THE EFFECT OF VARYING LEVELS OF CALCIUM INTAKE ON THE CALCIUM BALANCE, SHELL THICKNESS, AND BLOOD CALCIUM LEVEL OF WHITE LEGHORN PULLETS

by

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INTRODUCTION

The dietary requirement of various farm animals for calcium, in terms of total ration, is about 0.29 per cent for the lactating cow, 0.45 per cent for the sow, and 0.23 per cent for the ewe, according to Mitchell and McClure (1937). The level of dietary calcium that will sustain a high level of egg production, promote good hatchability and satisfactory shell texture has been set at 1.65 per cent by Norris et al (1934), 2.25 per cent or less by Miller and Bearse (1934), and 4.5 per cent by Bird (1937).

Although it is quite apparent that the calcium requirements of laying hens are considerably greater than those of other farm animals, the findings of the various investigators show wide disagreement. Undoubtedly these differences are due to variations in the fowls and in the experimental procedures used, variations existing in number of factors studied, sources of calcium, duration of test, and possibly others. The value of these experiments in establishing the minimal level of calcium required by the laying hen must be questioned, however, because no consideration has been given to the changes which occur in the calcium stores during the period of egg production.

That the changes in calcium stores must be considered in an attempt to arrive at the calcium requirements of laying hens was first suggested by Halnan (1925), as quoted by Russel and McDonald (1929), when he reported that birds in heavy production may be in a negative calcium balance, or in other words drawing on their calcium stores to obtain calcium for shell formation. Similar results have been reported by Russel and McDonald (1929) and by Common (1933, 1936, 1938).

The extent to which a laying bird may deplete her calcium stores has been studied by a few investigators. Deobald et al (1936), Edin and Andersson

(1937), and Common (1938) have employed different experimental procedures in arriving at values of 10, 20, and 25 per cent respectively as the amount of body calcium which a bird may withdraw for shell formation.

The present experiment was conducted to determine by means of calcium balance studies the amount of calcium which a bird may withdraw for shell formation, and to study the effect of varying levels of calcium intake on the calcium balance, shell thickness, and blood calcium level of laying pullets. It was hoped that the data obtained would present more evidence concerning the calcium requirements of laying hens.

REVIEW OF LITERATURE

Evidence has been presented by Common (1933, 1936, 1938), Morgan and Mitchell (1938), Deobald, Lease, and Hart (1936), and Edin and Andersson (1937) which suggests that chickens in production may use the calcium stored in their skeletons for egg shell formation.

Common (1933, 1936) showed that calcium was retained or stored in the bone as tricalcium phosphate and that it was lost from the body also as a calcium phosphate salt. His data confirm the view that when the calcium carbonate intake is sufficiently high, lime is transferred directly from the gut to the oviduct via the blood stream and that no extra phosphorus excretion occurs under these conditions. When the calcium carbonate intake is low, an insufficient amount of dietary calcium is available during the period of shell formation to provide calcium for an egg shell. Under these conditions phosphorus excretion was accelerated. This would indicate that in lieu of dietary calcium, previously stored calcium in the form of tricalcium phosphate was utilized for shell formation and this accentuated the excretion of the liberated phosphorus. These results were confirmed by Morgan and Mitchell (1938). They concluded also that the ability of a hen to utilize calcium seems to be an individual matter, the higher producing hens as a rule utilizing the greater percentage of the ingested calcium.

Halnan (1925) as quoted by Russel and McDonald (1929) concluded that calcium was not stored to any great extent in advance of sexual maturity and that the bird's need for calcium after the onset of laying was almost completely met by the increased utilization of calcium from the available food supply. Negative calcium balances were recorded during periods of heavy egg production. Common (1938) noted, however, that as much as 9.348 grams of calcium was stored during the 20-day period just preceding the laying of the first egg when a ration containing approximately 3.0 per cent calcium oxide was fed.

Russel and McDonald (1929) obtained a few slightly negative but chiefly slightly positive balances with laying pullets on a 1.4 per cent calcium ration for a period of five weeks. The birds, however, were in comparatively low production. Morgan and Mitchell (1938), however, demonstrated by balance studies that laying birds consuming liberal quantities of calcium in the form of oyster shell could have cumulative negative calcium balances as great as 9.46 grams at the conclusion of 10 fifteen-day periods.

The differences in the calcium intake and calcium outgo through egg shells of birds receiving a low calcium diet have been studied by Edin and Andersson (1937). They concluded that birds may withdraw 20 per cent of their body calcium for shell formation before production decreases to the point where the intake and outgo are the same.

Deobald, Lease, and Hart (1936) found that 10 per cent of the calcium of the bone could be used for egg shell formation since the percentage ash content of the breast bones and tibias of Brown Leghorn hens on a low calcium diet was 10 per cent less than that of the control groups receiving adequate amounts of calcium. It must be pointed out, however, that while a decrease in the percentage skeletal ash can serve as a measure of calcium loss from the skeleton, an accurate measurement of the loss can be made only by determining the total amount of calcium present in the body before and after depletion. The conclusion of Deobald and coworkers is valid provided that (1) the initial percentage of ash was the same for both birds and (2) the control bird neither stored nor lost calcium.

Common (1938) conducted calcium balance studies with White Wyandotte pullets using high and low levels of dietary calcium. Data were obtained for calcium retention (total calcium intake during the experiment less total calcium voided in eggs and the excreta). Over a period of 73 days one of his birds on the low calcium diet (.365 per cent CaO) had a negative calcium oxide balance of 9.344 grams. A slight positive or negative balance was maintained by the birds on the high calcium diet. At the conclusion of the balance trials the carcasses of the birds were analyzed for calcium content, and from the values obtained, together with the balance data, he was able to calculate the initial amount of calcium in the bodies of the birds at the beginning of the experiment. A knowledge of the initial body calcium and the gain or loss of calcium made it possible to determine the percentage loss or gain of body calcium during the experimental period. On the basis of the data obtained for his birds LM1 and LM4 he concluded that laying birds may utilize 25 per cent of their body calcium for shell formation, and this within a surprisingly short period of time. As Common points out, however, the data for one of his birds may not be of great value in determining the maximum possible loss of body calcium. This bird had not been laying for some time before she was killed and therefore had stored calcium to replace that which she had lost.

EXPERIMENTAL PROCEDURE

Balance studies were conducted on a plan similar to that used by Common (1938). However, all birds were kept on the experiment for 30 days, and the calcium balance was determined by three-day periods. As a result, it was possible to calculate the maximum loss or gain rather than the loss over the entire experimental period.

Experimental data were obtained for 18 Single Comb White Leghorn pullets of the Kansas State College flock. In order that the birds might become accustomed to the environmental conditions peculiar to the experiment, and in order to determine the date on which the birds laid their first egg, those pullets used were taken from the range and placed in the experimental cages a short time previous to sexual maturity.

The cages used in the experiment consisted of a series of 16 hen batteries separated each from the other by solid partitions. Individual feces containers were placed under each bird. Feed and water containers were constructed in such a manner as to minimize the loss of feed.

The birds were fed an all mash basal ration consisting of ground yellow corn 35 parts, ground wheat 25, ground oats 10.3, wheat shorts 10.3, wheat bran 8.25, dehydrated alfalfa 2.6, meat and bone meal 2.6, fish meal 2.6, soybean oil meal 2.6, salt 0.5, and cod liver oil[1] 0.25. To three lots of the basal ration, which contained 0.562 per cent calcium, was added sufficient calcium carbonate[2] to increase the calcium content of the three diets

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[1]The cod liver oil used contained 400 A.O.A.C. units of vitamin D and 3,000 U.S.P. units of vitamin A per gram.

[2]Limeolith, a precipitated calcium carbonate analyzing 99.43 per cent calcium carbonate, was used.

to 1.142, 2.111, and 3.174 per cent respectively. The rations contained 0.70 per cent phosphorus and 370 p.p.m. of manganese. Although the calcium to phosphorus ratio was different in each diet it was believed that there was sufficient vitamin D in the rations to insure the proper utilization of the calcium and phosphorus regardless of the ratio of the two.

Sixteen pullets, which were divided into four lots of four birds each, were started on the experiment. Lot 1 was fed the basal ration only. Lot 2 received the diet containing 1 per cent calcium, Lot 3 the diet containing 2 per cent calcium, and Lot 4 received the 3 per cent calcium ration. The rations, in pellet form, and distilled water were before the birds at all times. One bird in each of Lots 1 and 4 ceased egg production entirely and were killed before the duration of the study. Two other birds were subsequently placed on experiment to replace these two birds.

A feces marker consisting of approximately 1 gram of iron oxide, given by means of a capsule, was used to determine the beginning of the balance study. Data for computing the calcium balance of each bird were obtained periodically for ten 3-day periods. Collection of data began with the first egg and during the period immediately following. Data on calcium intake and the calcium outgo, through egg shells, egg contents and feces, were obtained.

Calcium oxide in the shells was determined by ashing the shells in covered porcelain dishes. A white ash of calcium oxide with constant weight was obtained by heating to 900° C. for three hours.

The weight of contents of the egg was determined by subtracting the weight of the damp shell and membranes, which had been wiped as free as possible from all adhering albumin, from the total weight of the egg. A 2-gram sample of a mixture of the contents of all eggs laid by each bird during each three-day period was analyzed for calcium. In analyzing, the sample was ashed, the ash dissolved in 2 cc. of concentrated HCl and diluted to 50 cc. with distilled water. Calcium was determined on a 5 cc. aliquot by the Wang (1935) method for determining the calcium of precipitated whole blood.

The collected feces were placed in 12-inch porcelain evaporating dishes and dried in a forced-draft electric oven at 100° C. for 24 hours. The dried feces were then ground in a Wiley mill and a two-gram sample taken for analysis. The calcium in the feces and also in samples of the feed was determined by the Association of Official Agricultural Chemists (1940) method for calcium oxide in mineral feeds.

At the conclusion of the balance study the birds were killed and the flesh carefully removed from the skeleton. In an endeavor to determine if the loss of calcium from the skeleton of the birds on the low calcium diet was from any particular portion of the skeleton or was lost equally from all portions, the skeleton was divided into its appendicular and axial parts. The bones were ground with a food chopper, ashed, the ash dissolved in 200 cc. of 1-3 HCl and diluted to 10 liters with distilled water. Calcium was determined on a 25 cc. aliquot by the same method used for analysis of the feeds. In view of the fact that Mitchell, Card and Hamilton (1931) have shown that the total calcium of both the flesh and feathers of four-pound White Leghorn pullets amounts to only 2 per cent of the total body calcium, and that Common (1938) has shown that the calcium in the flesh of birds on both high and low calcium intake is virtually the same, the calcium in the flesh and feathers of the birds was not used in calculating the percentage loss of body calcium.

Blood calcium was determined by a slight modification of the Wang (1935) method, the modification being made in order to take into consideration the difference in blood calcium between laying birds and man.

Shell thickness was determined for each shell with membranes intact by means of a micrometer caliper calibrated to thousandths of an inch. Four measurements were taken near the equatorial plane of each shell. Body weights were recorded at the end of every six days.

Warren and Schnepel (1940) and Conrad (1939) reported that temperatures greater than 90° F. will adversely affect egg shell thickness and the blood calcium level of fowls. For this reason the temperature of the room in which the birds were kept was maintained as nearly as possible between 70 and 75 degrees Fahrenheit. Temperatures were recorded on a Tycos thermograph.

EXPERIMENTAL RESULTS

Calcium Balance Studies

The balance data which are presented in Tables 7 to 24 of the appendix show that the birds which received 0.5 or 1.0 per cent calcium in the diet were in negative cumulative calcium balance, whereas all birds receiving the 2 and 3 per cent calcium diets, with the exception of Nos. 58(1) and 59, were in positive cumulative calcium balance.

Table 1 presents a summary of the balance data for each lot. The birds in Lot 1 had an average negative balance of 7.556 grams and withdrew an average of 46.7 per cent[3] of their skeletal calcium for shell formation. Individual withdrawals varied from 41 to 51 per cent. The birds in Lot 2 utilized an average of 34.38 per cent of their skeletal calcium, and had average negative calcium balances of 5.618 grams. Individual variations were from 27 to 44 per cent. In Lots 3 and 4 positive balances of 1.817 grams

[3]Percentage loss or gain of skeletal calcium was determined from loss or gain from period of greatest negative or positive cumulative balance and not from total balance for 10 periods.

and 2.838 grams respectively were maintained. This represents gains in skeletal calcium of 12.28 and 21.36 per cent respectively. In Lot 3 the variations were from -0.4 to +28 per cent, and in Lot 4 from -24 to +50 per cent. The bird on negative balance in Lot 4 was No. 58(1) and she ceased laying after four periods and was killed.

Figures 1 and 2 show that the average rate of withdrawal of skeletal calcium in Lots 1 and 2 was rapid. The maximum withdrawal in Lot 1 had been nearly reached by the end of the fifth period. Figures 3 and 4, however, show that the rate of deposition of skeletal calcium in Lots 3 and 4 was slow and was distributed over the entire experimental period.

Table 1. The balance data for groups of birds receiving varying amounts of calcium.

	Lot 1	Lot 2	Lot 3	Lot 4
Av. Total Ca Intake, grams	18.575	40.266	59.618	71.979
Av. Ca in Shells, grams	17.184	32.831	34.261	29.040
Av. Ca in Egg Contents, grams	.243	.490	.492	.381
Av. Ca in Feces	8.704	12.563	23.048	39.720
Av. Ca Outgo	26.131	45.884	57.801	69.141
Av. Ca Balance	-7.556	-5.618	+1.817	+2.838
Av. Percentage Loss or Gain of Skeletal Ca*	-46.74	-34.38	+12.28	+21.36

*Percentage loss or gain of skeletal calcium was determined from loss or gain from period of greatest negative or positive cumulative balance and not from total balance for 10 periods.

That 46.7 per cent is not the maximum skeletal calcium which White Leghorn pullets may withdraw is indicated by the fact that in most cases when the birds in Lot 1 started laying after having been out of production for a few days they withdrew more calcium for the formation of egg shells than that which they had stored while out of production. The fact that some of the birds on the basal diet were still laying at the close of the experiment suggests that further depletion might be possible.

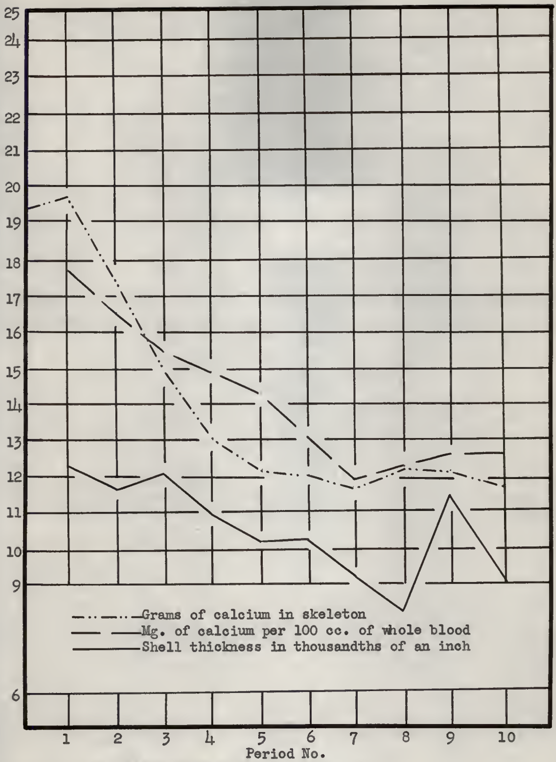

Fig. 1. Mean experimental data for Lot No. 1.

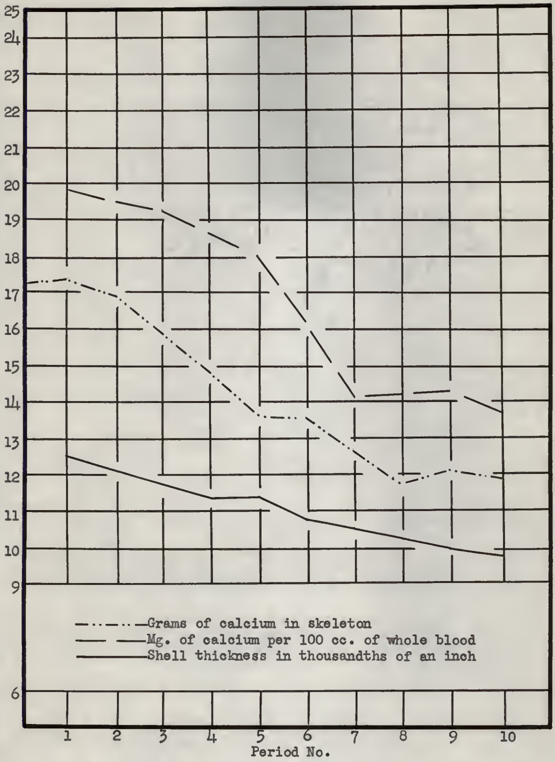

Fig. 2. Mean experimental data for Lot No. 2.

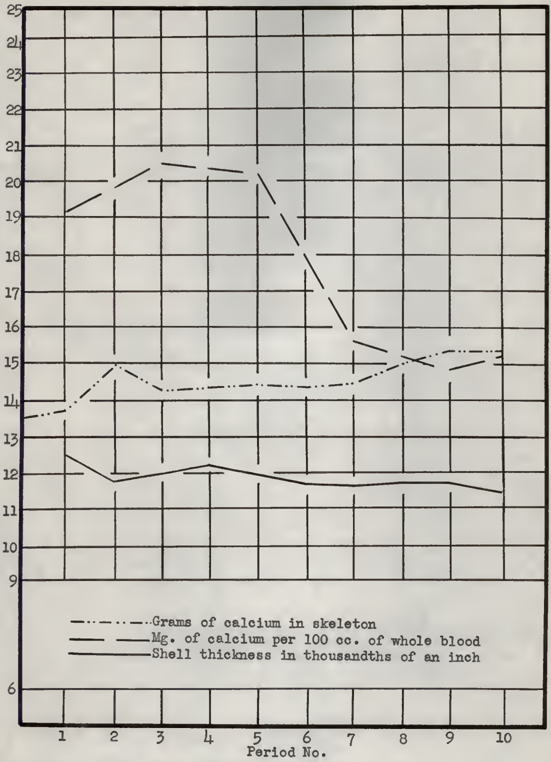

Fig. 3. Mean experimental data for Lot No. 3.

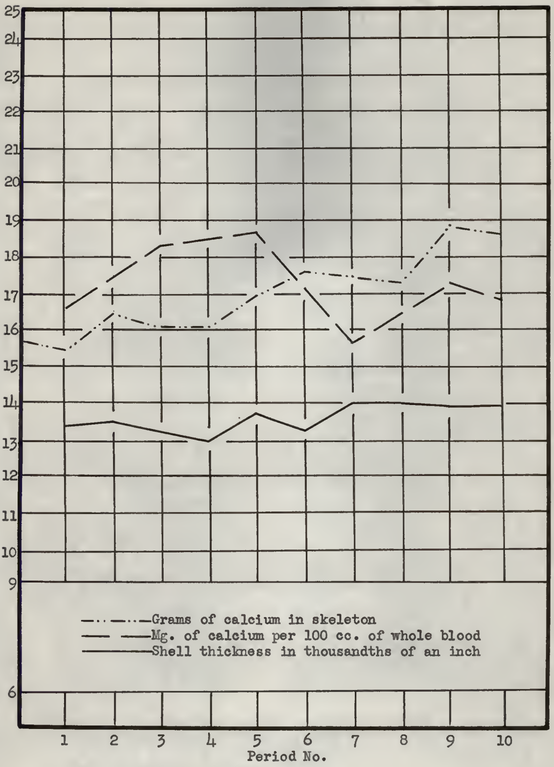

Fig. 4. Mean experimental data for Lot No. 4.

The average percentage loss of calcium for the birds in Lot 1 is much greater than that reported by Edin and Andersson (1937), Deobald et al (1936), or Common (1938). The results of the present experiment are in variance with those of Deobald et al and Edin and Andersson probably due to differences in experimental methods and methods of statistical analysis employed. These differences have been previously enumerated. The fact that the percentage loss in the present study was calculated from a point not more than three days from which the bird was in its greatest negative balance, rather than from the loss resulting during the entire experimental period is probably responsible for a withdrawal value nearly twice that reported by Common. The procedure followed in the present study eliminated the factor of calcium storage which results when birds are not laying. In the event that laying was not resumed so as to afford an opportunity to convert the stored calcium into egg shell the extent of withdrawal would be greatly obscured. That the stock used by Common was inferior with respect to egg laying ability is suggested by the fact that two birds on the low calcium diet produced only 13 and 11 eggs in 73 and 48 days respectively and hence had little opportunity to deplete their body reserves of calcium. In the present experiment the birds produced an average of 12 eggs in 30 days even though losing more skeletal calcium than did the birds of Common. The level of calcium in the diet may also have been a factor since the diet used by Common contained only 0.261 per cent calcium. It is possible that this level of calcium in the diet was so low as to provide insufficient calcium for the normal physiological processes of the birds. This could result in reducing the rate of lay, which in turn would make it impossible to determine the amount of calcium actually susceptible of being withdrawn.

That the skeletal calcium of the birds in negative calcium balance was withdrawn equally from all portions of the skeleton is shown by the data presented in Table 2.

Table 2. Percentage calcium in appendicular and axial skeleton of birds receiving varying amounts of calcium.

Lot No.	Bird No.	Percentage Calcium Appendicular	Percentage Calcium Axial
1	57	57.2	42.8
	71	58.7	41.3
	73	57.4	42.6
	77(1)	54.4	45.6
	77(2)	54.1	45.9
	Average	56.4	43.6
2	55	57.7	42.3
	60	56.7	43.3
	61	54.9	45.1
	78	59.0	41.0
	Average	57.1	42.9
3	56	55.8	44.2
	59	55.7	44.3
	62	55.5	44.5
	76	56.0	44.0
	Average	55.8	44.2
4	58(1)	56.4	43.6
	58(2)	55.4	44.6
	72	54.7	45.3
	75	58.6	41.4
	79	57.2	42.8
	Average	56.5	43.5

The ratio of calcium in the axial or appendicular skeleton to the total calcium present in the skeleton was approximately the same for all birds, the average ratio of all skeletons being 43.6 per cent axial and 56.4 per cent appendicular. This indicates that calcium is withdrawn from and stored in both divisions of the skeletal structure at the same rate.

Egg Production

The effect of a low calcium diet on subsequent laying performance was studied in growing pullets by Massengale and Platt (1930). A diet unsupplemented by calcium was fed during the first ten weeks. Two per cent of limestone was added to the ration for the next eight weeks following which the limestone was again removed from the ration. The average production of this group at 33 weeks of age was 0.5 egg per bird, whereas birds which had received 4 per cent or more of oyster shell in the ration for a corresponding period laid an average of 20.8 eggs per bird.

Buckner and Martin (1920) observed that the feeding of limestone or oyster shell ad lib increased egg production 69.4 per cent over a control group of pullets receiving an unsupplemented diet. The birds fed no mineral supplements continued to lay eggs until there was a general depletion of magnesium, phosphorus, and calcium in their bones, the breakdown occurring in about six months. These results were confirmed by Buckner, Martin and Peter (1923, 1925). In 1925 they observed that the production of yearling hens which were fed a low calcium diet for two months and then given calcium carbonate ad lib for 31 days increased the rate of production from 11 to 58 per cent.

Deobald, Lease, and Hart (1936) have reported that birds virtually ceased laying within 12 days after the abrupt removal of calcium supplements from their diet. Similar results had previously been reported by Buckner and associates (1930). They fed White Leghorn hens for 14 days on a diet consisting of yellow corn, skim milk, and calcium carbonate fed ad lib and then removed the calcium carbonate from the diet of half of the birds, the other half being continued on the calcium supplemented diet. In a period of six weeks the production of the birds deprived of calcium carbonate decreased 78 per cent while birds kept on the calcium supplemented diet increased 30 per cent in production. Likewise, Buckner, Martin, and Peter (1925) observed that when oyster shell was removed from the diet of confined hens having received it for six months, egg production decreased 50 per cent within 3 months. Birds which had access to range and were suddenly deprived of the calcium continued to lay at a satisfactory rate. Apparently sufficient mineral matter was obtained from the range to maintain good egg production. The addition of oyster shell to the diets of hens not having received any calcium supplements for six months trebled egg production within 3 months.

Inability to consume and utilize sufficient calcium to maintain a positive balance of calcium was shown by Morgan and Mitchell (1938) to result in a cessation of laying.

Edin and Andersson (1937) reported that when the body calcium of a bird on a low calcium intake has decreased to 80 per cent of normal, due to loss of calcium through egg shells, egg production decreases. Decreases in production of from 65 to 8 per cent were noted within 25 days.

Common (1938) observed that the normal egg laying cycle is markedly interfered with from the outset in birds on low calcium diets where dietary calcium must be supplemented with body calcium for egg formation. He suggests as a reason for this the more rapid mobilization of ingested calcium as compared with the mobilization of skeletal calcium for egg shell formation.

The results of the present study show that the average production per bird in Lots 1, 2, 3, and 4 for the 30-day period was 12, 21, 20, and 15 eggs respectively.

All birds in Lot 1, which received the 0.5 per cent calcium diet, ceased production for periods of at least 5 or more days one or more times during the experiment. The average number of days that the birds were on experiment before production ceased was 13 with the greatest number of days for any individual bird before production ceased being 16. The results obtained with birds 57, 71, and 73 as shown in figures 5, 6, and 7 of the appendix indicate that after the birds ceased laying, they stored calcium and when sufficient was stored, egg production continued until there was a depletion of the accumulated reserve. In some instances a return to the laying condition resulted in a withdrawal of calcium which exceeded the amount stored during sexual inactivity. Normal egg production was maintained by all the birds in Lots 2 and 3. The average production of Lot 2 was increased due to the high production of bird 61. The lower average production of the birds in Lot 4 is difficult to understand. The fact that bird 75 laid 20 eggs, and that other non-experimental birds kept under identical conditions, except for experimental handling, and fed the 3.0 per cent ration laid at a satisfactory rate indicates that the low production was due to some factor such as an unfortunate choice of birds.

These results suggest that rations containing as little as 1.0 per cent calcium will maintain satisfactory egg production for a period of at least 30 days following date of onset of sexual maturity.

Egg Shell Thickness

Buckner and Martin (1920) reported a decrease in shell weight from 5.1 grams to 2.9 grams when hens were limited for a period of six months to the calcium naturally occurring in the food. However, the percentage of calcium and phosphorus in the egg shells remained constant thus indicating that there is a stable equilibrium between the two elements. These results were confirmed by Buckner and coworkers (1930) when they observed that feeding a non-calcium supplemented diet decreased shell weight 21 per cent in a period of six weeks. Deobald et al (1936) reported a gradual diminution of the CO_2 free ash of the eggshells until the ash content of the shell was less than 25 per cent of normal when birds were abruptly deprived of the calcium in the ration. However, they were unable to obtain any shell-less eggs before production ceased.

The results obtained by Common (1936) also show that birds on a low calcium diet (0.365 per cent calcium oxide) lay eggs whose shells become progressively thinner. His data show that a bird on a low calcium ration will lay two or three eggs which contain about two grams, or a normal amount of calcium oxide, and then may decrease as much as from 2.039 grams to 0.714 grams.

Edin and Andersson (1937) observed that when the calcium content of the diet of laying birds was abruptly reduced, a decrease in the relative calcium content of the eggs is noticeable even the day after reduction of calcium in the feed. They report a decrease of 48 per cent in the calcium oxide content of eggs within a period of 25 days.

Riddle and Hanke (1921) observed in their studies on egg-laying ring doves that thin-shell eggs cannot be attributed to an inadequate supply of calcium in the feed.

In the present experiment the quality of the eggshell is expressed in terms of (1) measured shell thickness, and (2) the milligrams of calcium present per square centimeter of surface area of eggshell. These methods should provide a more accurate measure of shell quality than total shell weight or weight of calcium in the shell because variability due to differences in egg size is eliminated.

A summary of the measured shell thickness (in thousandths of an inch) and also calculations of the amount of calcium per square centimeter of surface area of the shells is presented in Table 3. Surface area of the shells, expressed as square centimeters, was computed by the formula $4.67W^{2/3}$, where W is the weight of the egg in grams. This method for determining surface area has been shown by Mueller and Scott (1940) to yield a smaller standard error than methods previously presented by Marshall and Cruickshank (1938) and Edin and Andersson (1937).

Table 3. Shell thickness of eggs from birds receiving varying amounts of calcium.

	Lot 1*	Lot 2*	Lot 3*	Lot 4
Av. Shell Thickness, thousandths of an inch, Period 1	12.0	12.1	11.4	12.1
Av. Shell Thickness, thousandths of an inch, Period 10	9.2	10.0	12.0	13.2
Av. mg. Ca per sq. cm., Period 1	26.1	26.8	25.6	28.5
Av. mg. Ca per sq. cm., Period 10	20.3	22.8	27.9	30.4

*Data for Lots 1, 2, and 3 begin with the period during which birds were first placed on their respective diets. It does not include Period 1 when on 3 per cent calcium.

Increases in average shell thickness of 5.3 and 9.1 per cent were obtained in Lots 3 and 4 respectively. The increase in Lot 4 was made even though Bird 79 of the lot had a considerable decrease in shell thickness. A decrease in shell thickness of 17.3 per cent resulted in Lot 2.

In Lot 1 the average decrease in thickness was from 12 to 9.2 thousandths of an inch. This represents a decrease of 23.3 per cent. Individual shells only 7.4 thousandths of an inch in thickness were obtained but in no case were shell-less eggs obtained before production ceased. This confirms the results of Deobald et al (1936).

The actual average thickness and amount of calcium per square centimeter of surface area also show that the level of dietary calcium affects the composition of the eggshells. The amount of calcium per unit of surface area increased slightly on diets containing 2.0 and 3.0 per cent calcium. Significant decreases in calcium were secured on the 0.5 and 1.0 per cent levels of intake. The mean milligrams of calcium per square centimeter was 20.3, 22.8, 27.9, and 30.4 for Lots 1 to 4 respectively.

That shell thickness tends to increase and decrease as skeletal calcium and blood calcium increase and decrease is shown in Figs. 1 to 22.

Calcium in Egg Contents

That the level of calcium in the ration does not appreciably affect the percentage of protein nor the percentage of calcium in the white and yolk of the egg was shown by Buckner, Martin and Peter (1925). Common (1936) and Buckner et al (1923) were unable to show any noticeable effect of dietary calcium on the amount of calcium oxide in the egg contents.

Data for the milligrams of calcium per gram of egg contents as obtained in the present study are shown in Table 4. The amount of calcium per gram of egg contents varies in many cases from period to period, and there is considerable variation between the averages of birds within the same lots. Lot 4, however, had an average of 14 per cent more calcium in egg contents than did Lot 1. Lot 2 averaged 5.2 per cent more calcium in egg contents than Lot 1, Lot 3 averaged 2.3 per cent more than Lot 2, and Lot 4, 4.4 per cent more than Lot 3. An analysis of the variance of the calcium in the egg contents of the eggs produced by the birds in the different lots is presented

Table 4. Milligrams of calcium per gram of egg content for birds used in this study.

Bird No.	Period 1	2	3	4	5	6	7	8	9	10	Mean	Average By Lots: Lot	Average
57	.380	.500	.481	.445	.386	.446	----	----	.529	.477	.466)		
71	.621	.696	.663	.583	.543	----	.486	.394	----	----	.573)		
73	.410	----	.474	.505	----	.414	.441	----	.412	.443	.448)	1	.493
77(1)	.474	.491	.487	.502	----	----	----	----	----	----	.488)		
77(2)	.452	.516	.502	.466	.458	.503	----	----	----	----	.489)		
55	.619	.600	.503	.559	.503	.450	.463	.476	.459	.500	.501)		
60	.593	.781	.472	.484	.409	----	.493	.434	.443	----	.502)	2	.518
61	.502	.604	.563	.636	.578	.515	.581	.600	.589	.619	.587)		
78	.471	.463	.467	.488	.509	.603	.452	.463	.497	.429	.484)		
56	.732	.712	.673	.645	.471	.486	.542	.542	.539	.659	.585)		
59	.523	.603	.569	.556	.514	.526	.538	.558	.573	.487	.547)	3	.540
62	.616	----	.667	.553	.563	.519	.537	.563	.491	.583	.559)		
76	.478	.493	.494	.392	.597	.485	.513	.490	.470	.461	.488)		
58(1)	.733	----	.620	.644	.612	.559	----	----	----	----	.634)		
58(2)	.552	.551	.563	.562	.569	.570	.535	.502	----	----	.550)		
72	.625	.666	.505	.458	.468	.549	.526	.544	.597	.621	.556)	4	.564
75	.555	.518	.540	.528	.551	.512	.564	.553	.577	.548	.545)		
79	.584	.562	.529	----	.547	----	.525	.524	.513	.509	.537)		

in Table 5. An F value, Snedecor (1938, p. 184), of 8.8 is highly significant with the degrees of freedom represented in the analysis.

Table 5. Analysis of variance of calcium in egg contents of eggs of 18 birds classified by individuals and lots.

Source of variation	Degrees of freedom	Sum of squares	Mean square
Total	149	.801	
Between Ca levels	3	.114	.038
Between birds within Ca levels	14	.121	
Within birds	132	.566	.0043

$F = .038/.0043 = 8.8$

Blood Calcium

Buckner and associates (1930) studied the effect of varying the level of calcium intake on the blood calcium of laying hens. They found that birds which were kept on a diet of yellow corn, skim milk, and calcium carbonate ad lib for six weeks maintained a constant blood calcium level, whereas the blood calcium of birds which were deprived of the calcium carbonate decreased from 20.9 mg. to 16.1 mg. per 100 cc. of blood.

Deobald et al (1938) demonstrated that as long as the calcium content of the ration was suboptimal and inhibited egg production, the blood calcium shows regular, almost cyclic, variations which are quite characteristic for each individual hen. When the calcium was removed from the ration of heavily laying pullets, laying continued for about ten days with the blood calcium content decreasing from 16 mg. to a level of about 10 mg. per 100 cc. of whole blood.

The effect of fasting upon the calcium content of blood plasma of laying hens was investigated by Laskowski (1934). Short fasts of 2 to 4 days did not interrupt egg laying or plasma calcium while fasts of 4 to 6 days interrupted egg laying for several days and reduced the plasma calcium level to that of non-laying birds.

In the present experiment all data for blood calcium are presented as milligrams of calcium per 100 cc. of whole blood. Data are not available for some periods because of difficulties arising in experimental procedure.

Results obtained for individual birds are presented in Tables 7 to 24 of the appendix. A summary of the results is presented in Table 6.

Table 6. Influence of different levels of calcium intake on blood calcium.

	Lot No.			
	1	2	3	4
Mean mg. blood Ca, Period 1	17.815	19.885	19.141	16.614
Mean mg. blood Ca, Period 10	9.356	13.764	15.481	16.992

Blood calcium values decreased from 17.8 mg. to 9.4 mg., from 19.7 mg. to 13.8 mg., and from 19.1 mg. to 15.5 mg. on diets containing 0.5, 1.0 and 2.0 per cent calcium respectively. Three per cent calcium in the diet enabled the hens of Lot 4 to maintain a high level of blood calcium, a slight increase being noted. The blood calcium for all birds in Lot 1 decreased from the first to the last period. In Lot 2 the blood calcium of all birds except No. 61 decreased. The records show that this bird consumed considerably more feed than the other birds in the lot. This would increase the calcium intake, making it possible to maintain a high level of blood calcium. The blood calcium of all individual birds in Lot 3 decreased. In two instances slight decreases in blood calcium were noted in Lot 4 birds. The remaining three birds of this lot showed slight increases in blood calcium.

Of particular interest are the increases in blood calcium associated with periods of non-laying and positive calcium balances. That variations in blood calcium may be related to periods of withdrawal and storage of calcium is suggested by the present study. This is clearly shown in Figs. 1 to 22 where it will be noted that the blood calcium curve tends to parallel the curve representing changes in skeletal calcium.

DISCUSSION

The data obtained in this experiment show that the level of calcium in the diet has a definite effect on the calcium balance, egg production, shell thickness, calcium in egg contents, and blood calcium level of laying pullets. Diets of 0.5 and 1.0 per cent calcium did not furnish sufficient calcium to meet the needs of the birds whereas 2.0 and 3.0 per cent calcium in the diet were sufficient to meet the requirements for the physiological processes studied except that 2.0 per cent calcium was insufficient to maintain the blood calcium level.

The present study has shown that laying birds are capable of storing dietary calcium in their skeletons and of drawing upon their skeletal calcium for shell formation. It was also shown that the amount of dietary calcium available to the bird determines whether calcium is stored or withdrawn and the rate at which these changes take place. Birds receiving a diet containing only 0.5 per cent calcium withdrew an average of 47 per cent of their skeletal calcium for shell formation and did so within about 15 days. Birds receiving 1.0 per cent calcium utilized an average of 34 per cent of their skeletal calcium and required an average of about 25 days to withdraw the greater portion of this. The birds fed 2.0 and 3.0 per cent calcium diets increased in skeletal calcium by 12 and 21 per cent, respectively.

Limiting the calcium in the diet fed to pullets just entering production to that occurring in feed materials with no calcium supplements has been shown in the present study to cause at least a temporary cessation of laying within 13 days. That birds can continue production for some time while in a negative calcium balance has been demonstrated, however. Birds with a cumulative negative calcium balance of 9 grams obtained over a period of 30 days continued satisfactory egg production during the entire period. No doubt, however, the birds which were in negative balance for the entire experiment, but continued to lay at a satisfactory rate, would have ceased laying, temporarily at least, when they had withdrawn all of their skeletal calcium which was available for shell formation.

A diet containing 0.5 per cent calcium decreased shell thickness from .012 inch to .0092 inch. A 1.0 per cent calcium diet likewise caused a decrease in shell thickness. Increases in shell thickness, however, resulted from feeding 2.0 and 3.0 per cent calcium diets. The average thickness of the shells obtained on the 3.0 per cent calcium diet was greater than the average thickness of those obtained on the 2.0 per cent calcium diet. The rate of increase in shell thickness during the experimental period, however, was practically the same in the shells obtained by feeding the two higher levels of calcium.

Although previous workers were unable to show an effect of dietary calcium on the amount of calcium in the egg contents, the results of the present study show significant differences as a result of varying the Ca level of the diet. The fact that a difference of 14 per cent between birds receiving 0.5 and 3.0 per cent calcium was noted and that graded increases in dietary calcium caused corresponding increases in the amount of calcium in the egg contents suggests that the level of dietary calcium influences the calcium found in the egg contents.

Although 2.0 per cent calcium in the diet was sufficient to maintain a positive skeletal calcium balance and shell thickness it was not sufficient to maintain the blood calcium level of the birds of this experiment. Feeding diets containing 0.5 and 1.0 per cent calcium likewise caused decreases in blood calcium. The greatest decrease occurred in the birds fed 0.5 per cent calcium, in which case the decrease in blood calcium tended to follow the withdrawal of skeletal calcium until the blood calcium content reached the level for non-laying birds, which resulted when they had withdrawn as much skeletal calcium for shell formation as possible. Three per cent calcium in the diet was sufficient to maintain normal blood calcium values.

On the basis of the results obtained it may be concluded that the calcium requirement of laying hens as measured by (1) balance studies, (2) egg production, (3) shell thickness, and (4) blood calcium, and under the conditions described, rests between the 2 and 3 per cent level.

SUMMARY

1. The effect of varying levels of dietary calcium on the calcium balance, egg production, shell thickness, calcium in egg contents, and blood calcium of White Leghorn pullets has been studied.

2. Laying pullets receiving 0.56 and 1.14 per cent dietary calcium were in negative calcium balance whereas birds receiving 2.11 and 3.17 per cent calcium were in positive calcium balance.

3. Data are presented which show that laying birds may utilize at least 47 per cent of their skeletal calcium for shell formation.

4. Satisfactory egg production was maintained for a period of 30 days by birds receiving 1, 2, and 3 per cent calcium. Birds receiving only 0.5

per cent calcium in the ration ceased production temporarily at least after a period of 13 days.

5. Shell thickness decreased 23 and 17 per cent respectively in the lots where 0.5 and 1.0 per cent calcium were fed and increased slightly in lots where 2 and 3 per cent calcium were included in the ration.

6. A difference of 14 per cent in the amount of calcium in the egg contents of eggs produced on the 0.5 and 3.0 per cent calcium rations was observed.

7. Blood calcium levels of birds receiving 0.5, 1.0, and 2.0 per cent calcium were decreased. The decrement was in direct relation to the amount of calcium in the diet. Three per cent calcium in the diet was sufficient to maintain the blood calcium level.

8. The calcium requirement of laying pullets confined to laying cages can be met by the inclusion of slightly more than 2.1 per cent calcium (5.2 per cent calcium carbonate) in the ration.

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LITERATURE CITED

Association of Official Agricultural Chemists, Washington, D. C.
Methods of analysis, 5th ed. 1940. 757 p.

Bird, S.
Fecundity and reproductive ability in closely confined fowl. Sci. Agr. 17:359-375. Feb. 1937.

Buckner, G. D., and Martin, J. H.
The effect of calcium on the composition of the eggs and carcass of laying hens. Jour. of Biol. Chem. 41:195-203. Feb. 1920.

Buckner, G. D., Martin, J. H., and Insko, W. M. Jr.
The blood calcium of laying hens varied by the calcium intake. Amer. Jour. Physiol. 94:692, 695. Sept. 1930.

Buckner, G. D., Martin, J. H., and Peter, A. M.
Calcium metabolism in the laying hen. Ky. Agr. Expt. Sta. Bul. 250:333-367. 1923.

__________. Effect of a calcium carbonate supplement in the diet of hens, on the weight, protein content, and calcium content of the white and yolk of their eggs. Amer. Jour. Physiol. 72:458-463. May, 1925.

Common, R. H.
Observations on the mineral metabolism of pullets. Jour. Agr. Sci. 23:555-570. Oct. 1933.

__________. Observations on the mineral metabolism of pullets II. Jour. Agr. Sci. 26:85-100. Jan. 1936.

__________. Observations on the mineral metabolism of pullets III. Jour. Agr. Sci. 28:347-366. July, 1938.

Conrad, Ralph M.
The effect of high temperatures on the blood calcium of the laying hen. Poultry Sci. 8:327-329. July, 1939.

Deobald, H. J., Christianson, J. B., Hart, E. B., and Halpin, J. G.
The relationship between blood calcium and blood phosphorus and the effect of variations in the calcium content of the ration on ovulation and blood calcium changes in the laying pullet. Poultry Sci. 17:114-119. March, 1938.

Deobald, H. J., Lease, E. J., and Hart, E. B.
Studies on the calcium metabolism of laying hens. Poultry Sci. 15:179-185. March, 1936.

Edin, H., and Andersson, A.
Kalkstensgrus, ostronskal och musselskal som kompletteringsfoder till kalkfattiga foderstater vid hög äggproduktion. Kungl. Lantbruks. Akademiens Handlingar och Tidskrift, 76:435-492. 1937.

Laskowski, M.
Calcium and phosphorus content of blood plasma of laying hens during fasting. Inter. Rev. Poul. Sci. 8:24. 1934.

Marshall, W., and Cruickshank, D. B.
The function of the cuticle in relation to the porosity of eggs. Jour. Agr. Sci. 28:24-42. Jan. 1938.

Massengale, O. N., and Platt, C. S.
Effect of calcium from different sources on the growth and egg production of poultry. Poultry Sci. 9:240-246. May, 1930.

Miller, Wayne M., and Bearse, G. E.
Phosphorus requirements of laying hens. Wash. Agr. Expt. Sta. Bul. 306:1-19. 1934.

Mitchell, H. H., Card, L. E., and Hamilton, T. S.
A technical study of the growth of white leghorn chickens. Ill. Agr. Expt. Sta. Bul. 367:83-139. 1931.

Mitchell, H. H., and McClure, F. J.
Mineral nutrition of farm animals. Natl. Res. Council Bul. 99. 135 p. April, 1937.

Morgan, C. L., and Mitchell, J. H.
The calcium and phosphorus balance of laying hens. Poultry Sci. 17:99-104. March, 1938.

Mueller, Clyde D., and Scott, H. M.
The porosity of the egg shell in relation to hatchability. Poultry Sci. 19:163-166. 1940.

Norris, L. C., Heuser, G. F., Ringrose, A. J., and Wilgus, H. S. Jr.
Studies of the calcium requirements of laying hens. Poultry Sci. 13:308-309. Nov. 1934.

Riddle, O., and Hanke, M. C. E.
Effect of feeding soluble calcium salts upon reproductive secretions and upon the total inorganic constituents of the egg shell. Amer. Jour. Physiol. 57:264-274. Sept. 1921.

Russel, Walter C., and McDonald, F. G.
The utilization of the calcium of calcium carbonate and citrate by laying and non-laying pullets. Jour. Biol. Chem. 84:463-473. Oct. 1929.

Snedecor, George W.
Statistical methods. Ames, Iowa. Collegiate Press. 378 p. 1938.

Wang, C. C.
Improvements in the methods for calcium determinations in biological matter. Jour. Biol. Chem. 111:443-453. 1935.

Warren, D. C., and Schnepel, R. L.
The effect of air temperature on egg shell thickness in the fowl. Poultry Sci. 19:67-72. 1940.

A P P E N D I X

Table 7. Calcium balance data for Bird No. 57 of Lot No. 1. All weights are in grams.

	Period										
	1*	2	3	4	5	6	7	8	9	10**	Total
Ca Intake	9.502	1.936	1.673	1.435	1.182	1.192	1.212	1.361	1.240	1.171	21.904
Ca in Shells	1.653	2.994	3.062	1.573	2.769	1.417	----	----	3.108	3.303	19.879
Ca in Egg Contents	.014	.041	.040	.019	.028	.019	----	----	.038	.062	.261
Ca in Feces	6.479	1.447	.534	.337	.213	.179	.325	.215	.190	.137	10.182
Ca Outgo	8.146	4.482	3.636	1.929	3.010	1.615	.325	.215	3.336	3.502	30.196
Ca Balance	+1.356	-2.546	-1.963	-.494	-1.828	-.423	+.887	+1.146	-2.096	-2.331	-8.292
Ca Balance from End of Period 1 . . .											-9.648
Ca in Skeleton, Final . . .											10.468
Ca in Skeleton, End of Period 1 . . .											20.116
Percentage Loss of Skeletal Calcium . . .											47.96
Feed Intake	299.3	344.3	297.6	255.2	210.2	212.0	215.6	242.1	220.6	208.3	2505.2
No. of Eggs	2	2	2	1	2	1	----	----	2	3***	15
Wt. of Eggs	83.3	91.4	92.9	47.2	80.7	48.0	----	----	97.8	151.0	692.3
Wt. of Shells	9.5	9.3	9.7	4.8	8.7	4.4	----	----	9.7	11.0	67.1
Wt. of Contents	73.8	82.1	83.2	42.4	72.0	43.6	----	----	88.1	140.0	625.2
Body Weight	1675	----	1700	----	1650	----	1600	----	1625	1460	
Av. Shell Thickness	12.6	11.8	11.9	10.9	10.8	10.7	----	----	11.2	8.1	
Blood Ca	13.056	----	12.524	----	12.276	----	13.130	----	12.060	6.736	

* The feed consumed during Period 1 contained 3.1747% calcium.

** Period 10 consisted of four days.

*** The last egg was removed from the oviduct.

Table 8. Calcium balance data for Bird No. 71 of Lot No. 1. All weights are in grams.

	Period										
	1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	5.238	2.062	1.678	1.655	1.781	2.053	1.564	1.526	1.589	2.137	21.283
Ca in Shell	3.034	1.599	3.575	4.684	2.875	———	2.760	2.173	———	1.206	21.906
Ca in Egg Contents	.046	.029	.058	.080	.053	———	.039	.035	———	.015	.355
Ca in Feces	2.153	1.744	.832	.631	.514	.625	.598	.447	.624	.717	8.885
Ca Outgo	5.233	3.372	4.465	5.395	3.442	.625	3.397	2.655	.624	1.938	31.146
Ca Balance	+.005	-1.310	-2.787	-3.740	-1.661	+1.428	-1.833	-1.129	+.965	+.199	-9.863
Ca Balance from end of Period 1 to end of Period 8 . . .											-11.032
Ca in Skeleton, Final . . .											11.646
Ca in Skeleton at end of Period 1 . . .											21.514
Percentage Loss of Skeletal Calcium . . .											51.28
Feed Intake	165.0	366.7	298.4	294.3	316.8	365.1	278.1	271.4	282.5	380.0	2676.3
No. of Eggs	2	1	2	3	2	———	2	2	———	1**	15
Wt. of Eggs	83.0	47.1	98.0	151.3	105.5	———	88.3	94.7	———	41.9	709.8
Wt. of Shells	9.1	5.0	10.5	14.0	8.8	———	8.4	6.7	———	3.7	66.2
Wt. of Contents	73.9	42.1	87.5	137.3	96.7	———	79.9	88.0	———	38.2	643.6
Body Weight	1750	———	1830	———	1775	———	1750	———	1675	1725	
Av. Shell Thickness	13.4	11.8	12.6	11.0	10.2	———	10.4	8.0	———	9.0	
Blood Ca	———	———	25.316	———	16.766	———	10.706	———	8.118	———	

* The feed consumed during Period 1 contained 3.1747% calcium.
** Egg was removed from oviduct.

Table 9. Calcium balance data for Bird No. 73 of Lot No. 1. All weights are in grams.

	Period										Total
	1*	2	3	4	5	6	7	8	9	10	
Ca Intake	7.130	1.522	1.230	.967	1.341	1.139	1.185	1.420	1.655	.966	18.555
Ca in Shell	1.712	——	3.765	2.206	——	1.690	2.234	——	1.728	1.787	15.122
Ca in Egg Contents	.018	——	.048	.029	——	.021	.048	——	.020	.022	.206
Ca in Feces	5.630	2.269	.437	.492	.528	.161	.144	.194	.258	.382	10.495
Ca Outgo	7.360	2.269	4.250	2.727	.528	1.872	2.426	.194	2.006	2.191	25.823
Ca Balance	-.230	-.747	-3.020	-1.760	+.813	-.733	-1.241	+1.226	-.351	-1.225	-7.268
Ca Balance from End of Period 1											-7.038
Ca in Skeleton, Final											10.141
Ca in Skeleton, End of Period 1											17.179
Percentage Loss of Skeletal Calcium											40.97
Feed Intake	224.6	270.6	218.8	172.0	238.5	202.6	210.7	252.6	294.3	171.8	2256.5
No. of Eggs	1	——	2	2**	——	1	2	——	1	1	10
Wt. of Eggs	48.3	——	112.1	56.5	——	57.1	115.6	——	53.5	54.0	497.1
Wt. of Shells	5.4	——	11.4	5.4	——	5.3	7.7	——	5.3	4.8	45.3
Wt. of Contents	42.9	——	100.7	51.1	——	51.8	107.9	——	48.2	49.2	451.8
Body Weight	1750	——	1775	——	1725	——	1625	——	1730	1700	
Av. Shell Thickness	12.9	——	13.1	11.9	——	11.4	8.0	——	11.9	10.4	
Blood Ca	16.632	——	12.726	——	19.296	——	12.736	——	12.000	11.312	

* The feed consumed during Period 1 contained 3.1747% calcium.

** One of the eggs was broken and the shell recovered from the feces container.

Table 10. Calcium balance data for Bird No. 77(1) of Lot No. 1. All weights are in grams.

	Period 1	2	3	4	5*	6	7	8	9	10	Total
Ca Intake	1.202	1.306	1.177	1.016	.044						4.745
Ca in Shell	1.101	3.318	3.442	3.019	——						10.880
Ca in Egg Contents	.019	.041	.044	.046	——						.150
Ca in Feces	1.765	.420	.254	.173	.019						2.631
Ca Outgo	-1.683	-2.473	-2.563	-2.172	+.025						-8.916
Ca Balance to End of Period 4											-8.891
Ca in Skeleton, End of Period 4											9.403
Ca in Skeleton, Initial											18.294
Percentage Loss of Skeletal Calcium											48.60
Feed Intake	213.8	232.2	209.4	180.6	7.9						843.9
No. of Eggs	1**	2	2	2	——						7
Wt. of Eggs	45.9	93.6	101.3	99.6	——						340.4
Wt. of Shells	3.5	9.7	10.5	8.9	——						32.6
Wt. of Contents	42.4	83.9	90.8	90.7	——						307.8
Body Weight	1525	——	1475	——	1250						
Av. Shell Thickness	9.9	12.0	12.0	10.6	——						
Blood Ca	21.412	——	13.332	——	9.306						

* The bird became lame and was killed at the end of Period 5.

** One egg laid previous to beginning of experiment.

Table 11. Calcium balance data for Bird No. 77(2) of Lot No. 1. All weights are in grams.

	Period 1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	7.556	1.636	2.012	2.345	2.296	2.049	1.934	2.099	2.010	2.453	26.390
Ca in Shell	1.855	4.679	3.262	3.155	2.783	2.399	———	———	———	———	18.133
Ca in Egg Contents	.019	.068	.046	.043	.043	.025	———	———	———	———	.244
Ca in Feces	3.517	1.090	.779	.861	.866	.664	.763	1.248	.726	.811	11.325
Ca Outgo	5.391	5.837	4.087	4.059	3.692	3.088	.763	1.248	.726	.811	29.702
Ca Balance	+2.165	-4.201	-2.075	-1.714	-1.396	-1.039	+1.171	+.851	+1.284	+1.642	-3.312
Ca Balance from End of Period 1 to End of Period 6 . . .											-10.425
Ca in Skeleton, Final . . .											17.749
Ca in Skeleton, End of Period 1 . . .											23.226
Percentage Loss of Skeletal Calcium . . .											44.88
Feed Intake	238.0	291.0	357.9	417.1	408.3	364.4	343.9	373.3	357.4	436.2	3587.5
No. of Eggs	1	3	2	2	2	1	———	———	———	———	11
Wt. of Eggs	46.4	145.7	101.8	103.1	101.8	52.7	———	———	———	———	551.5
Wt. of Shells	5.3	14.2	9.8	9.8	8.8	4.0	———	———	———	———	51.9
Wt. of Contents	41.1	131.5	92.0	93.3	93.0	48.7	———	———	———	———	499.6
Body Weight	1980	———	2000	———	2020	———	1950	———	2050	2100	
Av. Shell Thickness	12.9	11.2	10.8	10.5	9.6	8.7	———	———	———	———	
Blood Ca	12.663	———	13.930	———	14.800	———	11.312	———	18.676	20.097	

* The feed consumed during Period 1 contained 3.1747% calcium.

Table 12. Calcium balance data for Bird No. 55 of Lot No. 2. All weights are in grams.

	Period 1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	7.562	3.649	3.061	2.986	3.659	3.540	3.351	3.298	3.568	3.874	34.488
Ca in Shell	1.742	3.158	3.060	2.999	4.423	3.055	2.790	2.690	2.686	3.761	30.364
Ca in Egg Contents	.023	.048	.042	.046	.063	.040	.041	.039	.039	.065	.446
Ca in Feces	5.047	1.553	.942	1.015	.789	.794	.965	.898	.620	.851	13.521
Ca Outgo	6.812	4.759	4.044	4.060	5.275	3.889	3.796	3.627	3.345	4.677	44.331
Ca Balance	+.750	-1.110	-.983	-1.134	-1.616	-.349	-.445	-.329	+.223	-.850	-5.843
Ca Balance from end of Period 1											-6.593
Ca in Skeleton, Final											9.105
Ca in Skeleton at end of Period 1											15.698
Percentage Loss of Skeletal Calcium											44.11
Feed Intake	238.2	319.4	267.9	256.1	320.3	309.8	293.3	288.7	312.3	339.1	2945.1
No. of Eggs	1**	2	2	2	3	2	2	2	2	3	21
Wt. of Eggs	42.2	89.9	92.4	92.0	138.3	97.2	96.2	90.9	93.3	142.0	974.4
Wt. of Shells	5.1	9.4	9.4	8.9	13.5	9.3	8.7	8.0	8.3	11.8	92.4
Wt. of Contents	37.1	80.5	83.0	83.1	124.8	87.9	87.5	82.9	85.0	130.2	882.0
Body Weight	1450	———	1510	———	1450	———	1450	———	1425	1400	
Av. Shell Thickness	13.1	12.0	12.0	11.2	11.4	11.3	10.0	10.3	9.6	9.4	
Blood Ca	22.220	———	———	23.836	18.786	———	9.494	14.256	———	11.514	

* The feed consumed during Period 1 contained 3.1747g calcium.

** One egg was laid previous to beginning of experiment.

Table 13. Calcium balance data for Bird No. 60 of Lot No. 2. All weights are in grams.

	Period 1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	3.892	2.800	2.707	3.536	4.264	3.833	4.046	4.186	3.378	4.443	37.085
Ca in Shell	1.564	1.291	5.359	3.913	3.614	3.441	4.944	3.151	3.111	2.727	33.115
Ca in Egg Contents	.026	.037	.070	.050	.042	.045	.076	.044	.046	———	.436
Ca in Feces	1.842	1.618	.799	.707	1.072	.989	.927	.821	.633	.839	10.247
Ca Outgo	3.432	2.946	6.228	4.670	4.728	4.475	5.947	4.016	3.790	3.539	43.798
Ca Balance	+.460	-.146	-3.521	-1.134	-.463	-.642	-1.901	+.170	-.412	+.877	-6.713
Ca Balance from end of Period 1											-7.173
Ca in Skeleton, Final											11.638
Ca in Skeleton at end of Period 1											18.811
Percentage Loss of Skeletal Calcium											38.13
Feed Intake	122.6	245.1	237.0	309.5	373.2	335.5	354.2	366.4	295.7	388.9	3028.1
No. of Eggs	1	1	3	2	2	2	3	2	2	2**	20
Wt. of Eggs	48.8	52.7	164.0	115.5	112.4	109.3	169.0	111.5	114.2	———	997.4
Wt. of Shells	5.1	5.3	15.8	11.7	10.9	10.0	15.3	9.5	9.5	———	93.1
Wt. of Contents	43.7	47.4	148.2	103.8	101.5	99.3	153.7	102.0	104.7	———	904.3
Body Weight	1850	———	1910	———	1850	———	1900	———	1900	1775	
Av. Shell Thickness	13.0	12.1	11.8	12.4	11.6	11.2	11.1	10.1	9.8	9.8	
Blood Ca	———	———	20.808	———	19.594	———	15.576	———	12.672	17.372	

* The feed consumed during Period 1 contained 3.1747% calcium.

** Eggs broke and the shells were recovered from the feces container.

Table 14. Calcium balance data for Bird No. 61 of Lot No. 2. All weights are in grams.

	Period										
	1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	6.364	4.489	4.907	4.715	4.450	4.649	5.084	4.804	4.719	5.006	49.187
Ca in Shell	3.243	4.267	4.591	4.505	3.456	3.106	4.724	4.764	3.312	4.427	40.395
Ca in Egg Contents	.056	.078	.088	.082	.051	.056	.084	.086	.062	.084	.727
Ca in Feces	2.954	.943	1.259	1.110	1.239	1.142	1.077	1.055	1.031	1.173	12.983
Ca Outgo	6.253	5.288	5.938	5.697	4.746	4.304	5.885	5.905	4.405	5.684	54.105
Ca Balance	+.111	-.799	-1.031	-.982	-.296	+.345	-.801	-1.101	+.314	-.678	-4.918
Ca in Skeleton, Final											13.172
Ca in Skeleton, Original											18.090
Percentage Loss of Skeletal Calcium											27.18
Feed Intake	557.0	392.9	429.5	412.7	389.5	406.9	445.0	420.5	413.0	438.2	4305.2
No. of Eggs	2**	3	3	3	2	2	3	3	2	3	26
Wt. of Eggs	102.0	153.2	152.0	156.0	107.1	104.0	154.0	159.8	110.6	158.7	1357.4
Wt. of Shells	10.1	14.2	14.1	13.8	8.8	8.0	13.9	14.5	10.1	13.8	121.3
Wt. of Contents	91.9	139.0	137.9	142.2	98.3	96.0	140.1	145.3	100.5	144.9	1111.0
Body Weight	1680	——	1750	——	1740	——	1820	——	1870	1890	
Av. Shell Thickness	11.1	10.7	10.1	9.8	10.5	10.0	9.9	10.1	10.3	9.9	
Blood Ca	16.716	——	17.200	——	17.776	——	17.052	——	17.136	——	

* Period 1 consisted of four days.

** One egg was laid previous to beginning of experiment.

Table 15. Calcium balance data for Bird No. 78 of Lot No. 2. All weights are in grams.

	Period 1	2	3	4	5	6	7	8	9	10	Total
Ca Intake	4.244	4.193	3.953	4.265	4.202	3.068	4.159	3.525	2.702	2.002	36.313
Ca in Shell	1.741	1.790	1.879	3.913	5.520	1.488	3.404	4.930	1.416	1.371	27.452
Ca in Egg Contents	.019	.020	.020	.045	.072	.028	.041	.066	.019	.019	.349
Ca in Feces	3.342	1.997	1.714	1.463	1.174	.841	1.063	.730	.663	.505	13.502
Ca Outgo	5.102	3.807	3.613	5.421	6.766	2.357	4.508	5.726	2.098	1.895	41.303
Ca Balance	-.858	+.386	+.340	-1.158	-2.564	+.711	-.349	-2.201	+.604	+.107	-4.990
Ca in Skeleton, Final											12.783
Ca in Skeleton, Original											17.773
Percentage Loss of Skeletal Calcium											28.08
Feed Intake	371.5	367.0	346.0	373.3	363.8	268.5	364.0	308.5	236.5	175.2	3174.3
No. of Eggs	1*	1	1	2	3	1	2	3	1	1	16
Wt. of Eggs	46.2	48.9	48.9	104.0	157.3	50.8	101.9	157.2	42.9	46.6	804.7
Wt. of Shells	5.3	5.9	5.6	11.5	16.5	4.5	10.0	14.8	4.7	4.3	83.1
Wt. of Contents	40.9	43.0	43.3	92.5	140.8	46.3	91.9	142.4	38.2	42.3	721.6
Body Weight	1950	———	1950	———	1950	———	1950	———	1950	1875	
Av. Shell Thickness	13.4	13.8	13.5	12.8	12.4	10.6	11.5	11.3	10.5	10.5	
Blood Ca	19.796	———	15.958	———	16.236	———	14.140	———	13.467	13.736	

* One egg laid previous to starting the experiment.

Table 16. Calcium balance data for Bird No. 56 of Lot No. 3. All weights are in grams.

	Period										
	1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	4.889	4.606	5.730	5.620	5.274	5.858	5.265	6.042	7.175	8.105	57.646
Ca in Shell	1.782	1.773	3.600	3.710	3.414	3.701	3.693	3.879	5.948	5.401	36.901
Ca in Egg Contents	.028	.028	.057	.056	.041	.042	.047	.049	.075	.091	.514
Ca in Feces	2.220	2.479	2.350	1.906	1.330	1.457	1.590	1.233	1.264	1.993	17.882
Ca Outgo	4.030	4.280	6.007	5.672	4.785	5.200	5.330	5.161	7.287	7.485	55.297
Ca Balance	+.859	+.326	-.277	-.052	-.489	+.658	-.065	+.881	-.112	+.620	+2.349
Ca Balance from End of Period 1 . . .											+1.490
Ca in Skeleton, Final . . .											13.944
Ca in Skeleton, End of Period 1 . . .											12.454
Percentage Gain in Skeletal Calcium . . .											12.47
Feed Intake	154.0	218.2	271.4	266.2	249.8	277.5	249.4	286.2	296.4	383.9	2652.1
No. of Eggs	1**	1	2	2	2	2	2	2	3	3***	20
Wt. of Eggs	43.0	45.0	95.8	98.3	98.4	97.7	97.4	102.4	156.5	154.7	989.2
Wt. of Shells	5.1	5.3	10.9	11.0	10.3	10.8	10.7	11.5	17.7	15.8	109.1
Wt. of Contents	37.9	39.7	84.9	87.3	88.1	86.9	86.7	90.9	138.8	138.9	880.1
Body Weight	1375	———	1375	———	1425	———	1435	———	1425	1475	
Av. Shell Thickness	14.0	14.1	13.4	13.3	12.4	13.0	12.9	13.1	12.9	11.7	
Blood Ca	20.806	———	22.848	20.402	24.240	———	14.746	———	18.414	14.544	

* Feed consumed during Period 1 contained 3.1747% calcium.

** One egg laid previous to beginning of the experiment.

*** Last egg removed from oviduct.

Table 17. Calcium balance data for Bird No. 59 of Lot No. 3. All weights are in grams.

	Period 1*	2	3	4	5	6	7	8	9	10**	Total
Ca Intake	6.448	4.898	2.160	4.188	6.086	4.283	5.269	4.663	5.554	6.768	50.317
Ca in Shell	2.819	2.833	3.098	1.003	3.362	3.379	3.460	3.159	3.301	4.699	31.113
Ca in Egg Contents	.037	.047	.043	.023	.044	.046	.046	.048	.049	.061	.444
Ca in Feces	3.834	2.548	.936	2.286	2.395	1.372	1.371	.975	1.372	1.976	19.065
Ca Outgo	6.690	5.428	4.077	3.312	5.801	4.797	4.877	4.182	4.722	6.736	50.622
Ca Balance	-.242	-.530	-1.917	+.876	+.285	-.514	+.392	+.481	+.832	+.032	-.305
Ca Balance from end of Period 1 . . .											-.063
Ca in Skeleton, Final . . .											14.988
Ca in Skeleton, End of Period 1 . . .											15.051
Percentage Loss of Skeletal Calcium . . .											.40
Feed Intake	203.1	232.0	102.3	198.4	288.3	202.9	249.6	220.9	263.1	320.6	2281.2
No. of Eggs	2	2	2	1	2	2	2	2	2	3	20
Wt. of Eggs	78.9	86.7	85.6	44.1	96.1	97.9	95.8	95.2	94.9	140.2	915.4
Wt. of Shells	8.9	8.9	9.2	3.4	10.0	10.0	9.7	9.7	9.8	14.3	93.9
Wt. of Contents	70.0	77.8	76.4	40.7	86.1	87.9	86.1	85.5	85.1	125.9	821.5
Body Weight	1475	——	1475	——	1485	——	1460	——	1480	1500	
Av. Shell Thickness	11.9	11.0	11.4	8.4	11.9	11.2	11.3	10.8	11.2	11.1	
Blood Ca	11.110	——	18.612	——	20.806	——	12.261	——	13.134	15.400	

* Feed consumed during Period 1 contained 3.1747% calcium.
** Period 10 consisted of four days.

Table 18. Calcium balance data for Bird No. 62 of Lot No. 3. All weights are in grams.

	Period										
	1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	5.622	7.114	7.619	7.032	5.808	6.369	4.471	4.898	5.421	7.522	61.876
Ca in Shell	1.360	.946	4.822	3.671	3.524	5.358	3.696	3.711	3.795	5.667	36.550
Ca in Egg Contents	.021	.047	.087	.053	.055	.078	.053	.056	.048	.070	.568
Ca in Feces	3.442	4.205	2.853	3.324	2.198	1.356	.956	1.012	1.377	1.795	22.538
Ca Outgo	4.823	5.198	7.762	7.048	5.777	6.792	4.705	4.779	5.220	7.532	59.636
Ca Balance	+.799	+1.916	-.143	-.016	+.031	-.423	-.234	+.119	+.201	-.010	+2.240
Ca Balance from End of Period 1											+1.441
Ca in Skeleton, Final											17.533
Ca in Skeleton, End of Period 1											16.092
Percentage Gain in Skeletal Calcium											8.97
Feed Intake	177.1	337.0	360.9	333.1	275.1	301.7	211.8	232.0	256.8	356.3	2841.8
No. of Eggs	1	1	3	2	2	3	2	2	2	3	21
Wt. of Eggs	38.4	42.2	144.8	106.6	107.8	167.5	110.1	110.6	110.4	163.2	1101.6
Wt. of Shells	4.2	3.0	14.7	11.3	10.9	16.5	11.5	11.2	11.7	17.3	112.3
Wt. of Contents	34.2	39.2	130.1	95.3	96.9	151.0	98.6	99.4	98.7	145.9	989.3
Body Weight	1675	———	1800	———	1780	———	1675	———	1560	1625	
Av. Shell Thickness	12.7	7.7	11.7	12.4	12.1	11.6	11.9	11.7	11.9	12.0	
Blood Ca	———	———	18.564	21.412	19.594	———	14.746	———	12.672	17.979	

* Feed consumed during Period 1 contained 3.1747% calcium.

Table 19. Calcium balance data for Bird No. 76 of Lot No. 3. All weights are in grams.

	Period 1*	2	3	4	5	6	7	8	9	10	Total
Ca Intake	10.197	8.018	6.884	7.051	7.294	5.744	5.886	6.063	6.808	4.689	68.634
Ca in Shell	1.343	1.272	3.108	4.766	3.677	3.301	3.274	3.322	5.125	3.293	32.481
Ca in Egg Contents	.018	.020	.043	.053	.066	.044	.046	.045	.065	.042	.442
Ca in Feces	9.162	3.757	4.319	2.784	2.597	2.611	1.937	1.756	2.116	1.667	32.706
Ca Outgo	10.523	5.049	7.470	7.603	6.340	5.956	5.257	5.123	7.306	5.002	65.629
Ca Balance	-.326	+2.969	-.586	-.552	+.954	-.212	+.629	+.940	-.498	-.313	+3.005
Ca Balance from End of Period 1											+3.331
Ca in Skeleton, Final											15.215
Ca in Skeleton, End of Period 1											11.884
Percentage Gain in Skeletal Calcium											28.03
Feed Intake	321.2	379.8	326.1	334.0	345.5	272.1	278.8	287.2	322.5	222.1	3089.3
No. of Eggs	1	1	2	3	2	2	2	2	3	2	20
Wt. of Eggs	42.3	43.7	96.3	149.0	122.3	100.5	99.8	101.8	152.5	101.0	1009.2
Wt. of Shells	4.1	4.0	9.2	14.2	11.1	9.8	9.6	9.8	14.3	9.9	96.0
Wt. of Contents	38.2	39.7	87.1	134.8	111.2	90.7	90.2	92.0	138.2	91.1	913.2
Body Weight	1720	——	1790	——	1750	——	1730	——	1800	1700	
Av. Shell Thickness	11.7	10.4	11.6	11.3	11.5	11.4	10.9	11.7	11.1	11.2	
Blood Ca	18.584	22.624	——	16.766	——	20.394	——	15.756	——	14.000	

* Feed consumed during Period 1 contained 3.1747% calcium.

Table 20. Calcium balance data for Bird No. 58(1) of Lot No. 4. All weights are in grams.

	Period 1	2	3	4	5	6	7	8*	9	10	Total
Ca Intake	7.702	6.899	7.140	7.556	6.934	5.381	2.848	2.299			46.759
Ca in Shell	3.400	3.531	3.567	5.328	3.587	1.749	———	———			21.162
Ca in Egg Contents	.063	.076	.053	.083	.052	.024	———	———			.351
Ca in Feces	4.374	3.031	3.893	4.199	3.299	3.032	4.942	3.175			29.443
Ca Outgo	7.837	6.638	7.513	9.610	6.938	4.805	4.942	3.175			51.458
Ca Balance	-.135	+.261	-.373	-2.054	-.004	+.576	-2.094	-.876			-4.699
Ca in Skeleton, Final . . .											14.648
Ca in Skeleton, Initial . . .											19.347
Percentage Loss of Skeleton Calcium . . .											24.29
Feed Intake	242.6	217.3	224.9	238.0	218.4	169.5	89.7	72.4			1472.8
No. of Eggs	3	2	2	3	2	1	———	———			13
Wt. of Eggs	136.0	93.2	96.3	145.0	95.7	47.9	———	———			614.1
Wt. of Shells	14.7	10.1	10.1	15.7	10.4	5.2	———	———			66.2
Wt. of Contents	121.3	83.1	86.2	129.3	85.3	42.7	———	———			547.9
Body Weight	1325	———	1200	———	1275	———	1170	1225			
Av. Shell Thickness	13.4	13.6	12.6	12.7	12.9	12.5	———	———			
Blood Ca	21.816	———	24.684	21.008	20.806	———	12.928	———			

* Killed at end of Period 8.

Table 21. Calcium balance data for Bird No. 58(2) of Lot No. 4. All weights are in grams.

	Period										
	1	2	3	4	5	6	7	8	9	10	Total
Ca Intake	6.861	9.000	9.524	8.540	8.969	9.108	8.086	7.664	8.886	8.972	85.610
Ca in Shell	1.662	1.888	5.941	4.039	2.122	4.309	4.313	4.055	———	———	28.329
Ca in Egg Contents	.022	.025	.081	.054	.027	.056	.052	.049	———	———	.366
Ca in Feces	3.919	4.064	5.489	4.245	4.884	5.069	3.785	3.687	6.641	9.044	50.827
Ca Outgo	5.603	5.977	11.511	8.338	7.033	9.434	8.150	7.791	6.641	9.044	79.522
Ca Balance	+1.258	+3.023	-1.987	+.202	+1.936	-.326	-.064	-.127	+2.245	-.072	+6.088
Ca in Skeleton, Final . . .											18.192
Ca in Skeleton, Initial . . .											12.104
Percentage Gain in Skeletal Calcium . . .											50.3
Feed Intake	216.1	283.5	300.0	269.0	282.5	286.9	254.7	241.4	279.9	282.6	2696.6
No. of Eggs	1	1	3	2	1	2	2	2	0	0	14
Wt. of Eggs	45.3	51.8	160.8	108.1	54.1	111.9	110.0	107.5	———	———	749.5
Wt. of Shells	5.2	5.6	17.5	11.9	6.3	13.1	12.5	10.5	———	———	82.6
Wt. of Contents	40.1	46.2	143.3	96.2	47.8	98.8	97.5	97.0	———	———	666.9
Body Weight	1800	———	1900	———	1840	———	1825	———	1870	1850	
Av. Shell Thickness	12.4	12.1	12.5	13.0	13.7	13.8	13.7	13.1	———	———	
Blood Ca	14.271	———	16.915	———	13.800	———	16.564	———	9.541	———	

Table 22. Calcium balance data for Bird No. 72 of Lot No. 4. All weights are in grams.

	Period										Total
	1	2	3	4	5	6	7	8	9	10	
Ca Intake	3.372	6.067	5.461	4.899	5.105	6.861	5.734	7.803	6.588	10.680	62.570
Ca in Shell	1.493	1.909	1.676	3.216	1.780	3.622	1.923	3.861	1.934	5.795	27.209
Ca in Egg Contents	.020	.032	.018	.033	.018	.045	.022	.048	.026	.084	.346
Ca in Feces	1.594	3.321	3.011	3.125	2.169	3.106	3.624	3.904	3.648	4.771	32.273
Ca Outgo	3.107	5.262	4.705	6.374	3.967	6.773	5.569	7.813	5.608	10.650	59.828
Ca Balance	+.265	+.805	+.756	-1.475	+1.138	+.088	+.165	-.010	+.980	+.030	+2.742
Ca in Skeleton, Final . . .											18.983
Ca in Skeleton, Initial . . .											16.241
Percentage Gain in Skeletal Calcium . . .											16.89
Feed Intake	106.2	191.1	172.0	154.3	160.8	216.1	180.6	245.8	207.5	336.4	1970.8
No. of Eggs	1	1	1	2	1	2	1	2	1	3	15
Wt. of Eggs	37.8	53.9	39.8	81.5	43.3	92.9	48.2	98.8	48.7	151.4	696.3
Wt. of Shells	4.5	6.5	4.8	9.5	5.2	10.7	5.7	11.4	5.7	16.8	80.8
Wt. of Contents	33.3	47.4	35.0	72.0	38.1	82.2	42.5	87.4	43.0	134.6	615.5
Body Weight	1525	——	1500	——	1525	——	1450	——	1450	1525	
Av. Shell Thickness	13.1	13.7	13.8	13.3	14.1	13.5	13.6	13.4	13.6	12.7	
Blood Ca	——	16.932	——	20.200	19.796	——	15.352	——	19.602	18.988	

Table 23. Calcium balance data for Bird No. 75 of Lot No. 4. All weights are in grams.

	Period 1	2	3	4	5	6	7	8	9	10*	Total
Ca Intake	4.111	6.727	7.803	9.077	8.207	8.023	8.359	8.092	8.594	10.607	79.600
Ca in Shell	3.569	2.024	3.988	4.279	4.277	4.210	4.402	4.161	4.477	6.399	41.786
Ca in Egg Contents	.047	.023	.049	.051	.054	.051	.056	.051	.058	.080	.520
Ca in Feces	2.157	3.698	4.441	4.424	3.228	2.661	3.392	3.018	3.012	3.922	33.953
Ca Outgo	5.773	5.745	8.478	8.754	7.559	6.922	7.850	7.230	7.547	10.401	76.258
Ca Balance	-1.662	+.982	-.675	+.323	+.648	+1.101	+.509	+.862	+1.047	+.206	+3.341
Ca in Skeleton, Final . . .											20.202
Ca in Skeleton, Initial . . .											16.861
Percentage Gain in Skeletal Calcium . . .											19.80
Feed Intake	129.5	211.9	245.8	285.9	258.5	252.7	263.3	254.9	270.7	334.1	2507.3
No. of Eggs	2	1	2	2	2	2	2	2	2	3	20
Wt. of Eggs	95.3	50.7	101.9	108.6	109.5	111.9	112.6	103.8	113.3	161.5	1069.1
Wt. of Shells	10.5	5.8	11.8	12.2	12.2	12.4	12.6	12.3	13.3	19.0	122.1
Wt. of Contents	84.8	44.9	90.1	96.4	97.3	99.5	100.0	91.5	100.0	142.5	947.0
Body Weight	1740	——	1650	——	1640	——	1610	——	1635	1625	
Av. Shell Thickness	12.8	13.7	13.3	13.0	13.6	13.2	13.8	14.3	14.1	13.9	
Blood Ca	14.652	——	17.372	——	20.703	——	15.124	——	15.000	15.352	

* Period 10 consisted of four days.

Table 24. Calcium balance data for Bird No. 79 of Lot No. 4. All weights are in grams.

	Period 1	2	3	4	5	6	7	8	9	10	Total
Ca Intake	6.892	9.863	8.257	7.365	10.159	10.686	8.153	8.242	8.911	6.807	85.355
Ca in Shells	4.353	4.250	3.990	———	2.140	———	2.128	4.159	1.849	3.844	26.713
Ca in Egg Contents	.054	.053	.049	———	.025	———	.024	.048	.022	.045	.320
Ca in Feces	3.233	5.636	4.036	4.708	6.877	8.447	5.405	4.988	4.714	4.058	52.102
Ca Outgo	7.640	9.939	8.075	4.708	9.042	8.447	7.557	9.195	6.585	7.947	79.135
Ca Balance	-.748	-.056	+.182	+2.657	+1.117	+2.239	+.596	-.953	+2.326	-1.140	+6.220
Ca in Skeleton, Final . . .											20.124
Ca in Skeleton, Initial . . .											13.904
Percentage Gain in Skeletal Calcium . . .											+44.09
Feed Intake	217.1	311.3	260.1	232.0	320.0	336.6	256.8	259.6	280.7	214.4	2688.6
No. of Eggs	2	2	2	———	1	———	1	2	1	2	13
Wt. of Eggs	104.7	108.3	104.4	———	50.8	———	51.9	103.9	48.3	99.4	671.7
Wt. of Shells	12.9	13.3	11.9	———	5.5	———	6.2	12.3	5.6	11.7	79.4
Wt. of Contents	91.8	95.0	92.5	———	45.3	———	45.7	91.6	42.7	87.7	592.3
Body Weight	1790	———	1780	———	1820	———	1870	———	1810	1800	
Av. Shell Thickness	15.4	14.5	13.8	———	14.9	———	14.9	15.1	13.9	15.1	
Blood Ca	———	15.400	———	12.726	———	18.473	———	13.250	———	10.050	

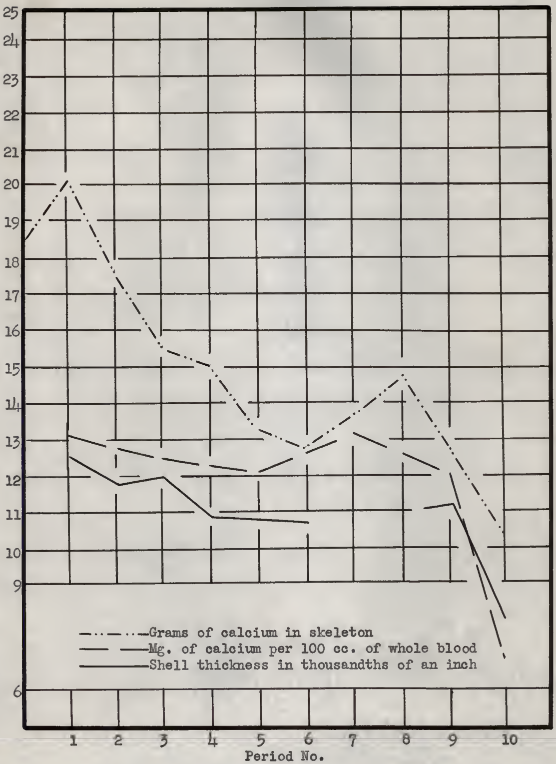

Fig. 5. Experimental data for Bird No. 57, Lot 1.

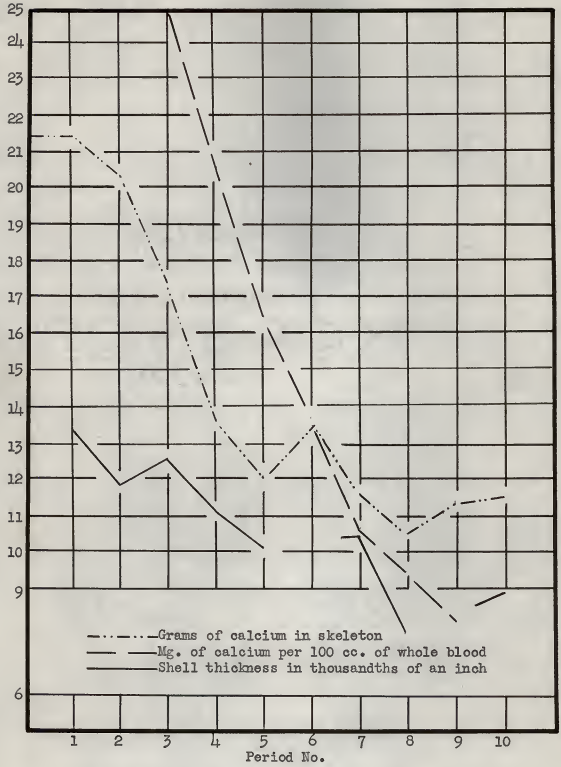

Fig. 6. Experimental data for Bird No. 71, Lot 1.

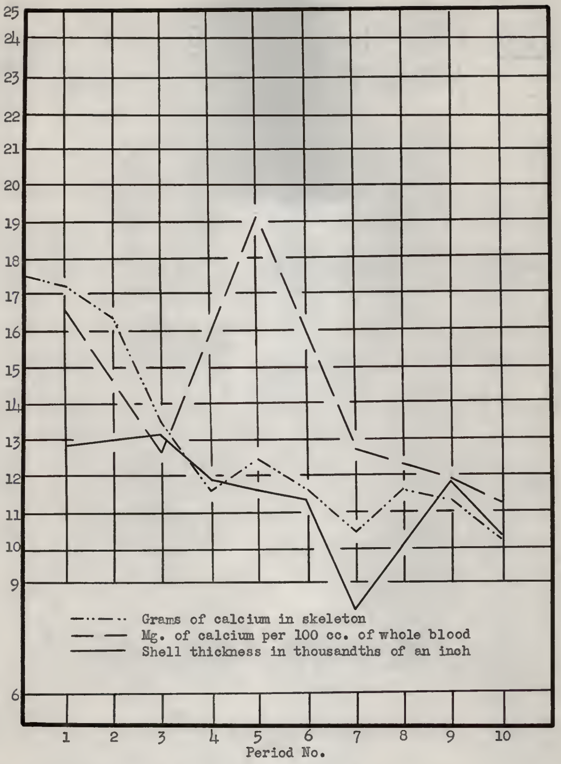

Fig. 7. Experimental data for Bird No. 73, Lot 1.

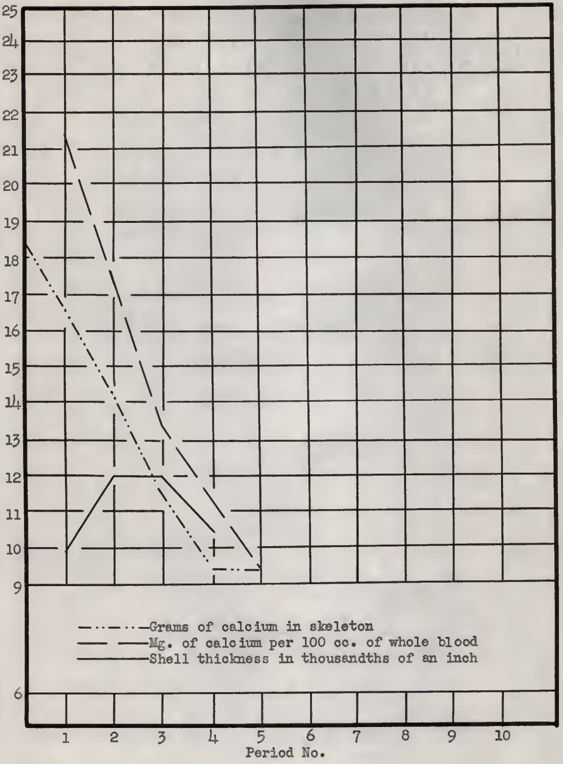

Fig. 8. Experimental data for Bird No. 77(1), Lot 1.

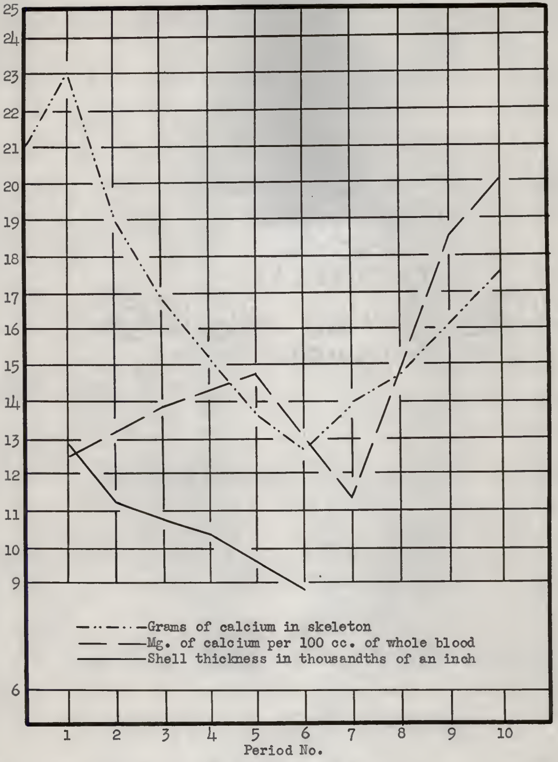

Fig. 9. Experimental data for Bird No. 77(2), Lot 1.

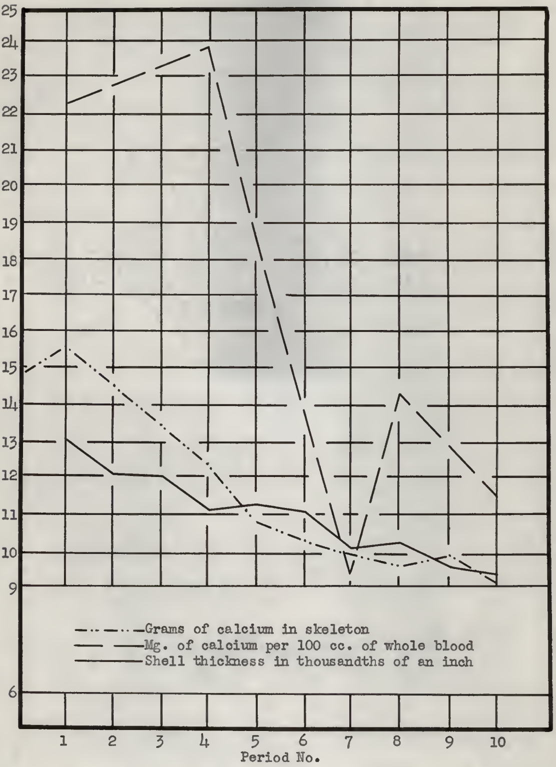

Fig. 10. Experimental data for Bird No. 55, Lot 2.

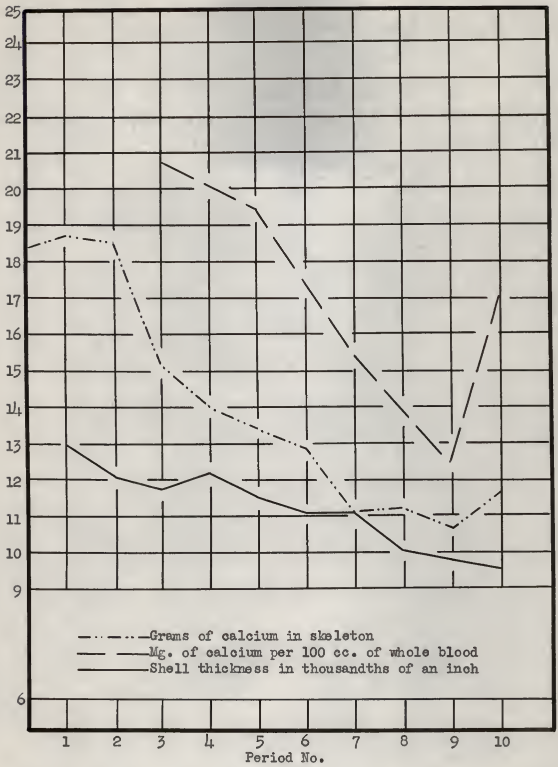

Fig. 11. Experimental data for Bird No. 60, Lot 2.

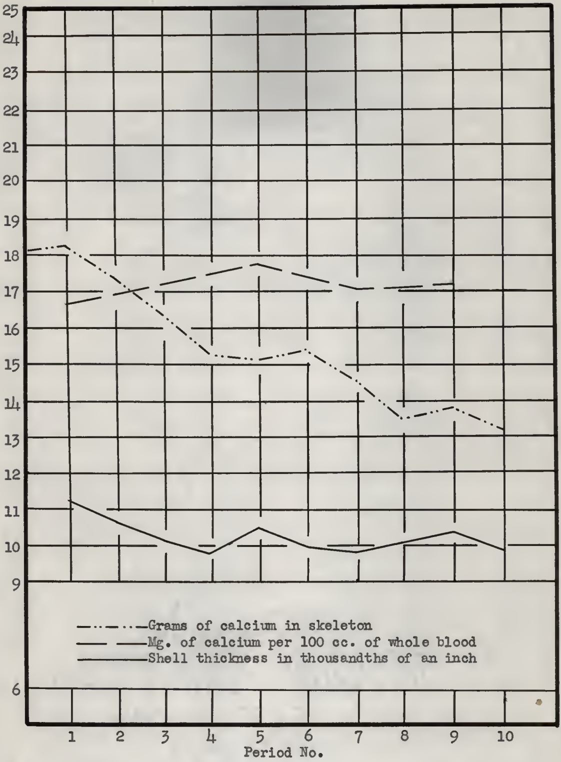

Fig. 12. Experimental data for Bird No. 61, Lot 2.

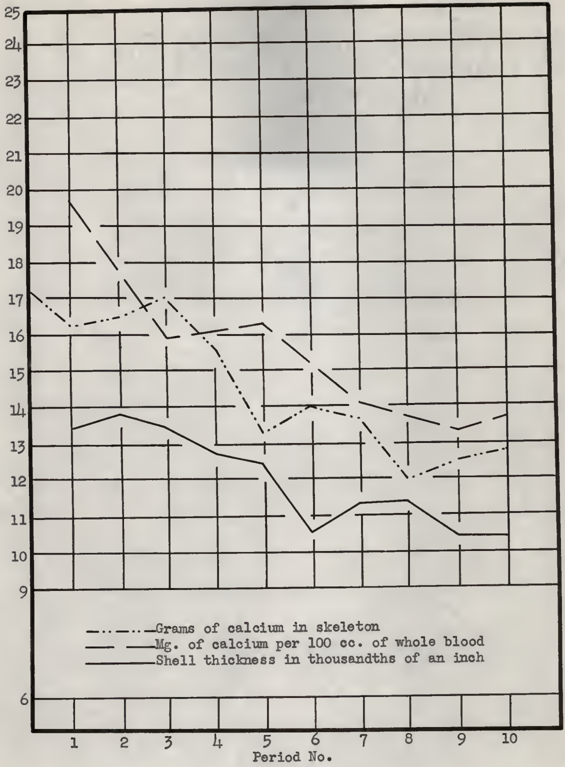

Fig. 13. Experimental data for Bird No. 78, Lot 2.

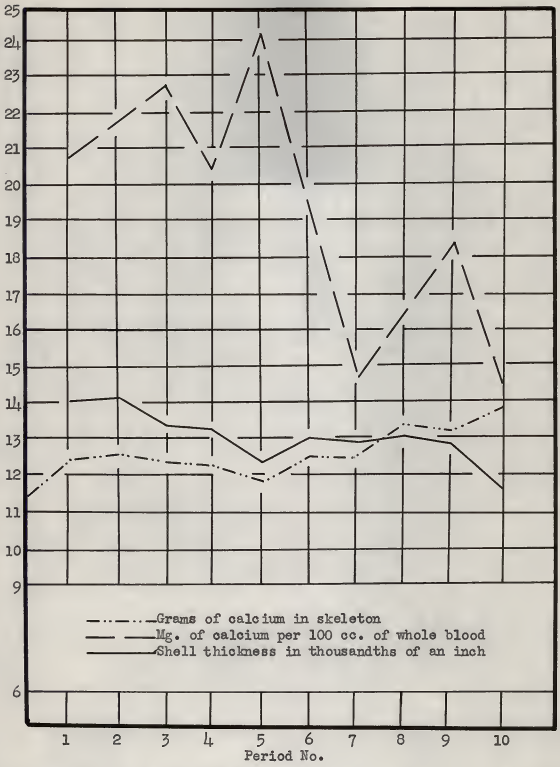

Fig. 14. Experimental data for Bird No. 56, Lot 3.

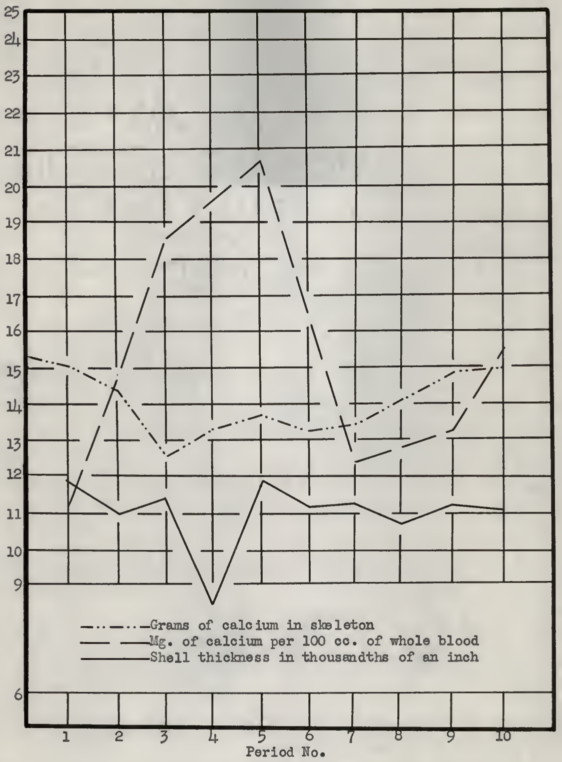

Fig. 15. Experimental data for Bird No. 59, Lot 3.

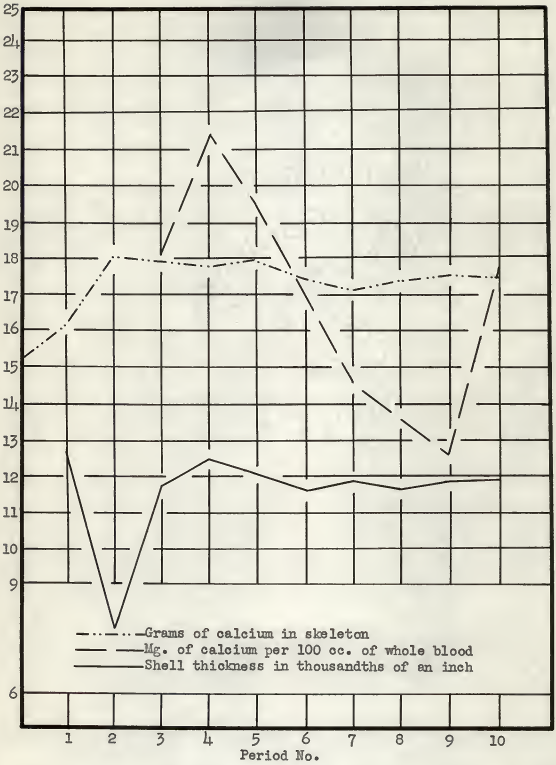

Fig. 16. Experimental data for Bird No. 62, Lot 3.

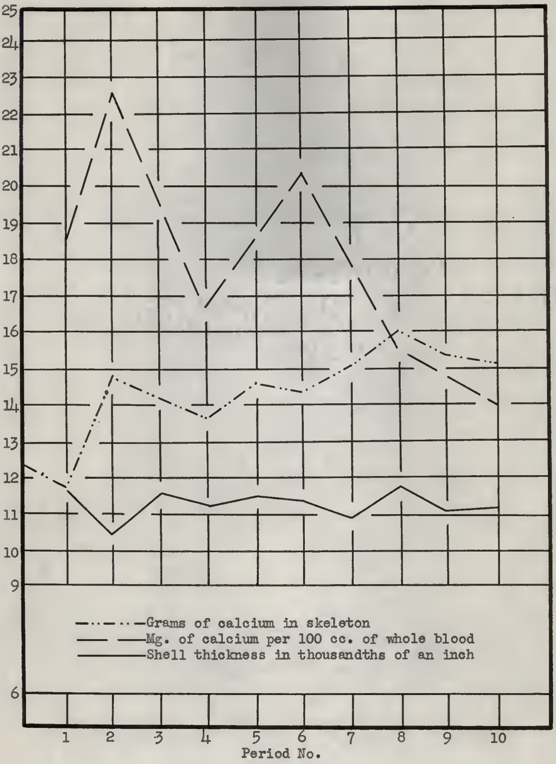

Fig. 17. Experimental data for Bird No. 76, Lot 3.

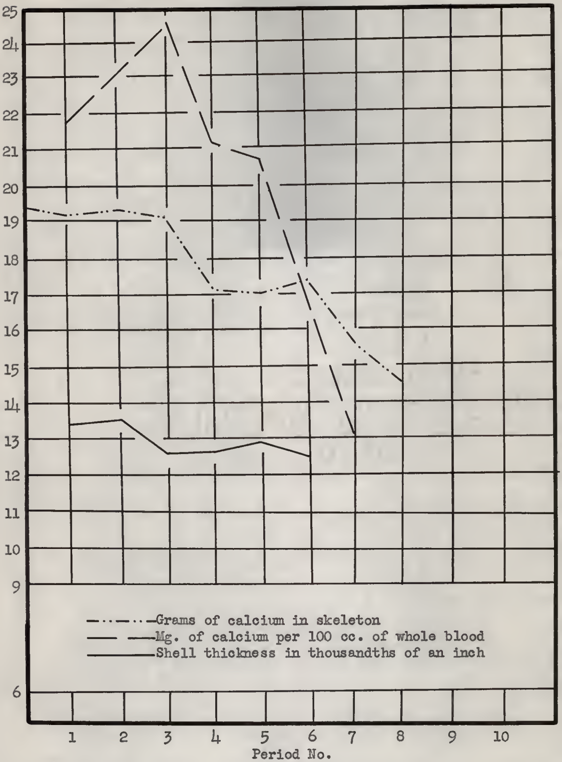

Fig. 18. Experimental data for Bird No. 58(1), Lot 4.

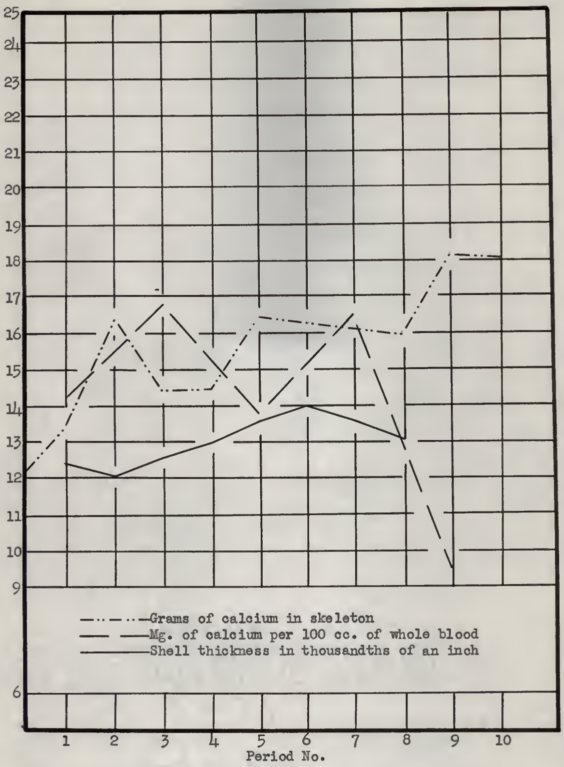

Fig. 19. Experimental data for Bird No. 58(2), Lot 4.

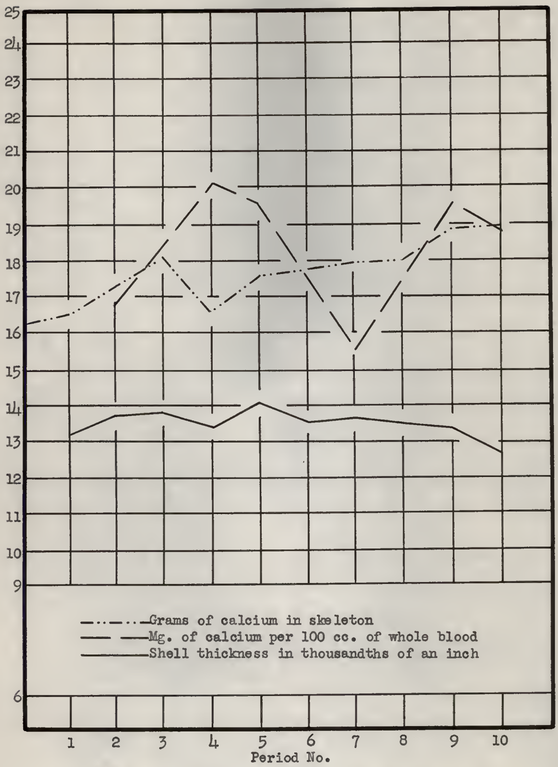

Fig. 20. Experimental data for Bird No. 72, Lot 4.

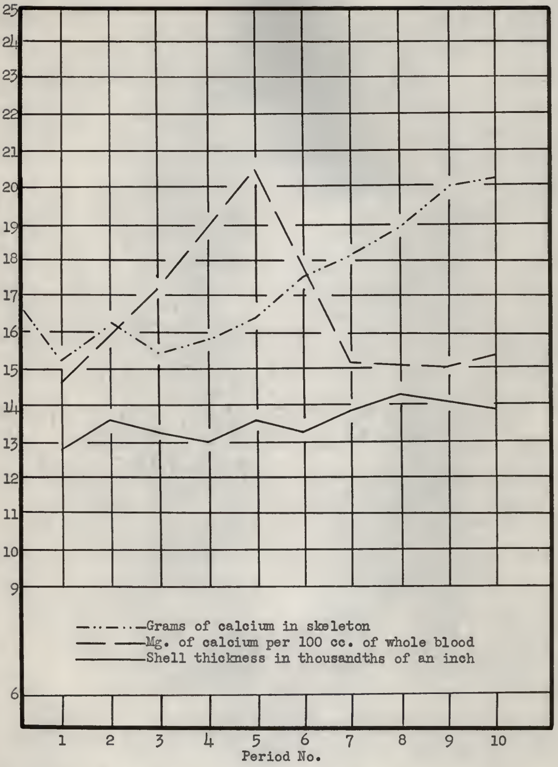

Fig. 21. Experimental data for Bird No. 75, Lot 4.

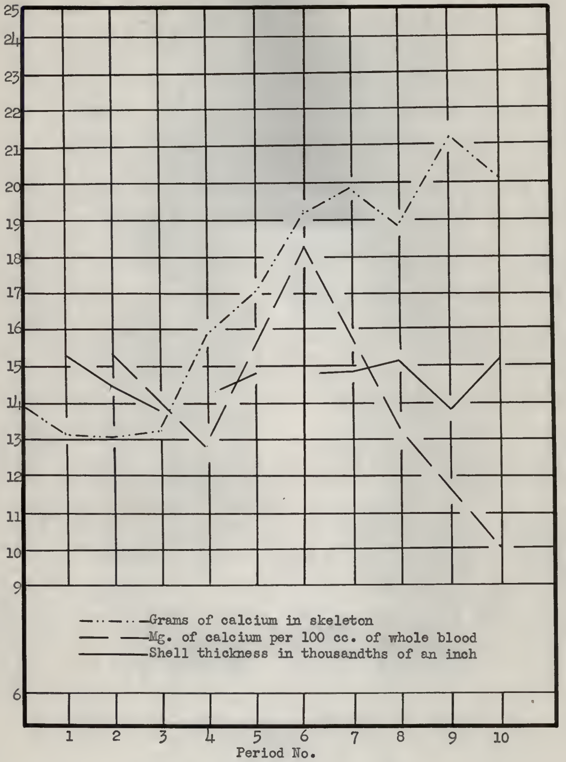

Fig. 22. Experimental data for Bird No. 79, Lot 4.