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EFFECT OF PREGNANT MARE SERUM GONADOTROPIN ON EGG PRODUCTION AND SOME BLOOD SERUM HORMONES OF PEKIN DUCKS

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SUMMARY

Sixty Pekin ducks at 20 weeks of age were used to study the effect of pregnant mare serum gonadotropin (PMSG) on egg production and some related blood serum hormones. Ducks were randomly divided into four equal groups. Birds of all groups were housed in floor pens, and feed and water were offered ad libtum. Birds were fed a diet contained 17% crude protein, 2800 Kcal ME/Kg diet, 3.7% calcium and 0.66% total phosphorous. The experiment started in December and extended up to 35 weeks of age.

Ducks of group one were served as control (which injected by 2ml saline solution/bird) at week 20 and 30. Those of groups two and three were subcuteonuosly injected by 100 IU of PMSG /bird/day for three consecutive days at week 20 and 30 respectively. While those of group 4 were twice injected at week 20 and 30.

Obtained results indicate that PMSG injection improved ($P<0.05$) egg production (weight and number) and feed conversion. Moreover, the injection reduced ($P< 0.05$) the level of triiodothyronine (T_3) and prolactin in blood serum. While it increased ($P<0.05$) serum progesterone, estradiol, luteinizing hormone and T_4/T_3 ratio. However, serum thyroxine (T_4) level did not affected by treatment.

According to the obtained results, it could be concluded that PMSG injection might have a useful effect on egg production of low producer birds.

Keywords: Pregnant mare serum gonadotropin, egg production, thyroid function, ovarian hormones, prolactin, and luteinizing hormone.

INTRODUCTION

Egg production is associated with intensive metabolic activities under the action of gonadotropins (Sturkie, 1986). These hormones affect egg production within physiological limits, to afford behoof metabolic activities for the action of gonadotropins and female sex hormones.

They collectively regulate egg formation from ovulation to oviposition (Armstrong, 1984). Pregnant mare serum gonadotropin (PMSG) is glycoprotein, with two α and β subunits, structurally similar to FSH and LH with higher carbohydrates content, especially sialic acid (Sherwood and McShan, 1977). The higher sialic acid

content in PMSG appears to account for its long half-life for several days. Thus, a single injection of PMSG can be of a biological effect, at the target gland for more than a week (Hafez, 1985).

The purpose of the present study was to determine the possible of accelerating the process of egg formation and thus increasing production rate of Pekin ducks (*Anas platyrhynchos*) by PMSG injection, in accordance with metabolic and sex hormones.

MATERIALS AND METHODS

Experimental design

A total number of 60 female Pekin ducks aging 20 weeks with an average body weight of 1721 ± 28g was randomly chosen from a large commercial flock. Birds were randomly assigned to four equal groups. Birds in group one (G1) were untreated and served as control, which injected at week 20 and 30 of age by 2 ml saline solution/bird for three days. While those of group two (G2), three (G3) and four (G4) were subcuteonusly injected (2 ml/bird) of an aquatic solution containing 100 IU of PMSG (Folligon, Intervet International B.V. Boxmeer. Holland) immediately dissolved prior to administration. Time of injection differed among groups for three consecutive days. Injection of birds of G2, G3 and G4 was applied at 20; 30 (50% egg production of control hens) and at both 20 and 30 weeks of age, respectively.

Birds of each group were divided into three replicates each of five. Birds were housed in a floor pen of 1×1.5 m per replicate and were fed a diet containing 17% crude protein, 2800 Kcal ME/kg diet, 3.7% calcium and 0.66% total phosphorous according to NRC (1994) requirements. The experiment was started in December and extended to March (15 wk), with natural day length.

Egg production (weight and number) was recorded from the first laid egg up to 35 weeks of age with five weeks interval. Age at sexual maturity (SM) was estimated as days at which bird laid its first egg.

Blood Sampling

Blood samples were collected just before injection from five birds chosen randomly within each group. Samples of about 3ml of blood were withdrawn from the brachial vein into collecting tube and immediately centrifuged at 3000 rpm for 15 minutes. Blood serum was then stored at -20°C until analyses. Blood samples were collected at 20, 21, 25, 30, 31 and 35 weeks of age. However, blood samples were betided every time between 7:00 to 7:30 am, while ducks are behaved to lay eggs before 9:00 am (normally between 8:00 to 9:00 am).

Hormonal assays

Direct radioimmunoassay technique was performed to determine the serum hormones using avian ready antibody coated tube kits (Diagnostic Product Corporation, Los Angelos) according to the procedure outlined by the manufacturer. Serum thyroxine (T_4), triiodothyronine (T_3), progesterone (P_4), estradiol (E_2), prolactin (PRL), and luteinizing hormones (LH) were determined.

Sensitivity value of T_3 kits , was reported to be 0.09 ng/ml. Standard curve ranged from 0.8 to 2.0 ng/ml, and intra- and inter assay variation coefficients were 4.9% and 5.8%, respectively. The cross reaction with othe thyroxin metabolities were 4.9%,

and 5.8% with L- and D- thyroxine, respectivily. Sensitivity value of T_4 kits , was reported to be 0.98 ng/ml. Standard curve ranged from 6.0 to 12.0 ng/ml. Intra- and inter assay variation coefficients were 4.6% and 4.7%, respectively. The cross reaction with each of L- & D- triiodothyronine was 7.7 and 2.1, respectively. Sensitivity value of P_4 kits , was reported to be 0.04 ng/ml. Standard curve ranged from 0.1 to 0.5 ng/ml. Intra- and inter assay variation coefficients were 3.2% and 6.7%, respectively. The cross reaction with other steroid hormones was less than 0.5 ng/ml. Sensitivity value of E_2 kits , was reported to be 0.2 ng/ml. Standard curve ranged from 4.2 to 24.0 pg/ml. Intra- and inter assay variation coefficients were 4.4% and 8.9 %, respectively. The cross reaction with anu of the other steroid hormones was less than 0.5 ng/ml. Sensitivity value of PRL kits , was reported to be 0.1 ng/ml. Standard curve ranged from 3.0 to 19.0 ng/ml. Intra- and inter assay variation coefficients were 1.8% and 3.2%, respectively. The cross reaction with FSH, LH, and TSH, hormones was less than 5.0 ng/ml for each. Sensitivity value of LH kits , was reported to be 0.15 mIU/ml. Standard curve ranged from 0.4 to 12.5 mIU/ml. Intra- and inter assay variation coefficients were 1.3% and 3.7%, respectively. The cross reaction with other FSH, PRL, and TSH, hormones was less than 5.0 ng/ml for each.

Statistical Analysis

Data collected were statistically analyzed by the analysis of variance with the General Linear Model (GLM) procedure of the SAS institute (SAS, 1992). All statements of significance are based on the 0.05 level of probability. Significant of PMSG injection effects were separated using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

1 - Productive performance

Data of productive performance of laying ducks as affected by PMSG injection are listed in Table 1. Obtained data reveal that injected ducks at week 20 of age maturated ($P < 0.05$) earlier by about 15 days than control. However, injected ducks, at week 20, recorded ($P < 0.05$) higher egg number and ($P < 0.05$) heavier egg weight throughout the experimental period than those of control and those which injected only at 30 weeks of age. While the highest egg number and egg weight were recorded for the ducks which injected twice at week 20 and 30. The laying intensity could be mainly related to metabolic activity. This observation may also indicate a more complex relationship between thyroid hormones and gonodal function of laying ducks. Since thyroidal hormones are essential for energy metabolism as reported by Queen *et al.* (1997).

Data of feed intake and feed conversion are listed in Table 1. It seems that injected ducks (G2 and G4) ate ($P<0.05$) higher and converted food to eggs ($P< 0.05$) better than control. It is possible that nutrient intake increased, in part to meet the additional metabolic requirements of estrogen-sensitive target tissues. This relationship provide as indirect evidence to support the proposal that the increase in nutrient intake following PMSG treatment was required for oviductal tissues development and function of the formation of yolk precursors (Zadworny and Etches, 1988).

2 - Hormonal changes

Thyroid hormones: Data of thyroid activity T_3, T_4 and T_4/T_3 are presented in Table 2. Obtained data indicate that serum T_3 decreased ($P<0.05$) due to PMSG injection, while serum T_4 had ($P<0.05$) unchangeable values. However, T_4/T_3 ratio increased ($P<0.05$) by PMSG injection. This is scientifically logic since the ratio is a relation between the concentration of the two hormones. The results of egg production and thyroid hormones demonstrated that thyroid activity was of essential importance for both initiation and maintenance of egg production. Since it is correlated with the energy metabolism needed for biological endothermic reactions. May (1989) and Queen *et al.* (1997) speculated that a higher metabolism may cause a rapid conversion from T_4 to T_3, which is considered the more potent thyroidal hormone. Such mechanism could have been responsible for the egg production levels observed in the current study of PMSG injected ducks. The decline observed in serum T_3 level could also represent other mechanisms being related to feed intake. Klandorf and Harvey (1985) and Queen *et al.* (1997) pointed out that plasma T_3 decreased ($P < 0.05$) by increasing feed intake and speculated that the decrease of T_3 may be also due to the advance of productive status.

Table 1. Least squares means ±SE for productive parameters of Pekin ducks as affected by PMSG injection

Item	Age (wk)	G1	G2	G3	G4
Age at sexual maturity (d)		157 ±2.3^a	142 ±1.8^b	156 ±2.2^a	143 ±1.8^b
Egg number	20 – 25	8.7 ±0.4^b	13.2 ±0.2^a	8.4 ±0.6^b	14.2 ±0.3^a
	25 – 30	12.2 ±0.8^b	20.1 ±0.4^a	10.3 ±0.8^b	19.3 ±0.7^a
	30 – 35	17.4 ±0.4^c	22.8 ±0.9^a	19.6 ±0.8^b	25.4 ±1.1^a
Egg weight (g)	20 – 25	74.4 ±1.2^b	77.3 ±1.2^a	73.3 ±1.1^b	78.2 ±1.4^a
	25 – 30	76.8 ±1.4^b	79.6 ±1.6^a	76.2 ±1.0^b	82.3 ±1.4^a
	30 – 35	79.2 ±1.5^b	84.8 ±1.8^a	88.9 ±1.2^b	86.4 ±1.7^a
Feed intake	20 – 25	218.2 ±6.4^b	250.1 ±7.3^a	210.8 ±7.1^b	256.4 ±8.2^a
gm/bird/day	25 – 30	243.7 ±8.5^b	284.6±10.2^a	238.4 ±8.3^b	289.5 ±9.9^a
	30 – 35	275.8±10.0^b	312.8±12.5^a	281.8±10.2^b	332.8±13.8^a
Feed conversion	20 – 25	4.2 ±0.07^a	4.0 ±0.02^b	4.4 ±0.06^a	3.9 ±0.02^b
K. feed:kg eggs	25 – 30	4.6 ±0.06^a	4.3 ±0.04^b	4.5 ±0.07^a	4.2 ±0.04^b
	30 – 35	5.5 ±0.09^a	4.8 ±0.07^{bc}	5.0 ±0.09^b	4.7 ±0.08^c

Least squares means in the same row with no common superscripts differ significantly ($P<0.05$).

G1: Control. G2: Birds injected by 100 IU of PMSG at week 20 of age.

G3: Birds injected by 100 IU of PMSG at week 30 of age.

G4: Birds injected by 100 IU of PMSG at week 20 and 30 of age.

Progesterone (P_4) and estradiol (E_2): Obtained data of P_4 and E_2 are shown in Table 3. It was found that both of P_4 and E_2 concentrations in blood serum increased ($P<0.05$) due to PMSG injection by about three and five times for P_4 and E_2, respectively when compared with controls. The strong positive relationship between gonadotropin and ovarian hormones as reported by Etches (1993) may be strictly vindicates the greater increase of P_4 and E_2 concentrations in the blood serum due to PMSG. Tixier-Biochard *et al.* (1990); Johnson (1993) and Lee *et al.* (1998) speculated that release of gonadotropins stimulated the ovarian follicles progesterone secretion. In addition, the variation in serum E_2 level may be a function of existed the rate of yolk protein biosynthesis and egg shell calcium deposition (Gruber, 1972).

Prolactin (PRL) and luteinizing (LH) hormones: Data of PRL and LH are presented in Table 3. Obtained data revealed that PRL level in blood serum decreased ($P<0.05$) due to PMSG injection. On the other hand, LH level showed a greater increases. The divergent reaction of PRL and LH, pituitary gland hormones, suggests that pituitary gland hormones may have distinctly different effect on egg production efficiency as well as indicates the production level. Prolactin is thought to act through the hypothalamo-hypophyseal-ovarian axis to inhibit egg laying. It is well known that there is an antagonistic action between prolactin and FSH, which may be a good reason for the decrease of prolactin in non-injected birds.

Table 2. Least squares means ± SE for the serum thyroid hormones in pekin ducks as affected by PMSG injection

Item	Age (wk)	G1	G2	G3	G4
T_3 (ng/ml)	20	3.0 ± 0.3^{a}	2.8 ± 0.3^{a}	2.7 ± 0.4^{a}	2.9 ± 0.2^{a}
	21	3.2 ± 0.2^{a}	1.8 ± 0.2^{b}	2.9 ± 0.4^{a}	1.6 ± 0.2^{ab}
	25	3.4 ± 0.2^{a}	1.6 ± 0.5^{b}	3.4 ± 0.1^{a}	1.8 ± 0.3^{b}
	30	4.1 ± 0.2^{a}	1.7 ± 0.3^{b}	3.2 ± 0.2^{a}	1.8 ± 0.2^{b}
	31	3.7 ± 0.4^{a}	1.9 ± 0.2^{b}	2.1 ± 0.3^{b}	2.0 ± 0.4^{b}
	35	3.4 ± 0.3^{a}	1.8 ± 0.3^{b}	1.9 ± 0.3^{b}	1.8 ± 0.2^{b}
T_4 (ng/ml)	20	14.5 ± 0.4^{a}	14.8 ± 0.3^{a}	14.6 ± 0.4^{a}	14.5 ± 0.4^{a}
	21	15.3 ± 0.8^{a}	14.8 ± 0.6^{a}	15.5 ± 0.5^{a}	14.7 ± 0.6^{a}
	25	12.9 ± 0.4^{a}	12.7 ± 0.8^{a}	12.8 ± 0.6^{a}	13.0 ± 0.8^{a}
	30	12.7 ± 0.8^{a}	12.5 ± 0.7^{a}	12.3 ± 0.6^{a}	12.4 ± 0.7^{a}
	31	11.2 ± 0.5^{a}	11.4 ± 0.6^{a}	11.2 ± 0.4^{a}	10.9 ± 0.9^{a}
	35	11.4 ± 0.5^{a}	11.3 ± 0.5^{a}	11.5 ± 0.6^{a}	11.4 ± 0.8^{a}
T_4 / T_3	20	4.8 ± 0.9^{a}	5.3 ± 1.0^{a}	5.4 ± 0.8^{a}	5.0 ± 1.1^{a}
	21	4.8 ± 0.8^{a}	8.2 ± 1.6^{a}	5.3 ± 0.9^{b}	9.2 ± 1.7^{a}
	25	3.8 ± 0.9^{b}	7.9 ± 1.2^{a}	3.8 ± 0.8^{b}	7.2 ± 1.1^{a}
	30	3.1 ± 0.7^{b}	7.4 ± 1.4^{a}	3.8 ± 0.7^{b}	6.9 ± 1.3^{a}
	31	3.0 ± 0.8^{b}	6.0 ± 1.1^{a}	5.3 ± 0.9^{a}	5.5 ± 0.8^{a}
	35	3.4 ± 0.7^{b}	6.3 ± 1.0^{a}	6.1 ± 0.9^{a}	6.3 ± 0.9^{a}

Least squares means in the same row with different superscripts differ significantly ($P<0.05$).
G1: Control. G2: Birds injected by 100 IU of PMSG at week 20 of age.
G3: Birds injected by 100 IU of PMSG at week 30 of age.
G4: Birds injected by 100 IU of PMSG at week 20 and 30 of age

However, PMSG may create a state of coordination along hypothalamo-hypophyseal-ovarian axis in the favor of an efficient egg formation processed that resulted in higher egg production in the injected birds. This observation is also supported by the findings of Bedecarrats *et al.* (1997) and Karatzas *et al.* (1997). They speculated that PRL may act at the neural or pituitary level to inhibit gonadotropin secretion or acts directly on the ovary because gonadotropin-stimulated ovulation and steroid production are inhibited by exogenous PRL. In the present study, PMSG injection could activate ovarian function which resultant in low PRL level in blood serum.

Table 3. Least squares means ± SE serum progesterone (P_4), estradiol (E_2), prolactin (PRL) and lutinizing hormone (LH) in Pekin ducks as affected by PMSG injection

Item	Age (wk)	G1	G2	G3	G4
P_4 (ng/ml)	20	0.106±0.01^a	0.106±0.01^a	0.107±0.01^a	0.104±0.01^a
	21	0.104±0.01^b	0.376±0.06^a	0.108±0.01^b	0.321±0.07^a
	25	0.118±0.01^b	0.381±0.04^a	0.123±0.02^b	0.362±0.04^a
	30	0.172±0.02^b	0.379±0.05^a	0.168±0.02^b	0.389±0.06^a
	31	0.185±0.02^c	0.380±0.07^b	0.414±0.06^a	0.519±0.08^a
	35	0.190±0.02^c	0.398±0.07^b	0.491±0.07^a	0.508±0.08^a
E_2 (pg/ml)	20	36.7±2.8^a	38.2±2.4^a	34.7±2.3^a	38.5±2.4^a
	21	38.3±3.7^b	218.7±6.4^a	40.6±2.4^b	215.8±5.8^a
	25	40.4±2.8^b	222.5±4.7^a	42.7±2.7^b	212.4±4.6^a
	30	48.8 ±3.1^b	203.7±4.5^a	53.3±2.3^b	208.5±4.2^a
	31	56.6±3.2^c	208.6±4.3^b	290.9±5.6^a	285.6±5.3^a
	35	80.5±2.8^c	186.9±3.7^b	289.7±4.9^a	274.8±5.9^a
PRL (ng/ml)	20	38.4±1.3^a	40.2±1.4^a	36.9±1.2^a	37.7±1.3^a
	21	20.7±1.2^a	8.4±0.9^b	21.3±0.4^a	7.9±0.5^b
	25	22.4±1.2^a	8.8±0.8^b	18.4±0.7^a	8.2±0.6^b
	30	18.4±1.1^a	9.7±0.7^b	18.8±0.8^a	8.7±0.4^b
	31	19.5±1.1^a	9.4±0.6^b	8.7±0.8^b	8.4±0.8^b
	35	17.9±1.1^a	8.8±0.4^b	7.4±0.8^b	7.8±0.8^b
LH (ng/ml)	20	0.3±0.01^a	0.3±0.01^a	0.3±0.01^a	0.3±0.01^a
	21	0.3±0.01^b	1.4±0.01^a	0.3±0.01^b	1.4±0.03^a
	25	0.4±0.01^b	1.5±0.04^a	0.3±0.01^b	1.6±0.02^a
	30	0.5±0.01^b	1.6±0.03^a	0.4±0.01^b	1.6±0.04^a
	31	0.5±0.02^c	1.5±0.04^b	1.4±0.03^b	1.8±0.03^a
	35	0.4±0.02^c	1.6±0.04^{ab}	1.5±0.04^b	1.8±0.05^a

Least squares means in the same row with no common superscripts differ ($P<0.05$).
G1: Control. G2: Birds injected by 100 IU of PMSG at week 20 of age.
G3: Birds injected by 100 IU of PMSG at week 30 of age.
G4: Birds injected by 100 IU of PMSG at week 20 and 30 of age.

Obviously, LH level is affected by the environmental conditions, especially, photo stimulation period. Alternately, under the current study, significant increases in LH levels were observed in the absence of any environmental changes. Likewise, changes in the activity of the hypothalamo-hypophyseal-ovarian axis, due to PMSG injection, luteinzing hormone-releasing factor (LHRF) stimulates the release of LH, which in turn stimulates the secretion of steroids in the ovary (Bedecarrats *et al.*, (1997). These hormonal activities are concomitant with the maturation of the reproductive tract, which leads to augment laying of eggs.

In conclusion, PMSG subcuteonusly injection for laying ducks in level of 100 IU/bird/day for three subsequent days, resulted in significantly increases of egg number, egg weight, feed intake and improved ($P < 0.05$) the conversion of food to egg. Moreover, the injection reduced ($P < 0.05$) the blood serum level of T_3 and PRL., while it increased ($P < 0.05$) the blood serum concentrations of P_4, E_2 and LH.

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تأثير الحقن بالهرمونات المنشطة للغدد الجنسية (سيرم الفرس الحامل) على إنتاج البيض و بعض هرمونات سيرم الدم فى البط البكينى

أكرم حمدى

قسم الإنتاج الحيواني – كلية الزراعة – جامعة المنيا – جمهورية مصر العربية

استخدم فى هذه الدراسة عدد ٦٠ بطة بكينى عمر ٢٠ أسبوع متماثلة فى الوزن بمتوسط وزن ١٧٢١± ٢٨ جم. قسمت الطيور إلى أربعة مجاميع، قسمت كل مجموعة إلى ثلاث مكررات. كل خمس بطات تم إسكانها فى مساكن مفتوحة بمساحة ١,٥×١م استمرت التجربة من الأسبوع الـ ٢٠ إلى الأسبوع الـ ٣٥ من العمر فى الفترة من ديسمبر الى مارس وهى فترة تزايد النهار طبيعياً.

غذيت الطيور على عليقة تحتوى على ١٧% بروتين خام، ٢٨٠٠ كيلو كالورى طاقة ممثلة/كجم عليقة، ٣,٧% كالسيوم و٠,٦٦% فوسفور كلى.

عوملت المجموعات كالآتى: المجموعة الأولى استخدمت كمجموعة مقارنة حيث تم حقنها بالمحلول الفسيولوجى (٢مل محلول فسيولوجى/طائر يومياً لمدة ثلاثة ايام متتالية عند ألأسبوع ال ٢٠ و ال ٣٠)

المجموعة الثانية حقنت الطيور بسيرم دم الفرسة الحامل بمستوى ١٠٠ وحدة دولية لكل طائر تحت الجلد ولمدة ثلاثة أيام متتالية فى الأسبوع الـ ٢٠ من العمر. طيور المجموعة الثالثة عوملت بالحقن كالمجموعة الثانية ولكن تم الحقن فى الأسبوع الـ من العمر ٣٠. بينما فى المجموعة الرابعة حقنت الطيور بنفس المعدل (١٠٠ وحدة دولية/٣ أيام متتالية) فى الأسبوع الـ ٢٠ والأسبوع الـ ٣٠ من العمر.

أوضحت النتائج أن الحقن زاد معنويا من عدد ووزن البيض وكذلك من كمية الغذاء المستهلك وحسن من معدل تحويل الغذاء إلى بيض. بينما قلل الحقن معنويا من مستوى هرمون PRL, T_3 فى سيرم الدم وزاد معنويا من مستوى تركيز البروجستيرون و الأستروجينو الهرمون المسبب للتبويض فى سيرم الدم. وتوصى الدراسة بأن الحقن بسيرم دم الفرسة الحامل له تأثير إيجابي على زيادة إنتاج البيض فى الطيور