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