EFFECTS OF LIVE BODY WEIGHT AND LEPTIN CONCENTRATION ON IRAQI CHICKENS FERTILITY AND HATCHABILITY

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ABSTRACT

This Study was conducted out at the Ministry of Agriculture's Poultry Research Station/Animal Resources Department/Agricultural Research Center. To see how body weight (BW) and leptin hormone (LEP) levels in breeder blood affect fertility and hatchability. 140 Iraqi local laying chickens (120 females + 20 males) aged 28 weeks were used in the study. Following the numbering of females, the birds were grown in individual cages and dispersed sequentially on cages. The experiment was divided into three periods, each lasting 28 days, during which the breeder's live body weight was recorded and divided into two categories (greater than 1.5 kg and less than 1.5 kg), and blood samples were collected at the end of each period to determine the concentration of leptin hormone in the breeders' blood. For comparison between mothers' performance, hormone concentration is separated into three groups: high, medium, and low. The percentage of fertile eggs (FE), the percentage of hatched chicks from total eggs (HAT), the percentage of hatched chicks from fertile eggs (HAF), and the percentage of mortality (MO) all showed a significant increase ($p<0.05$), and a linear relationship was discovered between the studied traits and hormone concentration levels. Leptin arrived at the best predictive values that reflect reality by computing regression and correlation coefficients and using a hypothetical technique in estimating prediction results. This study concludes that body weight and leptin levels have unknown impacts on hatching fertility rates.

Keywords: *LEP*, LHB, Mortality. artificial insemination.

تأثير وزن الجسم الحي وتركيز هرمون اللبتين في نسب الخصوبة والفقس في الدجاج العراقي

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المستخلص

أجريت هذه الدراسة في محطة أبحاث الدواجن / قسم الثروة الحيوانية / دائرة البحوث الزراعية / وزارة الزراعة للمدة ولغاية ولتحديد علاقة تأثير تركيز هرمون اللبتين في دم الأمهات ومعدلات أوزانها في النسب الجنسية الاولية والثانوية. استعمل في التجربة 140 طير من امهات الدجاج البياض المحلي (120 انثى + 20 ذكرا) بعمر 28 اسبوعا، جهزت من محطة ابحاث الدواجن، ربيت الطيور في اقفاص فردية، ووزعت الطيور بالتسلسل على الاقفاص بعد ترقيم الاناث. قسمت التجربة على ثلاثة مدد كل مدة 28 يوماً سجل في أثنائها الوزن الحي للأمهات وقسمت على أساسه على فئتين (أكبر من 1.5 كغم وأصغر من 1.5 كغم)، ومن ثم حسب المعدل العام لكل صفة مدروسة. وسحب الدم عند نهاية كل مدة لقياس تركيز هرمون اللبتين في دم الأمهات، ومن ثم وزعت الطيور على اساس تركيز الهرمون الى ثلاث مجموعات: المرتفعة، المتوسطة، المنخفضة، وحسب التداخل بين وزن الجسم وتركيز اللبتين للمقارنة فيما بين اداء الامهات. أشارت النتائج الى وجود ارتفاع معنوي ($p<0.05$) في النسبة المئوية للبيض الخصب، النسبة المئوية للأفراخ الفاقسة من البيض الكلي، النسبة المئوية للأفراخ الفاقسة من البيض الخصب، النسبة المئوية للأجنة الهالكة، ومن ذلك تبينت علاقة خطية فيما بين الصفات المدروسة ومستويات تركيز هرمون اللبتين، بحساب معامل الانحدار والارتباط، واعتماد المنهج الفرضي في تقدير نتائج التنبؤ للوصول الى أفضل القيم التنبؤية التي تقرب من الواقع. ونستنتج من هذه الدارسة أن لوزن الجسم وتركيز هرمون اللبتين تأثيرات في نسب الخصوبة الفقس، غير محددة الاتجاه.

كلمات مفتاحية: مستقبلات اللبتين، الهرمون اللوتيني، الأجنة الهالكة، التلقيح الاصطناعي.

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INTRODUCTION

Leptin is a peptide hormone that regulates and balances energy levels, as well as appetite and food metabolism (11). According to (1), leptin is important in regulating the functions of the reproductive axis in birds because it is a catalyst in stimulating the development of the reproductive system and sexual puberty in chickens, and its concentration rises after sexual puberty, and it also stimulates GnRH and the release of LH and FSH hormones (6).

GnRH secretion can be regulated by leptin; however, leptin receptors cannot be expressed in the neurons that produce GnRH. Leptin has an indirect effect on GnRH secretion (20). In addition, there is evidence that leptin raises levels of sexual hormones, controls the onset of puberty, and prevents apoptosis in the three major ovarian follicles (17, 18). In addition, leptin restores the natural order of the ovarian follicle hierarchy and corrects any disruptions in it (8). While doing so, leptin also boosts the synthesis of ovarian steroidogenesis (22), which in turn helps granulosa cells perform their tasks better (12). Egg production rates improved as a direct consequence of increased estrogen release, which was boosted by leptin (23). In their study on birds, (21) came to the conclusion that the hormone leptin has a significant role in both the process of ovarian growth and development as well as the process of folliculogenesis. In light of the fact that the primary effect of leptin is seen in its regulatory functions of energy metabolism, body weight, and a number of reproductive roles, the aim of this study was to investigate the influence of body weight and leptin on fertility and hatchability features.

MATERIALS AND METHODS

This study was conducted at the Poultry Research Station / Department of Livestock / Agricultural Research Department / Ministry of Agriculture for the period up to and to determine the relationship of the effect of the concentration of leptin hormone in the blood of mothers and their weight rates on the primary and secondary sex ratios.

In the experiment, 140 domestic laying hens (120 females + 20 males) at 28 weeks of age were used, which were prepared from the poultry research station. The data were recorded in three periods, each period is 28 days, and then according to the general average for each trait studied. Blood was drawn 4-5 hours before the default date of ovulation at the end of each period to measure the concentration of leptin hormone in the mothers' blood, and then the birds were distributed on the basis of the hormone concentration into three groups: high, medium, and low, to compare between the mothers' performance.

Fertility and hatchability ratios

Semen collection from roosters, as indicated by (2, 5), as the collection method requires the presence of two people, the first holds the bird and puts its head back and the assembly forward with both hands. As for the second person, he massages the dorsal ventral region (the back of the bird to the tail hall) quietly, until the erection of the papilla and the flow of semen, then collecting the semen using a plastic box, then diluting the semen with a normal saline 9% (3, 4)

Fertility percentage

The hatching process was carried out three times throughout the duration of the experiment, with one hatching for each 28 days of the experiment. Fertilized eggs were collected in the five days following the second day of the insemination process, and the eggs were stored in the station's hatching egg store at a temperature of 15.5 °C, and the eggs were incubated in A hatcher of the Belgian type (Petersime), belonging to the hatchery of the poultry research station of the Agricultural Research Department / Abu Ghraib.

- After completing the hatching process, record the number of dead embryos by cracking the non-hatched eggs and placing them in plastic boxes and keeping them by freezing for the purpose of conducting analyzes, and then calculating the fertility rate and the percentage of dead embryos according to the following two equations:

$$\mathbf{FE}\ \% = \frac{\mathbf{fertilized\ eggs}}{\mathbf{total\ eggs}} \times \mathbf{100}$$

$$\mathbf{MO}\ \% = \frac{\mathbf{dead\ embryos}}{\mathbf{fertilized\ eggs}} \times \mathbf{100}$$

hatching percentage:

The hatching percentage was calculated after calculating the number of hatched chicks, according to the following two equations:

$$\textbf{HAT}\ \% = \frac{\textbf{hatching chicks}}{\textbf{total eggs}} \times \mathbf{100}$$

$$\textbf{HAF}\ \% = \frac{\textbf{hatching chicks}}{\textbf{fertilized eggs}} \times \mathbf{100}$$

Blood collection:

Blood was collected from all females 4-5 hours before the expected date of ovulation at eight o'clock in the morning, at the end of each period, from the cutaneous ulnar vein, or what is known as the pterygoid or humeral vein, using a 5 ml syringe equipped with a 25-gauge needle, as mentioned by (10). If the bird is placed lying on its back and one of the wings is extended, the feathers covering the area of the ulnar vein are removed quickly to avoid causing pain to the bird, and the skin is wiped with alcohol to increase the visibility of the vein. Then the blood is withdrawn from the vein with the syringe equipped with a 25-gauge needle, after puncturing the vein towards the top, and the blood is withdrawn by creating a decompression (photo) then the blood is emptied after removing the needle from the plastic syringe into glass tubes containing a gel capacity of 10 ml, and these tubes are placed in Centrifuge at 6000 rpm for 10 minutes, to separate the serum from the cell fraction. After samples are transferred to plastic tubes of 5 ml and kept at a temperature of 20-C until tests are performed.

ELISA test:

Third: Examination of the concentration of leptin hormone in the blood serum:

The concentration of leptin hormone in the blood serum was measured using a special kit manufactured by (Bioassay Technology Laboratory) (Chicken Leptin Elisa Kit NO E0026Ch) in the laboratories of Al-Fadil Foundation in Babylon Governorate. The ELISA assay is a modern immunoassay used to titrate any antigen with a high accuracy of up to 0.0005 microml. The basis of this test is the association of antibodies directed against a specific antigen in the serum sample to be examined with special antigens attached to the microtiter plate.) prepared with the kit of standard solutions, and after washing to remove unwanted substances, the conjugate is added, which contains antibodies directed against the first antibody to bind to it and tagged with Horse Radish peroxidase (HRP), and after the second washing, the sweetener is added Or the substrate, which is a colorless substance that is transformed by the aforementioned enzyme into a brown substance that can be measured by means of a spectrophotometer for the ELISA test.

First: The step of preparing the Serum Dilution Dish:1. A Serum Dilution Plate, which is a dish consisting of 96 antigen-free holes, is prepared by adding 0.3 ml of Dilution Buffer to all the holes of the dish except for the last three holes.

2. Add 0.006ml of normal control serum prepared with the kit to the first three holes of the plate.

3. 0.006 ml of blood serum samples are added to each pit of the dish except for the first and last three pits as control. Leave the dish for 5 minutes under room temperature.

Secondly, the step of preparing the micro-calibration dish:

1- 0.05 ml of buffer solution is added to each pit of the microtiter plate, which consists of 96 U-shaped holes containing specific antigens, especially for the disease to be examined.

2- . 0.05 ml of the prepared positive control serum is added

3- With Kit)) to the last three holes of the dish.

4- 0.05 ml is transferred from each hole of the serum dilution dish pit to the corresponding pit of the micro-titration dish.

5- Leave the plate for 30 minutes at room temperature.

Third: The washing step:

1. Empty all the contents of the pits of the micro-calibration dish into a container containing a disinfectant liquid.

2. Add 0.3 ml of washing solution to each pit of the dish and wait for 3 minutes, and this process is repeated twice.

Fourth: The step of adding the coupling and the substrate:

1. 0.1 mm of conjugate is added to each pit of the micro-titration dish and left

The dish is under room temperature for 30 minutes, after which we carry out the washing step as mentioned previously.

2. 0.1 ml of the substrate solution is added to each pit of the micro-calibration dish and incubated under room temperature for 15 minutes, after which the washing step is performed as mentioned previously

Fifth: The step of stopping the reaction:

1. 0.1 ml of a stop solution is added to each pit of the dish.

2. The samples are read by a spectrophotometer (AWARENESS of American origin) for the examination at a wavelength of 405-410 nm.

RESULTS AND DISCUSSION

Percentage of fertile eggs (FE%):

Table 1 shows that there was a significant increase ($P<0.05$) in the FE% eggs produced by chickens in group A2 throughout the first period of the trial when compared with group A1. Although there was not a significant difference found between groups A1 and A2 in the FE% during the second, third, or overall average of the study, did find that there was a difference. Table 1 shows that there was a significant increase ($P<0.05$) in the FE% in the medium group (B2) of leptin concentration when compared with the high group of the same hormone concentration, both during the first and second periods of the study.

On the other hand, there were no significant differences found between the B1 groups as well as B2 and B3. The FE% produced during the third period did not significantly differ across the three groups, nor did it significantly deviate from the overall average. Table 1 reveals that during the first and second periods, there was a significant increase ($P<0.05$) in the percentage of fertile eggs in A2B2 compared with A1B3, which did not show any significant differences in the same trait compared with the other four Interactions. This is something that was noticed.

Percentage of hatched chicks out of total eggs (HAT%):

The results of the first periods of the experiment are presented in Table 2, which reveals that the HAT% was significantly increase in group A2 compared to group A1 ($P<0.05$). While it is true that there are no significant differences between groups A1 and A2 during the second and third periods and the overall average, it should be noted that there are also no disparities between these groups and the general average. In the second period, it was shown that there was a significant increase ($P<0.01$) in the HAT% in the group with the intermediate concentration of leptin hormone in the blood (B2), in comparison with groups B1 and B3. During the first and third periods, as well as when compared to the overall average, there were no significant differences found between the three groups.

Table 1. Effect of body weight and concentration of leptin hormone and the interaction between them on the percentage of fertile eggs of local Iraqi chickens

Affecting factors			Percentage of fertile eggs (FE%)							
			1st Period	sample size	2nd Period	sample size	3rd Period	sample size	overall average	sample size
BW (A)	less than1.5 Kg (A_1)		84.04±1.39 B	66	79.15±1.52	67	84.44±1.28	66	83.49±0.77	65
	Greater than1.5kg (A_2)		87.96 ± 1.28 A	53	82.72 ± 1.64	52	83.46 ± 1.35	53	83.52 ± 0.89	54
	Significant		0.05	119	N.S	119	N.S	119	N.S	119
concentration of leptin (B)	Low (B_1)		86.07±1.59 AB	50	80.57±2.42 AB	32	84.95±1.77	34	83.47±1.91	28
	Medium (B_2)		88.33±1.40 A	42	83.84±1.49 A	49	82.56±1.43	54	82.94±0.76	60
	High (B_3)		81.30±2.10 B	27	76.80±2.06 B	38	85.48±1.66	31	84.82±0.96	31
	Significant		0.05	119	0.05	119	N.S	119	N,S	119
interactions AB	less than 1.5 Kg (A_1)	Low (B_1)	85.90±2.55 AB	26	81.86±3.33 AB	17	85.70±2.43	19	84.41±1.95 A	15
		Medium (B_2)	86.01 ± 1.78 AB	23	81.90 ± 1.74 AB	28	82.17 ± 2.11	30	83.03 ± 0.95 A	31
		High (B_3)	78.53±2.78 B	17	73.56±3.07 B	22	87.06±1.83	17	83.84±1.52 A	11
	Greater than1.5kg (A_2)	Low (B_1)	86.25±1.85 AB	24	79.11±3.57 AB	15	84.00±2.60	15	69.44±0.25 B	12
		Medium (B_2)	91.14 ± 2.18 A	19	86.43 ± 2.56 A	21	83.06 ± 1.85	24	82.83 ± 1.22 A	30
		High (B_3)	86.00±2.99 AB	10	81.25±2.38 AB	16	83.57±2.91	14	85.36±1.23 A	20
	Significant		0.05	119	0.05	119	N.S	119	0.05	119

The varying letters within the same column indicate significant differences between the means. N.S: There are no significant differences.

There was also a significant increase (P<0.05) in the HAT% of A2B2 and A2B3 in comparison to A1B3, and these interactions did not show any significant differences in comparison to A1B1, A1B2, and A2B1, during the first period. During the second period, all of the A1B2, A2B2, and A2B3 interactions showed a significant increase (P<0.01) in the HAT% in comparison with the A1B1, A1B3, and A1B1 interactions. In contrast to A1B3, both A1B1 and A2B1 demonstrated a significant increase (P<0.01) in the HAT% successfully developed into chicks. During the overall average, it was found that the HAT% was significantly increase in A1B3 when compared to A2B1. On the other hand, the differences between the interactions A1B3 and A2B1 and the other interactions were not significant. During the third time period, the HAT% did not demonstrate any significant changes across any of the six different interactions.

Percentage of hatched chicks from fertile eggs (HAF%): According to Table 3, it was discovered that during the third period: a significant increase (P<0.05) was seen in the HAF%, in chickens of the big size group A2 in comparison to A1. During the first and second periods, as well as when compared to the overall average for the study, neither A1 nor A2 showed any significant differences.

Table 3 shows that there was a significant increase (P<0.05) in the HAF% in B3 during the first period, in comparison to B1. However, there were no significant differences between B2 and B1 and B3, respectively. During the third period, both B1 and B2 showed a significant increase in the percentage of successfully hatched chicks that originated from fertile eggs. During the second period, there was not a significant difference between B1, B2, or B3 and the overall average. In contrast, there was a significant difference between the overall average. Except for the interaction A1B3, which did not show significant differences with either the interaction A2B3 or the other interactions, it was noticed that the interactions A2B3 recorded a significant increase (P<0.05) in the HAF% compared with the other interactions. This was observed during the first period. Significant differences (P<0.05) were also discovered in favor of A1B1, A1B2, and A2B3 in comparison with A1B3 and A2B1 in this study. During the second phase of the interactions, it was found that there were no statistically significant differences between A2B2 and the other interactions in terms of the HAF%, When compared to the other interactions, A2B2 showed a significant decrease in the incidence of HAF% during the third period. While A1B3 and A2B1 demonstrated a significant increase in the overall average of the HAF% chicks when contrasted with the results of the other interactions, these two interactions lagged far behind the others.

Mortality (MO%):

Table 4 shows that during the third period of the trial, there was a considerable decrease in the MO% that were classified as belonging to category A2 compared to that of category A1. There were no significant differences between the performance of the two groups throughout the first and second period of the experiment, as well as the overall mean for the whole thing. Table 4 study results make it abundantly clear that Group B1 experienced a significant increase (P<0.05) in the MO% when compared with Group B3, whereas Group B2 did not demonstrate a significant difference when compared with Groups B1 and B3 during the second term. During the third period, it was discovered that B1 and B2 showed a significant decrease (P<0.05) in the relative weight of the stillborn fetuses when compared with B3. This was in contrast to B3, which showed no such change.

Table 2. Effect of body weight and leptin hormone concentration and the interaction between them on the average percentage of hatched chicks from total eggs of local Iraqi chickens

Affecting factors			The percentage of hatched chicks from the total eggs (HAT%)							
			1st Period	sample size	2nd Period	sample size	3rd Period	sample size	overall average	sample size
BW (A)	less than1.5 Kg (A_1)		72.53± 1.56 B	66	69.15±1.57	67	71.54±1.66	66	72.57±0.77	65
	Greater than1.5kg (A_2)		77.45± 1.54 A	53	69.97±1.73	52	74.34±1.64	53	72.06±0.88	54
Significant			0.05	119	N.S	119	N.S	119	N.S	119
concentration of leptin (B)	Low (B_1)		73.43± 1.82	50	68.18±2.57 B	32	75.10±2.25	34	71.56±1.78	28
	Medium (B_2)		76.59± 1.71	42	74.05±1.61 A	49	72.99±1.73	54	72.21±0.76	60
	High (B_3)		74.20± 2.36	27	64.78±1.97 B	38	69.89±2.25	31	73.05±1.01	31
Significant			N.S	119	0.01	119	N.S	119	N.S	119
interactions AB	less than 1.5Kg (A_1)	Low (B_1)	72.12± 2.56 AB	26	72.84±3.74 B	17	76.32±3.31	19	71.70±1.90 AB	15
		Medium (B_2)	74.93± 2.54 AB	23	75.24±1.99 A	28	71.22±2.43	30	72.19±1.01 AB	31
		High (B_3)	69.90± 3.12 B	17	58.56±2.36 C	22	66.76±2.94	17	75.10±1.15 A	11
	Greater than1.5kg (A_2)	Low (B_1)	74.86± 2.59 AB	24	62.89±3.33 B	15	73.56±2.95	15	69.44±1.04 B	12
		Medium (B_2)	78.60± 2.21 A	19	72.46±2.65 A	21	75.21±2.43	24	72.22±1.16 AB	30
		High (B_3)	81.50± 3.19 A	10	73.33±2.99 A	16	73.69±3.43	14	71.92±1.41 AB	20
Significant			0.05	119	0.01	119	N.S	119	0.05	119

The varying letters within the same column indicate significant differences between the means. N.S: There are no significant differences.

Table 3. Effect of body weight and leptin hormone concentration and the interaction between them on the average percentage of hatched chicks from fertile eggs of local Iraqi chickens

Affecting factors		The percentage of hatched chicks from fertile eggs (HAF%)							
		1st Period	sample size	2nd Period	sample size	3rd Period	sample size	overall average	sample size
BW (A)	less than1.5 Kg (A_1)	87.22±1.36	66	88.53±1.33	67	84.75±1.45 B	66	87.48±0.80	65
	Greater than1.5kg (A_2)	88.14 1.22 ±	53	86.41 1.63 ±	52	88.65 1.29 A ±	53	86.95 0.83 ±	54
	Significant	N.S	119	N.S	119	0.05	119	N.S	119
concentration of leptin (B)	Low (B_1)	86.10±1.52 B	50	86.20±2.21	32	88.04±1.91 A	34	86.91±1.90	28
	Medium (B_2)	87.02±1.45 AB	42	89.15±1.36	49	88.06±1.38 A	54	87.52±0.72	60
	High (B_3)	91.42±1.88 A	27	86.80±1.98	38	82.04±2.04 B	31	86.77±1.06	31
	Significant	0.05	119	N.S	119	0.05	119	N.S	119
interactions AB	less than 1.5Kg (A_1) Low (B_1)	86.03±2.17 B	26	89.22±2.91 A	17	88.86±2.76 A	19	86.04±1.96 B	15
	less than 1.5Kg (A_1) Medium (B_2)	87.03±2.26 B	23	91.96±1.59 A	28	87.00±2.56 A	30	87.26±1.04 B	31
	less than 1.5Kg (A_1) High (B_3)	89.31±2.75 AB	17	83.64±2.62 B	22	86.39±1.91 A	17	90.20±1.18 A	11
	Greater than1.5 kg (A_2) Low (B_1)	86.18±2.14 B	24	82.78±3.32 B	15	90.14±1.96 A	15	91.56±0.25 A	12
	Greater than1.5 kg (A_2) Medium (B_2)	87.02±1.71 B	19	85.40±2.29 AB	21	77.25±3.09 B	24	87.81±1.00 B	30
	Greater than1.5 kg (A_2) High (B_3)	95.00±1.86 A	10	91.15±2.96 A	16	87.86±2.26 A	14	84.89±1.45 B	20
	Significant	0.05	119	0.05	119	0.05	119	0.05	119

The varying letters within the same column indicate significant differences between the means. N.S: There are no significant differences.

Table 4. Effect of body weight and leptin hormone concentration and the interaction between them on the average percentage of dead fetuses in local Iraqi chickens

Affecting factors		Percentage of dead fetuses (%): 1st Period	sample size	2nd Period	sample size	3rd Period	sample size	overall average	sample size
BW (A)	less than1.5 Kg (A_1)	12.78±1.36	66	11.47±1.33	67	15.25±1.45 A	66	12.52±0.80	65
	Greater than1.5kg (A_2)	11.86±1.22	53	13.59±1.63	52	11.35±1.29 B	53	13.05±0.83	54
Significant		N.S	119	N.S	119	0.05	119	N.S	119
concentration of leptin (B)	Low (B_1)	13.90±1.52 A	50	13.80±2.21	32	11.96±1.91 B	34	13.09±1.90	28
	Medium (B_2)	12.98±1.45 AB	42	10.85±1.36	49	11.94±1.38 B	54	12.48±0.72	60
	High (B_3)	8.58 ± 1.88 B	27	13.20 ± 1.98	38	17.96 ± 2.04 A	31	13.23 ± 1.06	31
Significant		0.05	119	N.S	119	0.05	119	N.S	119
interactions AB	less than 1.5Kg (A_1) Low (B_1)	13.97±2.17 A	26	10.78±2.91 B	17	11.14±2.76 B	19	13.96±1.96 A	15
	less than 1.5Kg (A_1) Medium (B_2)	12.97±2.26 A	23	8.04±1.59 B	28	13.61±1.91 B	30	12.74±1.04 A	31
	less than 1.5Kg (A_1) High (B_3)	10.69±2.75 AB	17	16.36±2.62 A	22	22.75±3.09 A	17	9.80±1.18 B	11
	Greater than1.5 kg (A_2) Low (B_1)	13.82±2.14 A	24	17.22±3.32 A	15	13.00±2.56 B	15	8.44±0.44 B	12
	Greater than1.5 kg (A_2) Medium (B_2)	12.98±1.71 A	19	14.60±2.29 AB	21	9.86±1.96 B	24	12.19±1.00 A	30
	Greater than1.5 kg (A_2) High (B_3)	5.00±1.86 B	10	8.85±2.96 B	16	12.14±2.26 B	14	15.11±1.45 A	20
Significant		0.05	119	0.05	119	N.S	119	0.05	119

The varying letters within the same column indicate significant differences between the means. N.S: There are no significant differences.

In comparison to the other interactions, A2B3 shown a significant decrease (P<0.05) in the MO% at the first term. On the other hand, A1B3 did not show any significant differences when compared to the other interactions. In the second period, it was discovered that A1B1, A1B2, and A2B3 recorded a significant decrease (P<0.05) in the MO% compared with A1B3 and A2B1, whereas A2B2 did not show significant differences compared with the other interactions. A1B3 and A2B1 did not show significant differences compared with the other interactions. At the end of the third period, it was discovered that A1B3 had a much lower MO% compared to the other therapies. This reduction was significant (P<0.05) When compared to the other four interactions, the MO% was found to be significantly lower for A1B3 and A2B1 when the overall average was calculated, this reduction was significant (P<0.05).

1) Correlation coefficient

It is clear from looking at Table 5 that a significant (P<0.01) was negative correlation coefficient between the BW and LPE, whereas the correlation coefficient of BW with FE% and HAT% significant (P<0.01) was positive. Although there was no significant correlation between LPE and the other traits, this does not mean that there is no correlation at all. With a significant negative correlation between FE and HAF, a significant positive correlation between FE, HAT, and MO, and a significant positive correlation between HAT and MO. In addition, a significant (P0.01) positive correlation can be seen between HAF and MO.

Table 5. Correlation coefficients for the productive and physiological characteristics of local Iraqi chickens during the first period.

Qualities	correlation coefficients				
	BW	LEP	FE%	HAT%	HAF%
LEP	-0.106*				
FE%	0.187**	0.009			
HAT%	0.150**	0.067	0.683**		
HAF%	0.002	0.082	-0.143**	0.610**	
Mo%	-0.002	-0.082	0.143**	-0.610**	-1.000**

***; ** The level of significance is 0.05 and 0.01 respectively**

2) Regression coefficient

Table 6 shows that the regression of BW and LPE on BW and LPE and (multiple regression) is a significant regression (P<0.01) and positive, and that the multiple regression HAT%, HAF%, and MO% on BW and LPE were significant (P<0.05) and positive. These findings can be seen by looking at the table.

Table 6. Multiple regression coefficients for productive and physiological traits on live body weight and leptin concentration in the blood of local Iraqi chickens during the third period.

Traits	Straight-line equation (expectation)	Significant	R-squared (R^2)
BW	$Y^ = 1.048 + 0.289 (X_1) + 0.020 (X_2)$	0.001	0.674
LPE	$Y^ = 0.141 - 0.050 (X_1) + 0.553 (X_2)$	0.001	0.690
FE%	$Y^ = 85.007 - 0.988 (X_1) + 0.216 (X_2)$	N.S	0.001
HAT%	$Y^ = 73.846 + 2.829 (X_1) - 2.607 (X_2)$	0.045	0.011
HAF%	$Y^ = 86.634 + 3.934 (X_1) - 2.955 (X_2)$	0.027	0.024
MO%	$Y^ = 13.366 - 3.934 (X_1) + 2.955 (X_2)$	0.027	0.024

The growth and development of the reproductive systems in the body are dependent, to a large extent, on the metabolic roles that are regulated by leptin. This hormone is involved for regulating food intake, which in turn controls energy expenditure and the amount of fat stored in the body (7). This contributes to the regulation of body weight, as leptin is raised in birds with dense fat in and around the abdomen. This also explains the association between body weight and leptin concentration, which was the basis for designing the study in the first place. The increase in the size of the fatty layers in the belly is to blame for the rise in overall body weight seen in chickens used for egg production (15). As leptin works to stimulate the neurons secreting GnRH hormone from the hypothalamus, in addition to providing direct stimulation to the hypothalamic pituitary-gonadal (HPGA) axis, it is possible that the primary roles played by leptin in the functions of the hypothalamic-pituitary-gonadal (HPGA) axis may be responsible for the improvement

in the characteristics of fertility and hatching. LH and FSH are both secreted by the distal portion of the pituitary gland (16), and this region also has an inhibitory connection with androgens (9). Which may suggest that the process of the metabolism of androgens to estrogen occurs by the effect of leptin, as estrogen works on the growth, maturity, and development of ovarian follicles, and then increases the ovulation-oviposition cycle, as estrogen is a response to the mechanism of action of the hypothalamic-pituitary-gonadal axis. In addition, leptin works on the growth, maturity, and development of ovarian follicle (19). Because of this, both fertility and hatchability see improvements as a result (Tables 1, 2, 3, 4). The results are summarized in Table 5, which shows that the correlations with leptin concentration are not significant but are significant with body weight. When reference to Table 6, it is easy to see that the investigated characteristics can be projected through their multiple regression on body weight and leptin concentration. However, this does not indicate that leptin does not play a significant role in the same characteristics, as the first effect of leptin appears in body weight (13), as leptin controls appetite and stimulates thyroid hormone, and since greater levels are dependent on the amount of feed that is ingested (14). It is possible to predict fertility and hatchability characteristics by using multiple regression of body weight and leptin concentration as the independent factors. This conclusion can be drawn from the previous information. And that both factors are crucial in the reproductive performance of hens, but that further research on both factors is still required.

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