Morphological Studies on Effects of the Bursa of Fabricius and Thymus of Chicks Hatched from Eggs in Treatment of Testosterone Propionate

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A considerable amount of literature has been published pertaining to the normal growth and role of bursa Fabricii in chicks by such authors as HAMMOND¹⁾ (1942), $GLICK^{2)3}$ (1955, 1956) CHANG, REEINS & WINTER⁴⁾ (1958), TEMPLE & JAAP⁵⁾ (1960) et al. Also, on the response of antibody reproduction in bursa were studied such authors as $GLICK^{6)7}$ (1960, 1961), SADLER & $GLICK^{8)}$ (1962), MAY & $GLICK^{9)}$ (1964), SATO & $GLICK^{10}$ (1965) et al. They suggested that bursa is a central lymphoid tissue that influences the immunological competence of a chicken. On the other hand, MAY & $GLICK^{9}$ (1964), WILSON & $GLICK^{11}$ (1966) has dealt with morphological studies on the bursa to induce testosterone propionate (TP) into fertile eggs. ASPINALL, MEYER & RAO¹²⁾ (1960) reported the effects of various androgen like steriods on the development of the bursa of Fabricius, the thymus and spleen were studied in chick embryo. These workers reported a significantly smaller bursa or no bursa in birds hatched from eggs in TP treatment.

The primary purpose of this study is to report the quantitative changes occuring in the weight of the bursa of Fabricius and thymus in chicks hatched from eggs in treatment in varying levels of TP solution. On the basis of these data it became of interest to make one study of the effects of steroids on the differentiation of the bursa.

The authors is deeply greatful to Mr. T. Tomita, Mr. H. Saito and Mr. M. Kunimitsu for technical asistance of laboratory work during the course of this study.

MATERIALS AND METHODS

The material used was fertilized ova of White leghorn strain. In the first experiment, 255 eggs were removed from incubator on the third days of incubation. The pointed end of the eggs was immersed in the treatment solution $(7-14^{\circ}C)$ for 7 seconds. The five trials incubated to the following treatment: 160 mg., 320 mg., 480 mg., 640 mg. and 1200 mg. testosterone propionate per 100 ml. ethyl alcohol according to MAY & GLICK's method⁹⁾ (1964). In the second experiment, 100 eggs were injected in 0.5 mg. and 1.0 mg. testosterone propionate per suspention distilled water for injection into the albumin portion on the five days of incubation. The bursa and thymus weights were recorded five days after hatching in dipped eggs and first

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day after hatching in injected eggs. Chick embryos in the still-development in two experiments was observed in morphology.

OBSERVATIONS

The date herein presented were based on observations with the quantitative changes on the bursa and thymus weights and the still-development embryos in the fertilized eggs from the two experiments.

In the first experiment, a number of the chicks hatched from eggs dipped in varying concentration of testosterone propionate was observed small in average number compared with control (Table 1). That is, average percents in the hatching

Section	Number of experiment	lst candling	2nd candling	Number of hatching
Control (Alcohol)	55	35	25	15(42%)
TP 160 mg.	55	37	24	9(24%)
TP 320 mg.	55	28	18	4(14%)
TP 480 mg.	55	27	17	6(22%)
TP 640 mg.	55	28	19	7(25%)
TP 1200 mg.	35	18	8	4 (22 %)

Table 1. Number of chicks hatched from testosterone propionate dipped eggs.

number of fertilized eggs at five trials in TP 160 mg., 320mg., 480 mg., 640 mg. and 1200 mg. treatment was 24, 14, 22, 25 and 22 percent at hatchability in each at to hatching from first candling of 5 days incubation. The control experiment of a simple ethyl alcohol treatment was 42 percent. Dipping eggs in TP solution of 640 mg. and 1200 mg. significantly reduced bursa size and weight at hatching. The development of the thymus no remarkable difference in weight was observed by five trials as sign Table 2. Chick embryos in the still-development in dipping eggs in TP solution

	Control		Level of TP, mg/100 m/. ethyl alchol			
	Control	160	320	480	640	1200
Body weight (g) 43.6	43.4	39. 5	43.0	44.6	43.2
Bursa of Fabricius (mg	.) 64. 5	53.6	54.8	42.3	28.3	20.0
Thymus (mg) 232.6	225.2	242.0	102.0	231.1	221.8

 Table 2.
 Average weight of the bursa of Fabricius and thymus of five days-old chicks hatched from testosterone propionate dipped eggs.

was observed various deformities. Namely, abnormality of brain development, abnormality of optic vesicle, three eyed, two beaks and abnormality of beak development was observed (Plate 1). Small number of chicks hatched from eggs dipped in TP solution was observed in abnormal beak, abnormal leg, abnormal eye ball and protrusion of the eye ball (Plate 2).

In the second experiment, number of the chicks hatched from eggs injected in TP 0.5 mg. and 1.0 mg. into the albumin portion was normal hatchability contrast with control (Table 3). TP injected eggs significantly reduced bursa size and weight at hatching. Especially, bursa of TP 1.0 mg. injected chicks was observed remarkable degeneration as seen in Table 4. Development of the thymus no remarkable

Section	Number of experiment	lst candling	2nd candling	Number of hatching
Control	50	42	38	35 (83%)
TP 0.5 mg.	50	44	44	39 (89%)
TP 1.0 mg.	50	42	37	33 (79%)

Table 3. Number of chicks from testosterone propionate injected eggs.

 Table 4. Average weight of the bursa of Fabricius and thymus of one day old chicks hatched from testosterone propionate injected eggs.

		Level of	ТР
	Control	TP 0. 5 mg.	TP 1.0 mg.
Body weight (g.)	34 • 74	34. 29	35.49
Bursa of Fabricius (mg.)	45.66	15. 81	4.86
Thymus (mg.)	229. 2	256. 1	192. 8

difference in weight contrast with control was observed similar to first experimental result. Chick embryos in the still-development in injected eggs was observed abnormal optical vesicle, abnormal legs and abnormal development of brain in morphology (Plate 1).

DISCUSSION

The present investigations have dealt with the one study of the bursa of Fabricius, particulary on morphological study on developmental effects of bursa and thymus of chicks hatched from eggs treated in testosterone propionate.

The papers has been extended to analyse the effects of steroids on the bursa with particular concern to the quantative reaction and on the thymus as similar to contraction of lymphoid tissue. MAY & $GLICK^{9}$ (1964), WILOSON & $GLICK^{11}$ (1966) reported the changes occuring in the weight of the bursa in birds hatched from eggs dipped in TP solution and dipping eggs in TP solution of 320 mg. or higher reduced bursa size at hatching. The present author has undertook morphological observations on the bursa and thymus from two experiments in TP dipped eggs and TP injected eggs. It was observed that the bursa are notable reduction in chicks hatched from higher unit of dipped eggs and injected eggs.

Only based on the data obtained from the present observations, the following assumption is no determinate that the bursa undergo effect of steriods in immediate reaction. But, based on the results of the present study the assumption is possibly made that TP hormone has a great influence on the differentiation of the developmental bursa in immature stage of chick. ASPINALL, MEYER & RAO^{12} (1960) reported the bursal inhibitory potency of these steroids depends upon their metabolism in the embryo. The degree of activity may be dependent upon the relative amount of each steroid that is metabolized to a more potent analogue.

According to the results of the present study, abnormal chick embryos in the still-development are generally more common in TP treatment eggs than in control eggs. The data indicated that the abnormal embryo was an occurrence through TP treatment in early stage of incubation. It is a study for further discussion on the ultimate cause of deformed embryos on this experiment.

SUMMARY

This study was undertaken to determine the quantitative changes occuring in the weights of the bursa and thymus in chicks hatched from eggs in treatment in varying levels of testosterone propionate. The data obtained in the presented investigations were described being separated into two parts.

1) Dipping eggs in TP solution of 640 mg. or 1200 mg. and TP injected eggs of 0.5 mg. or 1.0 mg. siginificantly reduced bursa size and weight at hatching. Especially, TP 1.0 mg. injected eggs is remarkable reduction as compared into the other experiments. On the other hand, the development of the thymus showed a tendency to no effects on chicks hatched from eggs in TP treatment.

2) Number of the chicks hatched from dipped in TP solution was observed small in average number compared with control. Chick embryos in the still-development in TP treatment various deformities were observed.

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テストステロンプロピオネート処理鶏卵の孵化雛のファブリシウス嚢 および胸腺の発育影響に関する研究

中 村 経 紀

鶏卵の孵卵3~5日目にテストステロンプロピオネート (TP 160mg., 320mg., 480mg., 640mg., 1200mg.) を浸漬または卵白内に注入 (TP 0.5mg., 1.0mg.) し, 孵化後の発育雛のファブリシウス嚢および 胸腺 の発育影響について研究した.

実験結果は(1) TP 浸漬実験区は各単位区とも 孵化率が極めて低く, TP注入実験区は対照区と比較 して正常であった.(2)ファブリシウス嚢および胸腺の発育影響は TP 640mg. および 1,200mg の浸漬 実験区および注入実験区で著しい退化を認め,特に TP 1.0mg. 注入卵は顕著であった. これは発育初期 のステロイド系ホルモン高単位処理によるファブリシウス嚢発育に影響をおよぼしたことを示した.他 方同一淋巴様組織である胸腺の影響は認めなかった.(3) TP 実験区の発生中止卵には異常発生をな した胚が多く観察され三眼鶏, 双嘴, 脳胞発育異常, 眼胞異常等が形態的に観察され, また孵化雛には 眼球異常, 肢異常等が認められた.

EXPLANATION OF FIGURES

- Plate 1. Abnormal chick embryos in the still-development in TP treatment.
- Fig. 1. Embryo of three eyed in TP treatment.
- Fig. 2. Embryo of abnormal optical vesicle in TP 1.0 mg. injection.
- Figs. 3-4. abnormal development of opticle vesicle in TP 0.5 mg. (Fig. 3) and TP 1.0 mg. (Fig. 4) injection.

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Plate 1



Plate 2. Abnormal chicks hatched from eggs in TP treatment and abnormal embryos.

- Fig. 5. Embryo of abnormal beak in TP 0.5 mg. injection.
- Fig. 6. Chick of abnormal eye in 320 mg. treament.

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- Figs. 7-8. Chicks of abnormal legs in TP 640 mg. treatment.
- Fig. 9. Color photograph in abnormal chick embryo of two beaks and three eyed in TP 1. 200 mg. treatment.

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