STUDIES ON THE EFFECT OF THYMECTOMY ON GROWING CHICKENS

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B.S., Kansas State Agricultural College, 1924

A THESIS

submitted in partial fulfillment of the

requirements for the degree of

MASTER OF SCIENCE

KANSAS STATE AGRICULTURAL COLLEGE

1929

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INTRODUCTION

The finding of Ackert (1924) that intestinal parasites decrease the size of the thymus glands of chickens and the announcement of Riddle (1924) that the thymi of pigeons are responsible for normal egg envelopes, raised the question of whether the thymus glands of chickens affect the formation of their egg shells.

As a preliminary test, Ackert (unpublished) removed the thymi from a mature Rhode Island Red hen, which had been laying about two months. Thirty days later she began again to lay. The first three eggs appeared to be normal; but the two which followed had such thin shells that the eggs dented on being handled. While the shells of the eggs layed subsequently became stronger, the results indicated that the thymus glands were a factor in the formation of normal egg shells.

One year later, two white leghorn pullets, raised in confinement, were used to obtain more data on the possible relation between the thymus glands and the formation of normal egg shells. After both of these pullets had layed many eggs they were thymectomized. The first pullet number 1349 died following the operation. The second number 1340, survived. Before thymectomy her 11 eggs were

normal. From the measurements made on the eggs layed before and after thymectomy there was no indication that the thymus glands control the formation of the egg envelope. Similar data were obtained on three hens (1373, 1374, 1379) four months later, the results failing to show a significant difference between the eggs layed prior and subsequent to thymectomy.

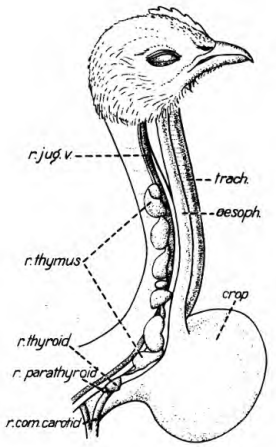
As the above tests were on birds which had matured before thymectomy the writer decided, at the suggestion of Doctor J.E. Ackert, to ascertain what the effect would be on chickens thymectomized at a week or so of age, the birds thus being without their thymi during the growing period.

THE THYMUS GLANDS

The thymus glands have a multiple origin, coming from the dorsal region of the gill pouches. They are present in all vertebrates. In some fishes the glands remain epithelial in character, while in higher forms they become lymphoid and highly vascular. The different embryonic elements of the thymus frequently fuse to form masses of tissue down either side of the neck. In birds, however, they remain as individual lobes, usually seven pairs on either side of the neck extending from the region of the larynx to the lower part of the crop.

By slitting the skin the glands can be readily exposed.

The complications experienced in the thymectomy of mammals are not encountered in birds, as there are no bones, muscles or pleura with which to contend. The greatest difficulty to be met is in the close proximity of the lobes to the jugular vein and vagus nerve. Beyond the last two thymus lobes lie the thyroids and and parathyroids. The thyroid appears more red than the thymus tissue and is smaller than the adjacent thymus lobe. The parathyroids are very small and posterior to the thyroids.



REVIEW OF PREVIOUS WORK ON AVIAN THYMECTOMY

Numerous attempts to study the function of the thymus glands have been made with many different types of animals, including a number of cases of extirpation of the thymi of birds. Tarulla and Lo Monaco (1897) performed total thymectomy on a group of very young chickens with heavy losses within a few days. The details of the symptoms and deaths indicate that the chickens also suffered with leg weakness (deficiency of vitamin D). Fischl (1907) being attracted by the works of Tarulla investigated the effect of thymus removal in fowl. He removed the thymi from 24 chicks from one to 12 days old, with a loss of only two chicks; the others recovered quickly and without ill effects.

Fischl concluded that the removal of the thymi was without significance, but postulated for it a function in fetal life which ceased at birth.

Basch (1908) vigorously assailed the work of Fischl as he had found the extirpation of the thymus in dogs had created a marked change in the skeleton, and a doubled calcium excretion. Basch concluded that it was impossible to remove the glands in birds, since he found thymus rests in the normal positions of the glands, three months after removal, and also that the tissue had the power of regeneration.

Soli (1906) states that the removal of the thymi from young cocks delayed the development of the testes. This author (1910) extended his investigations to pullets, to determine the effect of thymectomy upon the deposition of calcium in the egg shell. He concluded that the calcium metabolism was modified in the hen by extirpation of the thymi, ascribing the failure in the calcification of the egg to diminished absorption of the calcium from the intestinal tract and a diminished utilization of the calcium circulating in the blood by the various tissues of the body.

Katsura (1922) performing thymectomy in chickens concluded that the thymus has one function which helps the growth of animals and another that retards the maturity.

He operated pullets when four months old to test the effect of thymectomy on the shell structure. Katsura found no significant microscopical difference and only one per cent of calcium less than the controls.

Riddle (1924) after total thymectomy in normal pigeons found certain marked changes in the eggs, with deficiency in the albumen and in the shell, with single rather than paired ovulations, diminished fertility and restricted hatchability. Autopsy showed small thymi. Riddle designated a hormone highly specific in action upon the oviduct of birds, for which he suggested the name "thymovidin". Riddle and Frey (1925) stated that a true age involution occurs in pigeons; that there is a decrease in both sexes at about the time of sexual maturity and a further decrease with old age. The thymus tissue remained in excess of the thyroid tissue.

By feeding thymus to normal birds Krizenecky and Nevalonnyj (1927) secured an increase in pigmentation or intensity of color in chickens which they attributed to hyperthymism. By feeding thyroid and thymus these authors concluded that there is an antagonism between the thymus and thyroid glands, hyperthyroidism tending toward depigmentation and hyperthymism stimulating pigmentation.

The lack of uniformity in the results obtained from

thymectomy and the absence of definiteness in the function of the thymus make it desirable to search further for a possible function of the thymus.

METHODS

The chickens used in this experiment were pure bred single comb white leghorns, secured from a commercial hatchery as day old chicks, and raised in confinement on a natural adequate diet. That chickens so reared are normal was shown by Herrick, Ackert and Danheim (1923). The ration consisted of:

Yellow corn	40 parts.
Cracked wheat	12 parts.
Otona	12 parts.
Powdered skim milk	6.4 parts.
Alfalfa leaf meal	4 parts.
Cod-liver oil	2 parts.
Bone meal	1.6 parts.
Powdered oyster shell	1.6 parts.
Salt (NaCl)	.8 parts.

At all times the chickens had access to the food and water.

When they were one to three weeks old the chicks were anaesthetized with ether and operated for the removal of the thymi. Several of the first operations involved in-

cisions on each side of the neck. This method was almost doubly hard on the chick, especially the very young whose necks were small. A second method used was a median, dorsal incision. This worked less hardship on the chicken, but caused the last lobes to be obscured and easily overlooked. Later exploratory operations revealed the mistake and made reoperation necessary. It was found best to make a median ventral incision, exposing the trachea and the crop, as two pairs of lobes may be hidden by the latter.

The task of removing the lobes was very tedious and required careful technique, each lobe being enveloped in fascia, which had to be separated from the gland, and very carefully from the jugular. More or less hemorrhage was caused when each lobe was separated from its blood vessels. Occasionally there was a considerable amount of hemorrhage, but at other times it was small and the operation might be termed bloodless. The fascia was separated by blunt dissection, a small curved scalpel and toothed forceps being used.

Another problem was presented by the fat which surrounds the glands. In artificial light it closely resembles them. However, the glands can be readily recognized after a little experience, by their peculiar pinkish color and smooth texture. To make sure the thymectomy was complete, much of the fat was also removed.

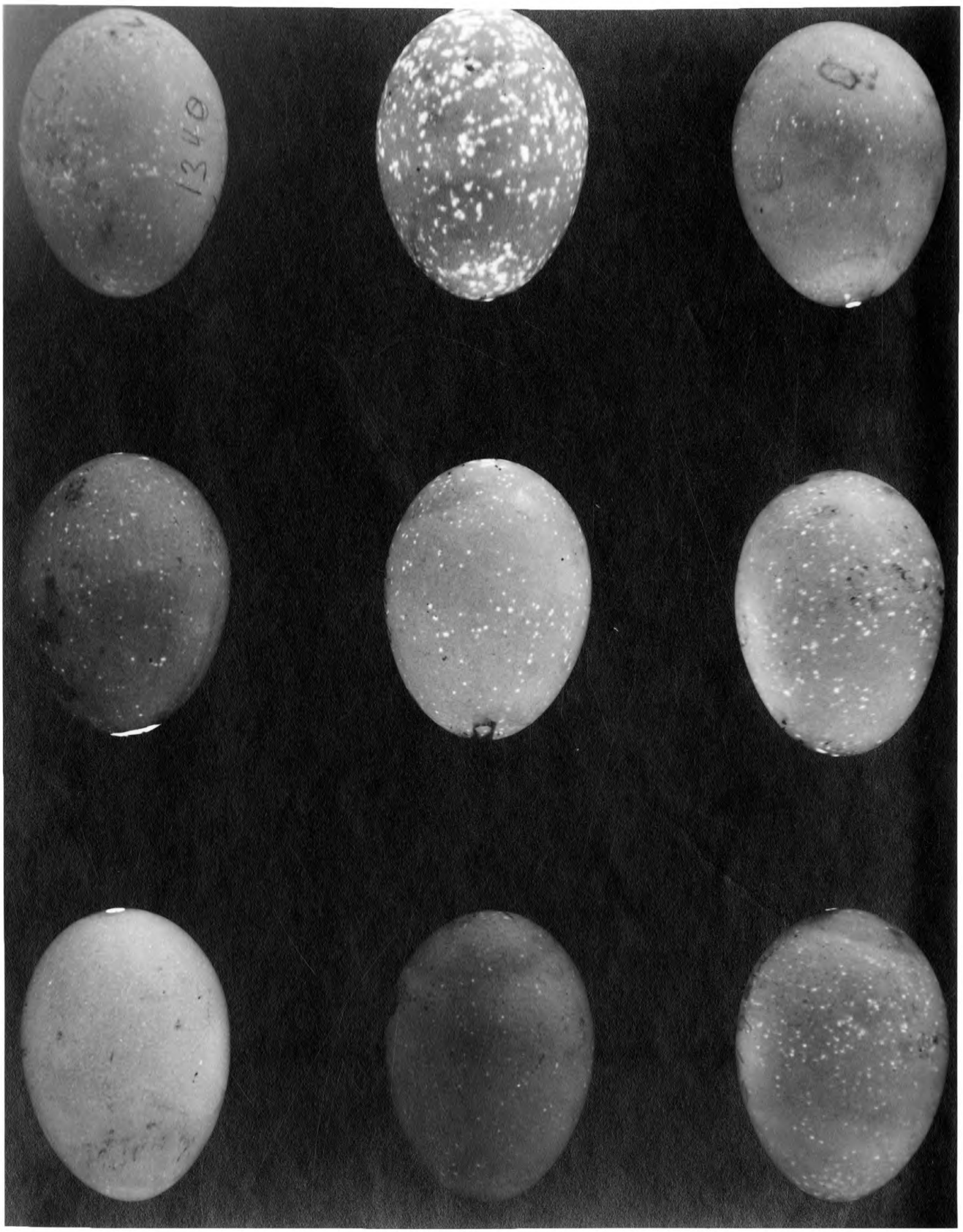
After the glands were taken out, the edges of the skin were brought together and sutured with black thread. As a final step, the wound was covered with pure vaseline, which aided in keeping out the air and dust, and in keeping the skin pliable. The incisions healed with remarkable rapidity; and in no case was there a death from infection. The operated bird was placed in a clean cage with food and water, for within a half-hour it would be up and recovered from the effects of the ether. When returned to the pen the following day, the chick would be as active as the others and within a few days could not be detected in the group.

EXPERIMENT I

The hens used in this experiment were secured as day-old chicks and raised in confinement. When the pullets began to lay they were trap-nested. Each egg was subjected to certain tests including candling for general texture of the shell. If the opacity of shell was quite uniform the appearance of the shell was said to be normal. If there were numerous semitransparent spots, it was described as pitted (Fig. 1).

Four of the hens(1340, 1373, 1374 and 1379) were first observed as normal hens, and data were taken on the eggs,

Fig. 1 Showing texture of egg shells.


1340

thus making each hen serve as a control or check on herself. After thymectomy similar data were taken for comparison (Table I). Before thymectomy the 5 eggs of hen 1340 were normal, their average weight was 47.8 gm., the shell thickness was 0.424 mm., and the percentage of calcium carbonate in the shells was 36.6 per cent. The 84 eggs which followed thymectomy were mostly pitted, had an average weight of 48.2 gm., a shell thickness of 0.3922 mm., a shell strength of 4049 gm., and the percentage of calcium carbonate in the shells was 37.05 in the first two weeks period following the operation, and 36.25 in the time following. The other three of the group were treated in the same manner and gave quite similar data (Table I). Hen 1380 furnished interesting and valuable data in that she was a control with a sham operation which left the glands intact. In the period prior to the operation this hen had an average shell strength of 3588 gm., while in the period following she had an average shell strength of 3218 gm. which is 370 gm. less. The calcium carbonate test prior to the sham operation was 36.89 per cent, as compared with 36.63 per cent in the two weeks following, again averaging 36.89 per cent for the rest of the period. This indicates a slight fall in the period following the sham operation. Hen 1372 was

Table I Giving data on eggs layed prior and subsequent to thymectomy of each hen.

Before Thymectomy

Hen No.	No. of eggs	Av. wt. gm.	Appearance of egg shell	Av. shell strength gm.	Av. shell thickness mm.	Average $CaCO_3$ per cent
1340*	5	47.8	Normal		.424	36.59
1374*	15	47.9	Slightly pitted	4638	.357	36.89
1380†	11	48.1	Pitted	3588	.341	36.89
1372++	15	44.6	Nearly normal	4153		35.65
1373=	19	47.1	Nearly normal	2791	.31	36.46
1379**	13	45.8	Nearly normal	4596	.34	35.65

After Thymectomy

						First 2 Weeks thymectomy	After 2 week period
1340*	84	48.2	Mostly pitted	4049	.3922	37.05	36.25
1374*	12	55.3	Pitted	4374	.364	(No eggs)	36.97
1380†	21	51.2		3218		36.63	36.89
1372++	45	48.9	Slightly pitted	5560			
1373=	31	49.6	Pitted	2776	.31	35.95	36.33
1379**	52	47.9	Pitted	3647	.33	36.21	36.72

*No regeneration of thymi.
†Sham operation - no lobes removed. Two soft shelled eggs layed after the operation.
++Cohtrol
=One small lobe - exploratory operation.
**Regenerated one small lobe of abnormal shape.

used as a control throughout this period, and showed an increase in both egg weight and shell strength. While the results from this experiment indicate no outstanding influences of thymectomy, they warrant further study of the problem.

EXPERIMENT II

The chicks of this group which hatched June 26, 1928 were thymectomized in the period between July 19 and August 4, the operations being completed by the time they were 39 days old. Exploratory operations in search of regenerated lobes were made on October 26, 27, 28, 1928.

An autopsy on cockerel 3218 revealed the fact that the last pair of lobes, which are situated at the base of the crop had been overlooked. These lobes of the thymus were removed during the latter half of November, 1928, so that all of the chickens were completely thymectomized by the time they were five months old, and before egg laying began.

Weekly weights of the birds in this experiment were made from the time the chickens were one and one-half months to six months of age. As single comb, white leghorns mature at approximately six months, weighing was discontinued at that time. Comparing the thymectomized

males with the control males it was found that at six weeks of age, the thymectomized males averaged 234 gm. (Fig. 2), and the controls 237 gm. Week by week there were slight fluctuations, the thymectomized running a few grams heavier until during the fifth month, when there was slight advantage in favor of the controls. This was doubtless due to the effects of reoperating on the thymectomized group in search of possible regenerated thymi. This advantage of the controls, however, was overcome during the sixth month, the average weight of the controls then being 1669 gm., and that of the thymectomized 1714 gm. This difference of 45 gm. is not significant.

Comparing the female birds in this group it was found that at the age of one and one-half months the thymectomized females averaged 230 gm. in weight and the control females 232 gm. The controls continued to outweigh the thymectomized pullets by a few grams each week until the close of the sixth month, when the thymectomized pullets averaged 1364 gm., and the control pullets 1335 gm. At this time the thymectomized pullets were reoperated in search of regenerated thymi, the effects showing up in the weights taken at the close of the week, when it was found that the controls were again slightly outweighing the thymectomized group. Also at the close of this week all birds were bled for a blood sugar test, 5 cc. of blood

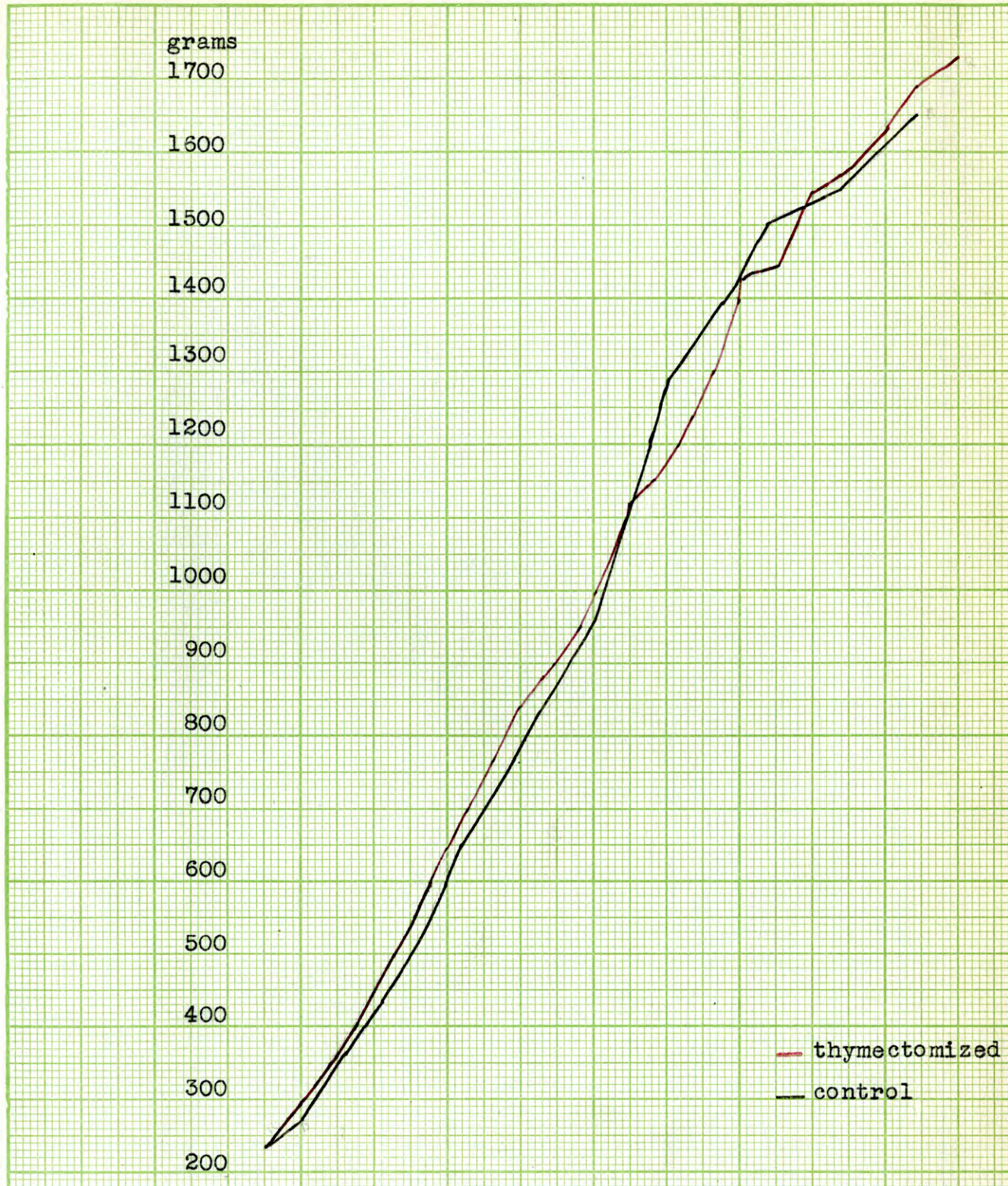


Fig. 2 Showing comparative weights of the thymectomized and control cockerels.

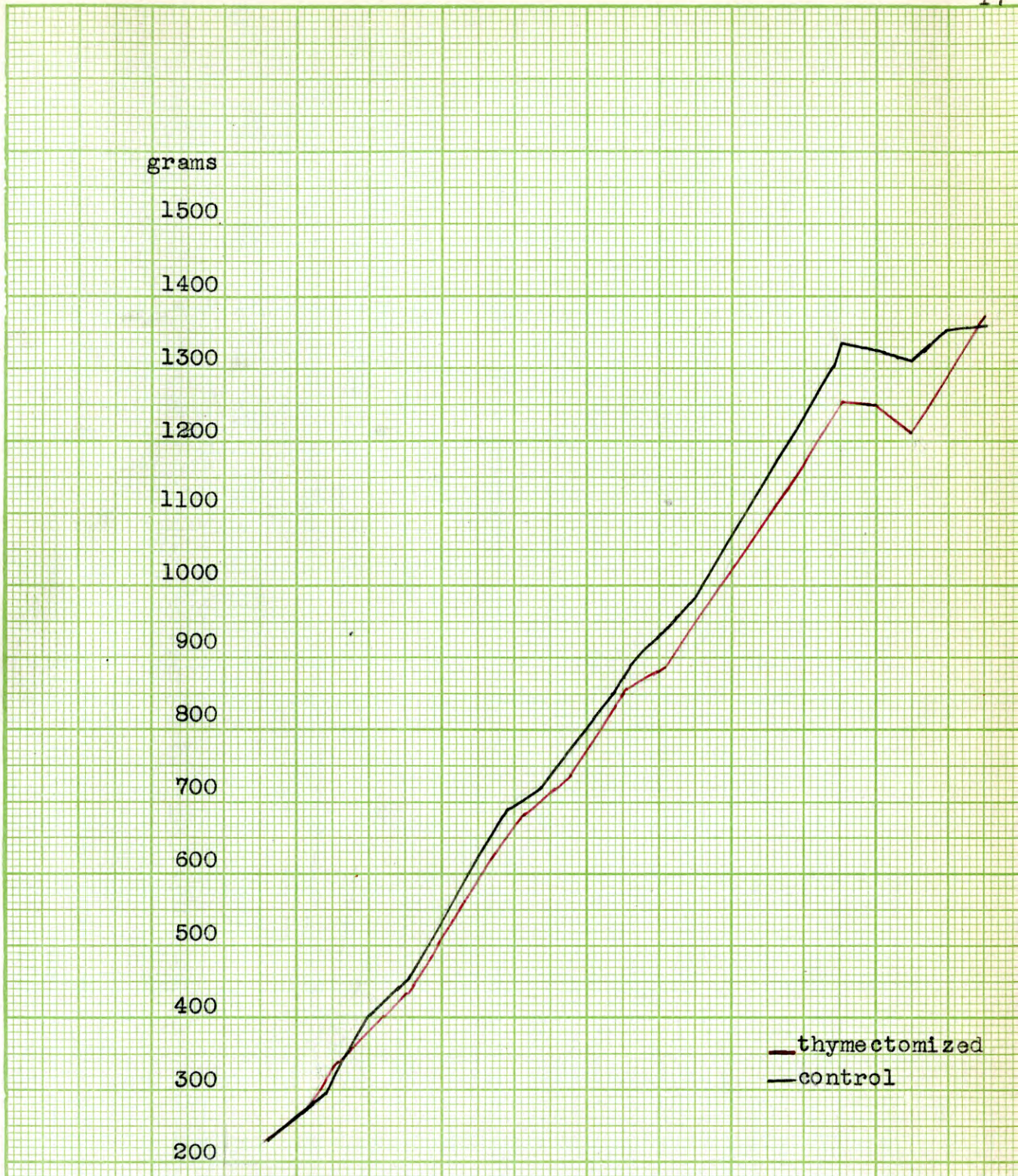


Fig. 3 Showing comparative weights of the thymectomized and control pullets.

having been withdrawn from each bird by cardiac puncture. The combination of reoperating and bleeding brought down the weights of the thymectomized group to an average of 1207 gm. as compared with an average weight of 1311 gm. for the control group, which of course was not reoperated (Fig. 3). The weekly weights were continued until the birds were seven months of age, at which time the thymectomized females averaged 1337 gm. and the control females 1362 gm. This difference of 25 gm. is not significant.

Daily observations on the behavior of the chickens in this group seemed to indicate that the thymectomized birds were somewhat more nervous. Both the males and the females appeared to be more restless and flighty, especially when handled, than were the controls. Though there was an indication that thymectomy may have resulted in increased activity of the birds, there is no indication of a direct influence on the body weight.

Egg Data on Trap-nested Hens

The hens were trap-nested, and as the eggs were layed they were taken from the nest, marked with hen number, date and egg weight in grams. The first eggs of each (Tables II-VI) bird were small since these were the

Table II Showing data on the eggs of thymectomized Hen No. 3212.

Egg laying date	Wt. of egg in gm.	Appearance of shell	Shell strength in gm.	Remarks
1-5-29	32	Pitted	4968	
1-7-29	38	Slightly pitted	5572	
1-8-29	36	Pitted	6270	
1-11-29	41	Slightly pitted	4720	
1-13-29	39	Slightly pitted	4720	
1-23-29	40	Pitted	5207	
1-24-29	41	Badly pitted	4445	
1-26-29	45	Slightly pitted	4617	
1-30-29	46	Badly pitted	4325	Very thin area on end.
1-31-29	41	Pitted	5300	
2-10-29	42	Pitted	5500	Spots of large area.
2-12-29	47	Pitted	3940	
Average	40.6		4965	

Table III Showing data on the eggs of thymectomized Hen No. 3226.

Egg laying date	Wt. of egg in gm.	Appearance of shell	Shell strength in gm.	Remarks
1-2-29	41	Slightly pitted.	5095	
1-4-29	45	Slightly pitted.	4565	
1-5-29	45	Normal	5725	
1-7-29	45	Slightly pitted.	5715	
1-9-29	46	Normal	5572	
1-10-29	45	Normal	5938	
1-12-29	46	Normal	6477	
1-14-29	46	Normal	4745	
1-17-29	49	Slightly pitted	5322	
1-19-29	48	Normal	5398	
1-22-29	48	Normal	5565	7-8 large pits in one spot on side of egg.
1-24-29	46	Pitted	5843	
1-26-29	48	Normal	4340	
1-28-29	49	Normal	5200	
1-29-29	46	Normal	5623	
1-31-29	49	Normal	5270	
2-2-29	48	Normal	5500	
2-4-29	48	Normal	4860	
2-5-29	46	Slightly pitted	5270	

Egg laying date	Wt. of egg in gm.	Appearance of shell	Shell strength in gm.	Remarks
2-7-29	51	Finely pitted.	6716	
2-8-29	47	Slightly pitted	4300	
2-10-29	49	Slightly pitted	6462	
2-13-29	49	Slightly pitted	5900	
2-14-29	45	Slightly pitted	3600	
2-16-29	48	Slightly pitted	5400	
2-18-29	48	Normal	4500	
Average	46.9		5226	

Table IV Showing data on the eggs of thymectomized Hen No. 3230.

Egg laying date	Wt. of egg in gm.	Appearance of shell	Shell strength in gm.	Remarks
1-11-29	41	Slightly pitted	4226	
1-12-29	40	Normal	3680	
1-14-29	40	Normal	4600	
1-15-29	40	Slightly pitted	3735	
1-18-29	41	Slightly pitted	3523	
1-20-29	38	Normal	3582	
1-21-29	42	Slightly pitted	4664	
1-23-29	44	Slightly pitted	3556	
1-25-29	43	Normal	3340	
1-26-29	42	Slightly pitted	3200	
1-28-29	45	Slightly pitted	3762	
1-29-29	42	Slightly pitted	3020	
1-31-29	43	Slightly pitted	3753	
2-1-29	44	Pitted	3543	
2-3-29	43	Badly pitted	3666	on one end.
2-4-29	44	Pitted	2676	
2-5-29	42	Badly pitted	2776	on one end.
2-7-29	45	Badly pitted	3242	on one end.
2-8-29	44	Badly pitted	3270	on one end.
2-10-29	45	Badly pitted	3329	on one end.
2-11-29	44	Slightly pitted	3188	
2-13-29	45	Badly pitted	2813	on one end.
2-14-29	42	Badly pitted	3400	on one end.
2-16-29	44	Badly pitted	2500	on one end.
2-18-29	45	Slightly pitted	3450	on one end.
Average	42.72		3459	

Table V Showing data on the eggs of control Hen No. 3242.

Egg laying date	Wt. of egg when layed in gm.	Appearance of egg shell	Shell strenth	Remarks
12-7-28	35	Slightly pitted	2855	
12-10-28	35	Slightly pitted	4005	
12-11-28	37	Slightly pitted	2912	
12-13-28	36	Normal	2976	
12-23-28	38	Slightly pitted	3098	
12-24-28	37	Slightly pitted	3791	
12-26-28	45	Slightly pitted	5081	
12-28-28	38	Slightly pitted	2603	Badly pitted in spots.
1-2-29	38	Slightly pitted	2455	
1-7-29	38	Slightly pitted	2059	Badly pitted at small end.
1-10-29	40	Pitted at end	2808	
1-12-29	40	Slightly pitted	2652	
1-14-29	44	Slightly pitted	2673	
1-22-29	44	Slightly pitted	2790	
1-23-29	40	Slightly pitted		
1-25-29	46	Normal	2771	
1-28-29	44	Pitted	2030	Weak on small end.
1-29-29	43	Pitted	3412	Weak spot on small end.
1-31-29	44	Pitted	2505	Weak spot on small end.
2-1-29	45	Pitted	2500	Badly on side of small end.
2-2-29	43	Badly pitted	2872	
2-3-29	45	Badly pitted	5662	
2-4-29	45	Pitted	2216	
2-5-29	45	Badly pitted	2780	
2-7-29	47	Pitted on end	2536	
2-8-29	43	Pitted on end	2986	
2-9-29	44	Badly pitted	3187	
2-12-29	42	Badly pitted	2173	
2-14-29	44	Pitted	3049	
2-15-29	44	Badly pitted	2750	
2-17-29	44	Pitted	2234	
2-18-29	41	Normal	3434	
Average	41.6		2963	

Table VI Showing data on control Hen No. 3273.

Egg laying date	Wt. of egg in gm.	Appearance of shell	Shell strength in gm.	Remarks
12-23-28	39	Pitted	4081	
12-26-28	39	Badly pitted	2487	Badly pitted on one end especially.
12-29-28	40	Badly pitted	5423	Uniformly thick shell.
12-31-28	45	Pitted	6912	
1-2-29	45	Badly pitted	4557	
1-3-29	62	Pitted	3924	Badly pitted and wrinkled on small end.
1-8-29	44	Slightly pitted.	5666	
1-9-29	45	Badly pitted	7187	Very thick shell.
1-11-29	46	Badly pitted	4640	
1-15-29	46	Pitted	6056	
1-17-29	44	Pitted	6986	
1-18-29	46	Badly pitted	6220	Especially at ends.
1-20-29	47	Badly pitted	5264	
1-22-29	46	Pitted	6603	
1-23-29	45	Badly pitted	6641	
1-25-29	49	Finely pitted	6172	
1-26-29	48	Badly pitted	4112	
1-28-29	45	Pitted	5000	
1-29-29	46	Badly pitted	4339	
1-31-29	46	Pitted	4582	
2-2-29	49	Finely pitted	4068	
2-5-29	48	Pitted	4530	
2-7-29	49	Slightly pitted	4000	
2-8-29	48	Pitted	6040	
2-10-29	49	Slightly pitted.	5518	
2-13-29	47	Slightly pitted.	4670	
2-14-29	47	Badly pitted	6487	
2-16-29	49	Finely pitted	5653	
2-18-29	49	Finely pitted	6318	
Average	46.4		5315	

first eggs of pullets. These eggs were tested for shell strength by placing the egg small end up in an apparatus which exerted pressure upon the small end of the egg by means of a wooden rod which supported a receptacle for physical sand. With the egg in position the sand was slowly placed in the container until the shell broke from the weight of the sand. This volume of sand was then weighed and its weight recorded as the shell strength. The appearance of the shell was determined by placing the egg in front of an egg testing apparatus, such as are commonly used. The structure as observed was then described as either normal, finely pitted, slightly pitted, pitted, or badly pitted. A glance at the Tables II-VI will show in every case, in the control as well as in the thymectomized, that there was a more or less pitted condition, so the factor causing this condition could not be attributed to the absence of thymus glands.

A summary of the data (Table VII) shows an average egg weight of 43.4 gm. in the thymectomized pullets and an average of 44 gm. in the control pullets, with a difference of only 0.6 gm.

The results on shell strength, although the averages are quite close, show a wide range. An average shell

strength of 2963 gm. in control pullet 3242 is less than the lowest average shell strength of the thymectomized pullets, which is 3459 gm. And an average of 5315 gm. in control 3273 is greater than the highest average shell strength of the thymectomized which is 5226 gm. Thus it will be seen that the shell strength of the thymectomized pullets averages higher and does not go as low as the shell strength of the controls, and that the thymus apparently has no effect upon the strength of the egg shell as was supposed by Riddle. The above statement is further strengthened by the autopsies which revealed that no lobes had been overlooked or regenerated.

As a further check of the condition of the shell, calcium carbonate tests were made on the egg shells from each hen which were dried, mixed, and ground to a powder in a mortar, and tested by an experienced chemist. The chemical analysis gave an average calcium carbonate test of 35.35 per cent in the thymectomized hens as compared with a calcium carbonate test of 35 per cent in the controls which showed essentially the same results.

From the results obtained in these experiments there is no indication that the thymus influences the production of normal egg envelopes.

Table VII Showing summary of data on the eggs of thymectomized and control hens.

Hen No.	Av. wt. of eggs in gm.	Av. shell strength in gm.	Per cent $CaCO_3$
Thymectomized Hens			
3212	40.6	4965	35.51
3230	42.72	3459	34.96
3226	46.9	5226	35.59
Average	43.4	4550	35.35
Control Hens			
3242	41.6	2963	35.25
3273	46.4	5315	34.75
Average	44	4139	35

Fertility and Hatchability of Eggs

Fertility and hatchability tests were made on eggs layed when the pullets were nine months old. Of the 19 eggs from the controls, 45 per cent were fertile; whereas, of the 40 eggs from the thymectomized birds, 65.8 per cent were fertile. The eggs of the control group were fertilized by a control cockerel, and those from the thymectomized group were fertilized by a thymectomized cockerel.

In a second fertility test made when the hens were 10 months old, 50 eggs from the thymectomized group and 30 eggs from the control group were incubated. The fertility of the thymectomized group was 84 per cent as compared with 63 per cent for the control eggs.

These eggs were continued in the incubator and tested for hatchability. The percentage of hatchability in the thymectomized group was 65 per cent as compared with 48 per cent for the controls. The chickens of both groups were normal, and were in a thrifty condition at the age of 6 weeks. It will thus be seen that in both fertility and hatchability the eggs from the thymectomized group were equal, if not superior to those of the control birds. As 60 per cent is considered a good average hatch, these results are up to standard for normal chicks.

Size of Body Parts

When the males were killed and autopsied certain observations and measurements were recorded for comparison (Table VIII). As Katsura held that the secondary sexual characters of the thymectomized males were more pronounced than those of the control males, measurements were made of the lengths of the left and right spurs of each bird. In the thymectomized males the left spurs averaged 9.78 mm., and the right spurs 10.75 mm.; whereas, in the controls the left spurs averaged 8.71 mm. and the right spurs 8.85 mm. By comparing the spur measurements of the thymectomized males with those of the controls, it was found that the former were longer, the difference being almost significant. As far as could be discerned there was no constant difference in the appearance of the scales of the legs, the feathers, or the comb and wattles between the two groups.

The measurement of the tibio-tarsus as a typical bone of the skeleton revealed almost identical average bone lengths and about the same range of lengths in the thymectomized as in the control males. In these cases, at least the thymus showed none of the influences upon the bony growth claimed by certain workers.

Table VIII Measurements of Body Parts of Cockerels

Chicken No.	Spurs mm. L.	R.	Tibio-tarsus mm.	Scales	Comb and wattles	Feathers	Intestinal tone	Testes wt. in gm.	Gizzard wt. in gm	Liver wt.in gm.	Urates	Remarks on thymus
					Thymectomized							
3209	10	9	151	y.r.s.	red	thick	good	17	39.5	19	absent	No thymus
3210	11	12	147	y.r.s.	"	"	"	9	28	19	"	"
3219	8	9	153	y.r.s.	"	"	"	13	34	19	"	"
3222	11	11.5	160	r. s.	"	"	"	6	34	20.5	"	"
3223	9	11	152	y.r.s.	"	"	"	13.5	31	17	"	"
3231	13	12	150	y.r.s.	"	"	"	7	31.5	17.5	"	"
3233	6.5	9	159	y.r.s.	"	"	"	12	41.5	22.5	"	One lobe (2x4mm.)
Av.	9.78	10.75	153					11.07	34.21	19.21		
					Controls							
3243	6	6	149	pale y r. s.	red	thick	good	10	37	21.5	absent	Very large
3244	9	9	153	y.r.s.	"	"	"	7.5	27	22	"	Medium
3246	8	10	155	rough irreg.	"	many pin feathers	"	13.5	29	20	"	Rather large
3250	12	11	150	y.r.s.	"	thick	"	9.25	31	18	"	Large
3278	10	12	157	y.r.s.	"	"	"	9	26	21.5	"	Medium
3280	8	7	154	y.r.s.	"	"	"	15.5	39	21	"	Large
3283	8	7	158	y.r.s.	"	"	"	4	30.5	23	"	Very large
Av.	8.71	8.85	153.71					9.82	31.35	20		

y.- yellow
r.- regular
s.- smooth

The weights and conditions of some of the organs were noted with interest. In all cases the intestinal tone was found to be good, and urates absent.

The average weight of 34.21 gm. for the gizzard of the thymectomized as compared with the average weight of 31.35 gm. in the controls, giving a difference of 2.86 gm. hardly seems sufficient to consider, especially since there was a considerable amount of variation in size.

The average weights of the testes of the thymectomized cockerels was 11.07 gm. while the average weight of the testes of the controls was 9.82 gm. The advantage of 1.25 gm. which the thymectomized have over the controls is not in accordance with the findings of either Soli or Katsura, who described delayed maturity in the thymectomized males.

The first six of these thymectomized males showed no regenerated thymus lobes and the seventh only one very tiny lobe which is contrary to the statement that thymectomy cannot be carried out in chickens.

Blood Sugar Tests

To note the effect of thymectomy on blood sugar, 5 cc. of blood were taken by cardiac puncture from each bird and tested in a chemical laboratory. The eight thymectomized cockerels had an average of 152.7 mg. of sugar per hundred cubic centimeters of blood, while the

11 control cockerels had an average of 152 mg. The ten thymectomized pullets had an average of 145.6 mg., and the eight control pullets an average of 151.1 mg. (Table IX). From these tests there is no indication that thymectomy affects the sugar content of the blood as the variability in the pullet groups places the difference within the limits of experimental error.

Effects on Resistance to Parasitism

To ascertain if there is a direct or indirect relation between the thymus gland and the resistance of the chicken to an intestinal parasite, 50 embryonated eggs of the large roundworm Ascaridia linneata(Schneider)were fed to each bird of both the thymectomized and control groups. At the end of three weeks of parasitism all of the cockerels were killed and examined. It was found that both groups were exceedingly resistant, the difference being not significant.

EXPERIMENT III

In this experiment thymectomy was begun when the chicks were eight days old and continued until they were 21 days old when one-half of the young birds had been thymectomized. Since the technique of the removal of the thymi had been so

Table IX Showing blood sugar data on thymectomized and control chickens.

Thymectomized			
Chicken No. Pullets	Mg. sugar in 100 cc. of blood	Chicken No. Cockerels	Mg. sugar in 100 cc. of blood
3211	151.4	3209	172.4
3212	130.2	3210	138.8
3216	133.2	3219	149.2
3224	156.2	3222	135.2
3225	122.4	3223	127.4
3226	154.4	3231	152.6
3227	156.8	3232	135.6
3228	148.6	3233	193.2
3229	146.0		
3230	156.8		
Average	145.6		152.7
		Controls	
3237	142.8	3243	182.6
3238	158.8	3244	148.2
3240	149.2	3246	128.6
3241	164.6	3247	146.6
3242	158.8	3250	130.2
3253	145.5	3257	141.4
3273	137.	3258	170.2
3284	154.4	3274	155.
		3278	166.6
		3283	146.6
		3280	
Average	151.3		152.

carefully developed, no exploratory operations for regenerated lobes was deemed necessary, and autopsy showed this supposition to be well grounded.

Weekly weights of the birds were made from the time they were nineteen days of age until they were five months old. Comparing the thymectomized males with the control males, it was found that at nineteen days of age the former averaged 85 gm., and the controls 99 gm. (Fig. 4). The controls maintained a slight advantage over the thymectomized birds until the eighth week, when the latter averaged a few grams heavier. This lead over the control males was maintained up to the fifth month when the weighing was discontinued. The average weight of 1291 gm. for the thymectomized birds as compared with 1239 gm. for the controls gives an average difference of 42 gm. which is not significant. By similar comparison of the pullets it was found that at nineteen days of age the thymectomized ones average 91 gm. in weight, and the controls 92 gm. (Fig 5). At the second weighing the controls had an average advantage of 9 gm. over the thymectomized ones by about the same amount until the twelfth week when the latter gained a slight advantage which they maintained until during the fifth month, at which point the controls again led slightly. At the close of the fifth month the average weight of the thymectomized pullets was 1212 gm. as compared with 1240 gm.

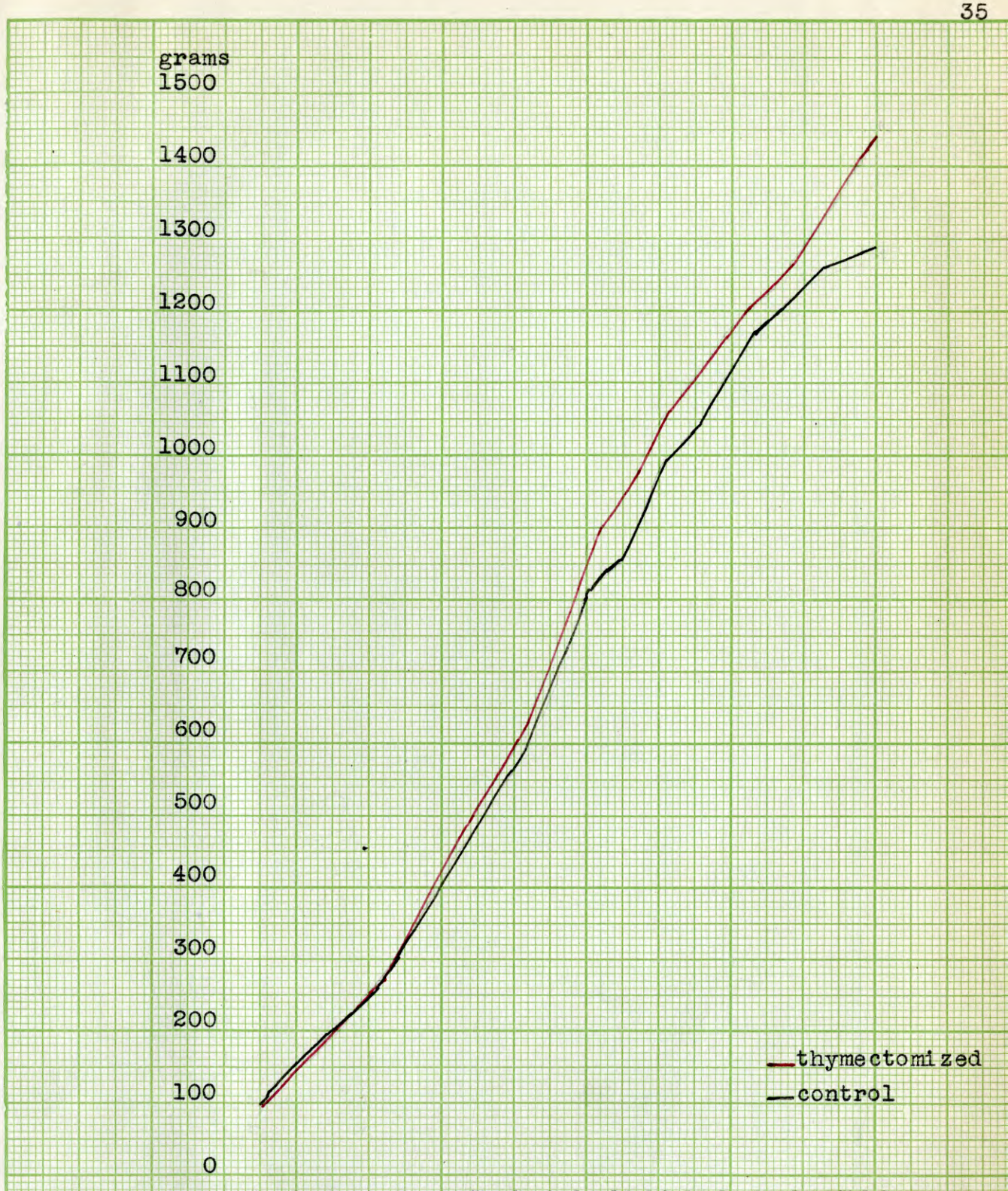


Fig. 4 Showing comparative weights of the thymectomized and control cockerels.

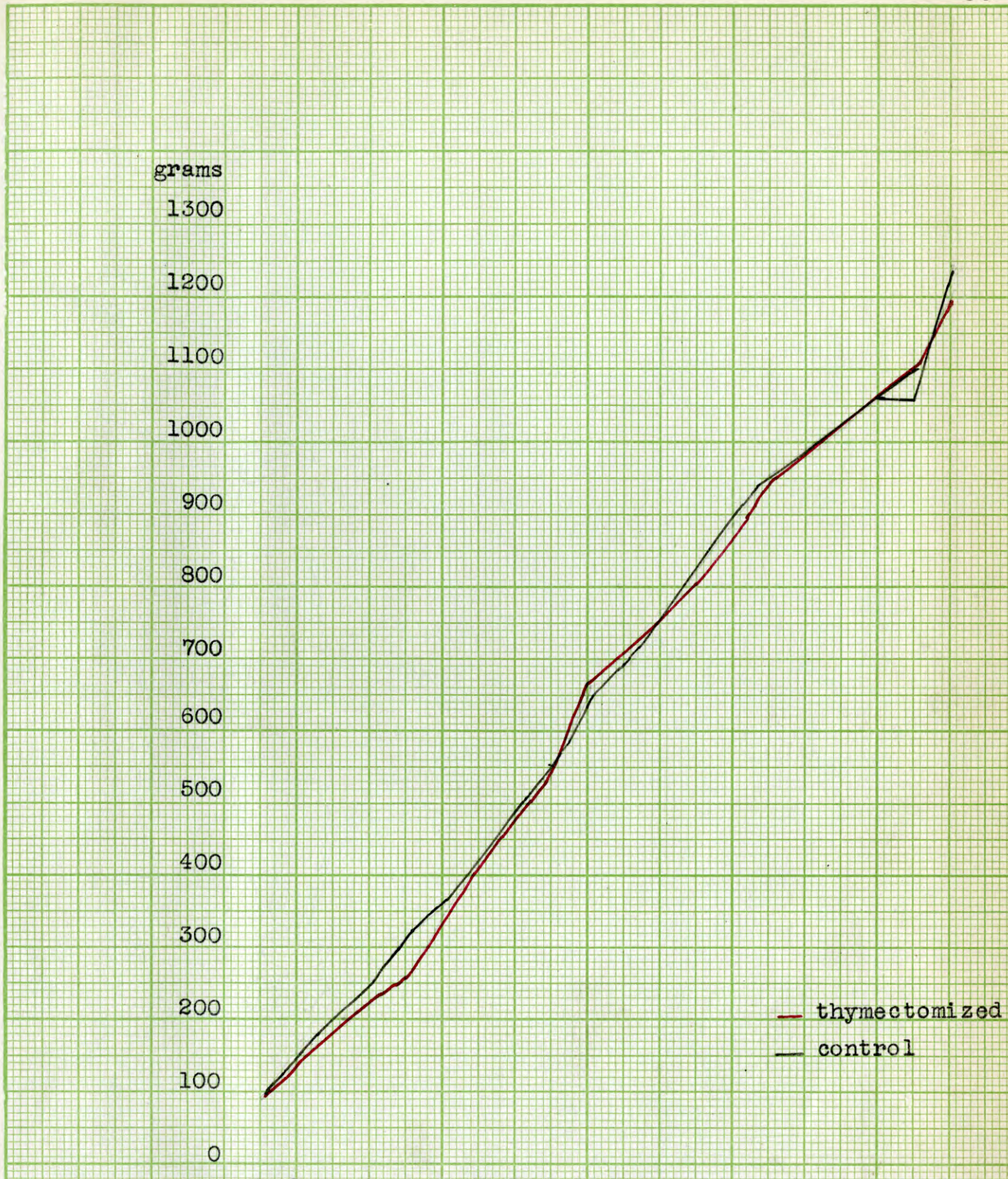


Fig. 5 Showing comparative weights of the thymectomized and control pullets.

for the controls. The slight difference in the weights of the two groups and the changing advantage, first in one group and then in the other, indicate that the thymus glands are not a factor in the development of growing chicks. The increased nervousness which appeared to be characteristic of the thymectomized birds in Experiment II was not apparent in the operated chickens of this experiment, as it was not possible to discern any definite difference in the behavior of the two groups under comparison. When the birds were nervous or flighty, it was noticed as much in the control chickens as in the thymectomized ones, and could probably be attributed to the normal behavior of this breed.

Size of Body Parts

A study of spur lengths of the cockerels in this experiment showed that those of the thymectomized males averaged 6.79 mm. as compared with 5.07 mm. for the controls. This difference of 1.72 mm. is 3.44 times its probable error and therefore would approach significance if the number of chickens were larger (Table X.).

A comparison of the appearance of the scales of the legs, color of the wattles and comb and thickness of the feathers gave no constant difference, and in most cases no difference at all.

Table X Measurements of body parts of cockerels.

Chicken	Spurs L.	Spurs R.	mm. Tibio tarsus mm.	Scales of legs	Comb and wattles	Feathers	Intestinal tone	Testes Wt.gm.	Gizzard Wt.gm.	Liver Wt.gm.	Urates Wt.gm.	Bursa Fabricii	Remarks
						Thymectomized							
3603	7	6	151	pale y. r. s.	red	thick	good	10	49.5	39	absent	2.1537	Slight regeneration
3604	6	7	157	y.r.s.	red	thick	good	21	25	31	absent		No thymi
3607	9	8	146	pale y. r. s.	red	fairly thick	good	9	31	21	absent	1.8110	No thymi
3608	9	8	146	y.r.s.	red	thick	good	16.5	25	21	absent		No thymi
3609	7	8	144	irregular y. rough	pale small	thick	good	12.5	39	30	absent	2.3350	No thymi
3611	3	3	154	y.r.s.	red	thick	poor caecum large	9	33	23	absent		One tiny lobe
3613	7	7	152	y.r.s.	red	thick	good	8	49	32	absent		No thymi
Average	6.79		150					12.28	35.92	28.14		2.0999	
						Controls							
3624	2	2	155	pale y. r. s.	red	fairly thick	good	7	31	24	absent	0.6660	Thymi medium
3628	6	6	142	y.r.s.	red	thick	good	13	34	25	absent	0.1117	Thymi large
3629	6	3	153	y.r.s.	red	thick	good	11	28	27	absent		Thymi large
3635	8	8	141	y.rough irregular	red	thick	good caecum large	15	32.5	25	absent	0.0921	Thymi medium to large
3637	1	2	154	y.r.s.	red	thick	good	5.5	34.5	27	absent		Thymi fairly large
3638	8	8	149	y.r.s.	red	thick	good	14	26	26	absent	0.2416	Thymi medium to small
3640	6	5	151	y.r.s.	red	thick	good	13.5	28	27	absent		Thymi medium
Average	5.07		149.28					11.28	30.57	25.85		0.27785	

r.-regular, y -yellow, s.-smooth.

The average length of 150 mm. for the tibio-tarsus in the thymectomized birds and of 149.28 mm. in the controls leaves a difference in length too small to consider. With but one exception the intestinal tone was good, and urates were entirely absent. The average weight of the gizzard in the thymectomized birds was 35.92 gm. while that of the gizzard of the controls was 30.57 gm. giving a difference of 5.35 gm. which again puts the thymectomized ahead of the controls. A similar comparison of the weights of the livers shows a small difference of 3.29 gm. in which the thymectomized again have the advantage. In comparing the weights of the testes although there is a range of 13 gm. in the thymectomized group and of 9.5 gm. in the control group, the averages of 12.28 gm. and 11.28 gm. gives the thymectomized a slight advantage of one gram.

An article by Riddle (1928) stating that the thymus glands and the bursa Fabricii are closely related in structure and function led to the observation and weighing of the bursa Fabricii of those birds which had not been previously autopsied. In the thymectomized cockerels the glands were fairly thick walled glandular pouches, which are dorsal to the cloaca, opening into it through a small duct. In the control cockerels the glands were small

fibrous masses. A consideration of the average weight of 2.0999 gm. of the bursa Fabricii of the thymectomized males and that of 0.2778 gm. in the controls gives a marked difference in the two groups, the bursa Fabricii of the thymectomized birds being over seven times as heavy as those of the controls. Perhaps these remarkable differences in appearance and weight may indicate as suggested a similar function which has been taken over by the bursa Fabricii in the absence of the thymi.

Observations of the different parts of certain pullets (Table XI) for the most part gave no outstanding differences. The spurs were small in each bird, the legs smooth and yellow, the comb and wattles red and the feathers thick. A comparison of the tibio-tarsus shows a length of 134 mm. in the thymectomized pullets, and a length of 126 mm. and 120 mm. in the controls. Body weights taken at this time were very close in the four birds, and this difference in the lengths of the tibio-tarsus is indicative of the findings of certain other workers. As a whole the internal organs were normal, the pancreas was a normal color, the intestinal tone was good, the spleen was normal and there were no urates. The livers in the two thymectomized were dark red and those of the two controls were brown. This brown color was found in other hens, especial-

Table XI Body parts of pullets.

Chicken No.	Spur mm. R	Spur mm. L	Tibio tarsus mm.	Scales of legs	Comb and wattles	Feathers	Pancreas	Intestinal tone	Ovary	Gizzard gm.	Liver gm.	Urates
3606*	3	3	134	y.r.s.	red	thick	normal	good	ova small	37	26	none
3641†	1	2	126	y.r.s.	"	"	"	"	laying	26	25	"
3605*	1	1	134	y.r.s.	"	"	"	"	ova small	24	24	"
3630**	2	2	120	y.r.s.	"	"	"	"	laying	24	27	"

* Thymectomized
† Control
**Control. Five large yolks. 1 egg in oviduct.

Weights of bursa Fabricii of the pullets.

Thymectomized	Bursa Fabricii in gm.	Controls	Bursa Fabricii in gm.
3606	3.4700	3630	0.63100
3617	0.5346	3636	0.22500
3619	1.3800	3641	0.10115
3621	0.7230	3642	0.84000
3622	1.7562	3645	0.44760
3623	2.3122	3647	1.7943C
Average	1.6960		0.67317

ly those that were laying. The weights of the livers were close and contribute no factor. The weights of the gizzards range in such a manner that they need not be discussed. Considering the degree of maturity of these four pullets it was found upon autopsy that the two controls were laying and there was a cluster of large yolks in each pullet. While in the thymectomized the ova were as yet very small. So it might be said that the maturity of the two thymectomized pullets was delayed as compared with the two controls.

The rest of the pullets of Experiment III were operated and the bursa Fabricii were removed and preserved in four per cent formalin. These glands were weighed, (Table XI) on an analytical balance, after being dried upon filter paper. Further drying during the weighing process was found to be very slight, and since each gland required about the same amount of time for weighing, the evaporation rate was not taken into consideration. A study of the table shows that the bursa Fabricii of the thymectomized average over two times the average weight of the control pullets. While the gland of control 3647 was larger than three of the thymectomized, on the other hand none of the glands of the controls were as large as those of thymectomized hens 3606 or 3623, and those of control 3636, 3641 and 3645 were appreciably smaller. These

thymectomized and bursectomized hens are being kept for further study.

Hemoglobin Estimate

Among other observations upon the conditions of these chickens it seemed desirable to have a test of the hemoglobin percentage in the blood. The individual tests show only slight variations in both the thymectomized and control birds. (Table XII).

The average of 8.06 per cent in the thymectomized females as compared with an average 8.13 per cent in the control females, gives a difference of only .07 per cent which is not significant. About the same condition exists between the cockerels, with an average of 9.25 per cent in the thymectomized and 9.9 per cent in the controls.

SUMMARY

1. Three experiments involving 90 single comb white leghorn chicks raised in confinement were carried out to ascertain possible effects of thymectomy upon the birds.

2. Of the surgical methods tried that of making a single, median, vertical, incision extending from the region of the larynx to the base of the crop was found to be the most satisfactory. With the aid of a hand magnifier all portions of the thymus lobes were readily removable.

Table XII Showing percentages of hemoglobin in the blood of thymectomized and control chickens.

Cockerels	Hemoglobin per cent	Pullets	Hemoglobin per cent
		Thymectomized	
3603	9.35	3605	7.18
3604	10.41	3606	8.77
3607	7.89	3617	7.74
3608	10.62	3618	9.39
3609	8.58	3621	8.16
3611	8.43	3622	7.45
3613	9.51	3623	7.72
Average	9.25		8.06
		Controls	
3624	8.77	3626	8.16
3628	9.99	3627	8.08
3629	9.99	3630	8.43
3635	9.51	3633	8.08
3637	11.22	3639	6.81
3638	10.23	3641	8.16
3640	9.58	3642	7.74
		3645	8.97
		3646	8.58
		3647	8.26
Average	9.90		8.13

3. The following tests were made upon thymectomized and control birds from the same hatches without securing significant differences:

a. Weight of eggs.

b. Strength of egg shells.

c. Calcium carbonate in egg shells.

d. Appearance of and thickness of shell.

e. Fertility and hatchability of eggs.

f. Lengths of spurs and tibio-tarsi.

g. Weights of testes, livers, gizzards, bursa Fabricii.

h. Hemoglobin estimate of blood.

i. Sugar in blood.

j. Resistance to intestinal roundworm, Ascaridia lineata (Schneider).

4. Tests on 19 chickens indicate a relationship between the thymus glands and the bursa Fabricii (cloacal diverticula).

ACKNOWLEDGMENTS

The writer wishes especially to thank Dr. J.E. Ackert, with whom these experiments were undertaken, and also Dr. R.W. Titus under whose direction the blood sugar tests were made; Dr. J.S. Hughes for the hemoglobin estimate and Dr. H.W. Brubaker for the calcium carbonate tests.

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