

EFFECT OF DIFFERENT ESTRUS SYNCHRONIZATION ON SERUM E₂, P₄, FSH AND LH DURING DIFFERENT ESTRUS PERIODS AND PREGNANCY IN EWES

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ABSTRACT

This research was designed to know the effect of different estrus synchronization protocols on Serum E, P, FSH, and LH during different estrus periods and pregnancy in Karadi and Arabi ewes. The study was carried out by using 30 ewes for each breed at 2-4 years. Ewes from each breed were equally and randomly distributed into three groups. The first group was treated with vaginal sponges saturated with (60 mg) of Medroxy progesterone acetate within a period of (14) fourteen days (T1). The second group was administrated with the MAP injected with a dose of 300 IU / head of the hCG (T2). The third group was treated with MAP treatment, and injected at a dose of 75 µg /head of GnRH (T3). The results of this analysis showed a significant increase in the estrogen concentration in the different reproductive stages; the significant level of estrogen in the estrus phase (60.96) pg/ml. The concentration of progesterone differed significantly in the different reproductive stages, with the highest concentration in the pregnancy period (16.33) pg/ml. In comparison, the FSH concentration differed significantly in the different reproductive stages with the highest concentration in the estrus period (3.243) mIU/ml. the highest significant level of serum LH is in the estrus period (5.230) mIU/ml. Thus, the study is concluded that hormones and phases may be effective in inducing follicular growth and ovulation and finally may increase pregnancy rate and production .

Keywords: Estrous synchronization; hormonal regulation, Karadi, Arabi.

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تأثير طرق توحيد الشبق المختلفة على هرمون الاستروجين ، البروجسترون، FSH و LH خلال فترات الشبق والحمل المختلفة في النعاج

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المستخلص

أجريت هذه الدراسة لمعرفة تأثير طرق مزامنة الشبق المختلفة على هرمون الاستروجين، البروجسترون، FSH و LH في المصل خلال فترات الشبق والحمل المختلفة في نعاج الكرادية والعربية. استخدمت 30 نعجة لكل سلالة بعمر 2-4 سنوات. تم توزيع النعاج من كل سلالة بالتساوي والعشوائية إلى ثلاث مجموعات. المجموعة الأولى عولجت بإسفنجات مهبلية مشبعة بـ (60 ملغ) ميدروكسي بروجيسترون (MAP) لفترة (14) يوماً (T1). المجموعة الثانية عولجت بـ MAP وتم حقنها بجرعة 300 وحدة دولية من هرمون hCG (T2). المجموعة الثالثة عولجت بـ الاسفنجات (MAP) تم حقنها بجرعة 75 µg / رأس من هرمون GnRH (T3). أظهرت نتيجة الدراسة زيادة كبيرة في تركيز هرمون الاستروجين في المراحل التناسلية المختلفة؛ وصلت إلى أعلى مستوى في مرحلة الشبق (60.96) pg / ml. اختلف تركيز هرمون البروجسترون بشكل معنوي في المراحل التناسلية المختلفة، مع أعلى تركيز في فترة الحمل (16.33) pg / ml. اختلف تركيز FSH بشكل كبير في المراحل التناسلية المختلفة بأعلى تركيز في فترة الشبق (3.243) وحدة دولية / مل. أعلى مستوى معنوي لهرمون LH كان في فترة الشبق (5.230) وحدة دولية / مل. وبالتالي، فإن الدراسة خلصت إلى أن الهرمونات والمراحل قد تكون فعالة في حث النمو الحويصلات والإباضة وأخيراً قد يزيد من معدل الحمل والإنتاج.

الكلمات المفتاحية: توحيد الشبق؛ التنظيم الهرموني، الكرادية، العربي.

INTRODUCTION

The majority of sheep reproduction management procedures focus on inducing and synchronizing estrus and ovulation to allow for out-of-season and/or synchronized lambing. (17). Studies in sheep considered management practices to improve the productive efficiency of herds in a technical and economical way, in which it is intended to eliminate the pharmacological manipulation of animals (25). These methodologies are founded on understanding reproductive events, socio-sexual factors, and the effects of nutrition (19,34) because at present, reproductive management protocols are based on the application of exogenous hormones that simulate the action of a corpus luteum (CL), such as progestogens (P4); and others manage to eliminate it, to induce a follicular phase and ovulation, such as prostaglandins (PGF2 α) (1). hCG and its receptor, LH/GCR, are expressed in numerous regions of the reproductive tract, both in gonadal and extragonadal tissues, stimulating oocyte maturation, fertilization, implantation, and early embryo development, according to a review of the worldwide literature and research investigations. Furthermore, hCG appears to have a role in solid organ transplantation as an anti-rejection substance (38). Furthermore, hCG has also been preferred over Human Menopausal Gonadotrophin because to its increased availability and lower cost (hMG). As a result, hCG was previously used to induce ovulation in sheep during the anoestrus season (20). The primary neuropeptide controlling reproductive function in all vertebrate species is gonadotropin-releasing hormone (GnRH) (20, 42). Ovarian steroids secreted by mature ovarian follicles control a pulsatile pattern of GnRH release from the hypothalamus, which stimulates a preovulatory production of luteinizing hormone (LH) by the anterior pituitary gland in females of spontaneously ovulating animals, such as sheep (6). The GnRH injection causes an LH peak in a tiny period of time and reduces the ovulation period (13). The use of gonadotropin-releasing hormone (GnRH) or the male effect in PGF2 α -based protocols was reported previously (27,28). Human chorionic gonadotropin (hCG)

and gonadotropin-releasing hormone (GnRH) were given to ewes of several breeds (Akkaraman, fat-tailed, Afshari, and Booroola-Merino crossbred) to improve reproductive performance (conception, lambing, twinning rate, and litter size), (2,5,12,29). In Afshari \times Booroola-Merino crossbred ewes, post-mating hCG-treated groups had a higher plasma concentration. (29). Injection of GnRH on the day of oestrus or at the time of mating and 7 or 9 days later increased serum progesterone concentration. The study's focus is to know the effect of using different estrus synchronization protocols on serum Estrogen, Progesterone, FSH, and LH during different estrus periods and pregnancy in Karadi and Arabi ewes in Erbil local.

MATERIALS AND METHODS

Animals and experimental design

The experiment was carried out in a private field in Trpespyan farm / Erbil governorate/ Kurdistan region/Iraq, between 20/7/2020 and 21/1/2021. The experiment included sixty Karadi and Arabi ewes (30 for each breed) aged 2-4 years old and live body weight 50 kg \pm 5 for Karadi and Arabi ewes, respectively. Each breed was randomly subdivided into three groups (10 ewes in each group), and Experimental animals were subjected to the same administrative and nutritional conditions prevailing in the field, the concentrated fodder 600 g / head/day) was provided into main two meals, morning and evening, with molds of mineral salts, suspended inside the barn, provided with the provision of free straw permanently, providing water and permitting For animals to graze in the morning and evening. In addition to that, ewes will weekly administrate with the animal's vitamin (AD3) to compensate for the lack of vitamins during the experiment. The first group (T1): - (10) ewes. Treated with vaginal sponges saturated with (60 mg) of Medroxyprogesterone acetate (MAP) will be inserted for a period of fourteen days. The second group (T2): - The second group: - (10) ewes will administrate with the same first treatment, and after withdrawal the sponge, They were injected with a dose of 300 IU per head of the Human Chorionic Gonadotropin (hCG). The third group: - (10) ewes will administrate with the same first treatment and after the sponge withdrawal was

injected at a dose of 75 micrograms/head of (GnRH).

Blood sampling and hormonal analysis

Blood samples were collected from ewes via Jugular vein-puncture using nonheparinized vacutainer tubes during various reproductive phases at 11.30 a.m. Blood samples were centrifuged (at 3000 rpm for 15 min), and plasma was separated and stored at -20°C until analysis. The biochemical constituents hormones such as estrogen, progesterone, LH, and FSH were measured by double-antibody enzyme immune-assay) using Modular Analytics E170, cobas e 602 analyzers. (Strasse 116, D-68305 Mannheim-USA) plates according to the method of (21) during various phases like Luteal, estrus, pregnant, and after parturition in ewes were analyzed.

Estrus detection and mating

Two fertile Karadi and Arabi rams were introduced to the ewes in each experimental group (one ram per 5 ewes) for estrus detection, and Rams were mating; starting at the sponge withdrawal day allowed to rotate among different ewes groups to avoid sire/group confounding effect. Painted-breast fertile rams were introduced to ewes for five days. Ewes with marked rumps were considered to be mated.

Statistical analysis

General Linear Model (GLM) within the statistical program SAS, 2005(33) was used to analyze the collected data to diagnose the significant effects of the available factors affecting the studied traits, and the experiment was designed as factorial-CRD. Duncan multiple range tests (11) were used to test the differences between the subclasses of each factor.

RESULTS AND DISCUSSION

Estrogen concentration

From table 1, it could be concluded that estrogen concentration differed significantly in the different reproductive stages; In the current study, the estrus phase contains a significant level of serum estrogen (60.96 pg/ml) with significant differences ($p \leq 0.01$) among for the three stages (37.73, 35.16 and 12.53) pg/ml respectively. Sangeetha and Rameshkumar (30) reported similar to our findings that a high level of serum estrogen is present in the ovulatory phase. In the luteal phase, the

highest concentration of estrogen was seen in group T3 (38.30 pg/ml) with significant ($p \leq 0.01$) differences in comparison with other groups T1 (37.20 pg/ml) and T2 (37.70 pg/ml). That may be related to the fact that the major regulator of the reproductive axis is gonadotropin-releasing hormone (GnRH). Its pulsatile secretion influences both endocrine function and gamete maturation in the gonads by determining the pattern of secretion of the gonadotropins follicle-stimulating hormone and luteinizing hormone. and stimulates the secretion of estrogen hormone (24). This result came in agreement with previous studies (3) Furthermore, the type of breed hadn't significant effect on estrogen concentration in general, with its arithmetic increase in the Karadi in all stages, there are differences between breeds in the Karadi breed, estrogen concentration increased non significantly in the estrus (61.07 pg/ml) in comparison with Arabi breeds. this difference may be attributed to the variation in the genetic abilities of the individuals of the two breeds (8) In the estrus period, estrogen concentration increased in all treatments group, with the highest concentration in the T3 group (61.90 pg/ml) while the lowest concentration was in the T1 group (59.70 pg/ml). Estrus is associated with the most ovarian follicular growth and increased estrogen release. Estrogen is a female sex hormone produced by the ovary that causes behavioral estrus in females. (18, 37). Gonadotropin-releasing hormone (GnRH) stimulates the secretion of estrogen hormone, which has a close relationship with the role of follicles in the ovaries (37). Estrogen is generated by internal theca cells in the antrum of the follicle. Finally, estrogen was absorbed and transported to the target organ via blood arteries (35). In the pregnancy period, estrogen concentration decreased in comparison with the estrus period. In spite of that, the highest concentration was noticed in the T3 group (36.10 pg/ml) with significant differences ($p \leq 0.01$) with other groups T1 (34.40 pg/ml) and T2 (35.00 pg/ml). After parturition, estrogen concentration highly decreased in comparison with other reproductive stages and showed the lowest concentration in the T1 group (11.90 pg/ml), while the highest concentration was in the T3 group (13.20 pg/ml) without any

significant differences among different treatments. Those results agree with the result of the previous study, which concluded that

estrogen concentration was increased during pregnancy and declined after birth (4).

Table 1. Effect of treatment, breed, and their interaction on Serum Estrogen levels (pg/ml) during different period

Periods	Breed	Treatments			Effect of Breeds	Effect of Periods
		T1	T2	T3		
Luteal	Karadi	37.60 ± 0.51 ab	38.20 ± 0.37 ab	38.60 ± 0.24 a	38.13 ± 0.24 a	37.733 ± 0.18 b
	Arabi	36.80 ± 0.37 b	37.20 ± 0.37 ab	38.00 ± 0.32 ab	37.33 ± 0.23 a	
	Effect of T.	37.20 ± 0.33 b	37.70 ± 0.30 ab	38.30 ± 0.21 a		
Estrus	Karadi	59.60 ± 0.51 b	61.60 ± 0.51 ab	62.00 ± 0.71 a	61.07 ± 0.42 a	60.967 ± 0.26 a
	Arabi	59.80 ± 0.66 ab	61.00 ± 0.32 ab	61.80 ± 0.37 ab	60.87 ± 0.34 a	
	Effect of T.	59.70 ± 0.39 b	61.30 ± 0.30 a	61.90 ± 0.38 a		
Pregnancy	Karadi	34.60 ± 0.51 ab	35.20 ± 0.37 ab	36.40 ± 0.51 a	35.40 ± 0.32 a	35.167 ± 0.21 c
	Arabi	34.20 ± 0.37 b	34.80 ± 0.37 ab	35.80 ± 0.37 ab	34.93 ± 0.27 a	
	Effect of T.	34.40 ± 0.30 b	35.00 ± 0.26 ab	36.10 ± 0.31 a		
After Parturition	Karadi	12.40 ± 0.51 ab	12.80 ± 0.58 ab	13.80 ± 0.58 a	13.00 ± 0.34 a	12.533 ± 0.23 d
	Arabi	11.40 ± 0.51 b	12.20 ± 0.37 ab	12.60 ± 0.51 ab	12.07 ± 0.28 a	
	Effect of T.	11.90 ± 0.38 a	12.50 ± 0.34 a	13.20 ± 0.41 a		

a, b, c, d, Means of each factor followed by different letters are significantly different ($p \leq 0.01$).

Progesterone concentration

According to the statistical analysis shown in table 2, the concentration of progesterone differed significantly in the different reproductive stages, with the highest concentration in the pregnancy period (16.33) pg/ml with significant differences ($p \leq 0.01$). Among the three stages (7.133, 0.803, and 0.940) pg/ml, respectively, this result was in accordance with the finding of (30, 4). The results showed a variation in progesterone levels. There is a nonsignificant increase in the progesterone concentration in different treatments T1, T2, and T3 during different stages, in the Pregnancy stage averaged 15.80, 16.10, and 16.50 pg/ml, respectively. Then it declined after parturition in the T1, T2, T3 (0.900, 0.950, 0.970) pg/ml respectively. In the luteal phase, progesterone hormone level increases because of the increased secretion from the corpus luteum; in the animals which ovulated, the plasma progesterone concentration either remained basal or rose to a lower level than that found during the luteal phase of the cycle. Those truths came in agreement with the results of the present by (21). Other experiments have shown comparable results using intravaginal sponges containing progestagens and hCG injection, with or without the addition of prostaglandins. (30, 4). The ability of hCG injection on day 7 post-estrus in female goats to stimulate accessory corpora lutea development and

increase progesterone was effectively reported (39). The hCG luteotrophic action was revealed later after breeding, according to plasma Progesterone concentrations obtained in hCG-treated mice in a prior study (45 days). Luteotrophic activity has also been previously observed by (15). Progesterone concentration in the blood plasma in goats increased to 57.5, 0.0, and 37.0 % of premature luteal regression in groups treated with hCG and GnRH groups, respectively (21). In an earlier study, the progesterone concentration in the peripheral plasma was measured sequentially in individual ewes during the estrous cycle and during gestation and parturition in intact and in ovariectomized ewes (4). During the estrous cycle, progesterone concentration was the lowest to 48 h after the onset of estrus. On the estrus, the progesterone level began to rise to reach a peak on the 10th day (3, 37). After a decline, the level rose to a second peak on the 14th or 15th day. Three to 4 days before the next onset of estrus, the concentration dropped sharply over a period of 48 h to a low basal level. During early pregnancy, the plasma progesterone concentration remained fairly constant at a level similar to the maximum level found during the cycle. A sharp rise started around the 80th day, reaching 15-20 ng/ml around the 110th day. This was followed by a second peak, then a decline in the plasma progesterone concentration before parturition, but the time at which this began

was variable, and even on the day of parturition, the level was generally 3 ng/ml (3, 4, 15). A basal level of (0.5 ng/ml) was reached within 24 h after parturition (32). Increasing progesterone concentration in a group treated with (hCG) may be explained by the ability of hCG to bind the LH receptor, stimulating Progesterone synthesis. Suggested that ewes receiving hCG had more corpus luteum and high serum Progesterone by increasing gene expression in maternal endometrium and promoted expression of proangiogenic factors in fetal extraembryonic membranes (9). Supplementing livestock with hCG may boost P4 levels and improve reproductive efficiency due to augmented CL number per ewe. This

may explain an increased level of progesterone during the pregnancy period in comparison with other reproductive periods. In cyclic ewes, ECG given after progestagen treatment reduces the interval to the onset of estrus when compared with ewes given only progestagen (10). Gonadotropin enhances estrogen concentration and induces estrous and LH surge. Furthermore, the type of breed didn't have a significant effect on estrogen concentration in general, and together the karadi surpassed the Arabi in all periods. this difference may be attributed to the variation in the genetic abilities of the individuals of the two breeds (8).

Table 2. Effect of treatment, breed, and their interaction on Serum Progesterone levels (pg/ml) during different periods

Periods	Breed	Treatments			Effect of Breeds	Effect of Periods
		T1	T2	T3		
Luteal	Karadi	7.000 ± 0.31 a	7.400 ± 0.24 a	7.400 ± 0.24 a	7.267 ± 0.15 a	7.133 ± 0.10 b
	Arabi	6.800 ± 0.20 a	7.000 ± 0.32 a	7.200 ± 0.20 a	7.000 ± 0.14 a	
	Effect of T.	6.900 ± 0.18 a	7.200 ± 0.20 a	7.300 ± 0.15 a		
Estrus	Karadi	0.820 ± 0.04 a	0.840 ± 0.02 a	0.860 ± 0.02 a	0.840 ± 0.02 a	0.803 ± 0.01 c
	Arabi	0.720 ± 0.04 a	0.780 ± 0.04 a	0.800 ± 0.03 a	0.767 ± 0.02 a	
	Effect of T.	0.770 ± 0.03 a	0.810 ± 0.02 a	0.830 ± 0.02 a		
Pregnancy	Karadi	16.00 ± 0.32 a	16.40 ± 0.24 a	16.80 ± 0.37 a	16.40 ± 0.19 a	16.133 ± 0.14a
	Arabi	15.60 ± 0.55 a	15.80 ± 0.37 a	16.20 ± 0.37 a	15.87 ± 0.19 a	
	Effect of T.	15.80 ± 0.20 a	16.10 ± 0.23 a	16.50 ± 0.27 a		
After Parturition	Karadi	0.940 ± 0.05 a	0.980 ± 0.06 a	1.000 ± 0.07 a	0.973 ± 0.03 a	0.940 ± 0.02 c
	Arabi	0.860 ± 0.05 a	0.920 ± 0.05 a	0.940 ± 0.07 a	0.907 ± 0.03 a	
	Effect of T.	0.900 ± 0.04 a	0.950 ± 0.04 a	0.970 ± 0.05 a		

a, b, c, Means of each factor followed by different letters are significantly different ($p \leq 0.01$).

FSH concentration

The results illustrated in the table 3 indicate variation in FSH concentration in the treated groups. In the luteal phase, FSH concentration increased insignificantly in the three treatments protocols T1, T2, T3 averaged 0.810, 0.830, 0.880 mIU /ml, respectively. While in the estrus phase, FSH concentration significantly increased in the T3 group to the value of 3.400 mIU /ml when compared to that of T1 (3.120) mIU /ml and T2 (3.210) IU /ml. Furthermore, no significant changes were noticed in the FSH concentration in the different groups in the pregnancy period. In spite of that FSH, the concentration remains at the highest level in the T3 when compared with the groups T1 and T2. After parturition, the concentrations of FSH in T1, T2, T3 groups dropped insignificantly in all groups with a slight difference in the three groups of treatments which averaged (0.039, 0.043, 0.048)

mIU/ml. Similarly, Theofanakis et al. (38), Sangeetha, and Rameshkumar (30). The increasing level of FSH in the luteal phase in the treated groups may be related to the usage of progesterone for decreasing the length of the proestration procedure by imitating the activity of the corpus luteum or a combination of both (7). These preliminary data in ewes showed that hCG treatment increased prolificacy but lowered fertility. More recent data support that ewes treated with hCG in resentment of inducing more synchronized ovulation than controls without gonadotrophin treatment, had a lower pregnancy rate after artificial insemination (23). Increased FSH in the estrus phase may be related to the effect of hCG hormone, which is known to stimulate ovulation mediated by increasing FSH and LH. Serum hCG concentrations decreased significantly after delivery, with an estimated maximal first half-life of 6.6 hr (36). Serum

FSH reduced only little in the first two days after birth, then increased to near the initial level by postpartum day five. Serum FSH concentrations were within the normal range (adult female premenopausal) in both lactating and nonlactating subjects at 6 weeks postpartum (14). Those findings are consistent with the findings of the present research. The highest concentrations of FSH hormone in different reproductive stages in the group that was treated with GnRH may be related to the regulatory effect of GnRH for FSH gene transcription (40). The hCG is used to promote ovulation during estrus synchronization in ewes may reduce fertility rates may be related to the high frequency of abnormal follicular development patterns, disruptions, and ovulation delays in the treated females, as well as the formation of follicular cysts (7). These findings preclude their practical application to induce ovulation concomitantly to estrous synchronization procedures. More recent data support that

ewes treated with hCG, in spite of inducing more synchronized ovulation than controls without gonadotrophin treatment, had a lower pregnancy rate after TAI (10). The concentration of FSH differed significantly in the different reproductive stages, with the highest concentration in the estrus period (3.243) mIU/ml with significant differences ($p \leq 0.01$) among the three stages (0.840, 0.773 and 0.043) mIU/ml, respectively. This result was in accordance with the finding of Sangeetha and Rameshkumar (30), Kohno et al. (21). Increased FSH in the estrus phase may be related to the effect of hCG hormone, which is known to stimulate ovulation mediated by increasing FSH (7). The type of breed didn't have a significant effect on FSH concentration in general; together the Karadi surpassed the Arabi in all periods. This difference may be attributed to the variation in the genetic abilities of the individuals of the two breeds (8).

Table 3. Effect of treatment, breed, and their interaction on Serum FSH levels (mIU /ml) during different periods

Periods	Breed	Treatments			Effect of Breeds	Effect of Periods
		T1	T2	T3		
Luteal	Karadi	0.820 ± 0.04 a	0.840 ± 0.04 a	0.900 ± 0.05 a	0.853 ± 0.02 a	0.840 ± 0.02 b
	Arabi	0.800 ± 0.04 a	0.820 ± 0.04 a	0.860 ± 0.02 a	0.827 ± 0.02 a	
	Effect of T.	0.810 ± 0.03 a	0.830 ± 0.03 a	0.880 ± 0.03 a		
Estrus	Karadi	3.140 ± 0.08 ab	3.240 ± 0.05 ab	3.420 ± 0.06 a	3.267 ± 0.05 a	3.243 ± 0.03 a
	Arabi	3.100 ± 0.09 b	3.180 ± 0.06 ab	3.380 ± 0.04 ab	3.220 ± 0.05 a	
	Effect of T.	3.120 ± 0.06 b	3.210 ± 0.04 b	3.400 ± 0.03 a		
Pregnancy	Karadi	0.760 ± 0.05 a	0.780 ± 0.04 a	0.820 ± 0.04 a	0.787 ± 0.02 a	0.773 ± 0.02 b
	Arabi	0.720 ± 0.04 a	0.760 ± 0.04 a	0.800 ± 0.03 a	0.760 ± 0.02 a	
	Effect of T	0.740 ± 0.03 a	0.770 ± 0.03 a	0.810 ± 0.02 a		
After Parturition	Karadi	0.042 ± 0.004 a	0.044 ± 0.005 a	0.048 ± 0.004 a	0.045 ± 0.002 a	0.043 ± 0.01 c
	Arabi	0.036 ± 0.004 a	0.042 ± 0.004 a	0.048 ± 0.004 a	0.042 ± 0.002 a	
	Effect of T.	0.039 ± 0.003 a	0.043 ± 0.003 a	0.048 ± 0.002 a		

a, b, c, Means of each factor followed by different letters are significantly different ($p \leq 0.01$).

LH concentration

Table 4 shows that there is insignificant increase in LH concentration in all treatments T1, T2, and T3 averaged 0.251, 0.257, 0.263 mIU /ml respectively in the luteal phase. In the Estrus phase LH increased according to the treatments with the highest level in the T3 (5.300 mIU /ml) treatments with insignificant increase in comparison with other treatments T1 and T2 averaged 5.150 and 5.240 mIU /ml respectively. Concentration of LH reduced in the pregnancy period in T1, T2, and T3 averaged 0.740, 0.770, 0.810 mIU /ml respectively non significantly among groups. After parturition the concentrations of LH

decreased sharply in all treatments. Furthermore, GnRH treatment showed the higher concentration of LH hormone in different reproductive stages. Those results showed that GnRH play a role in the stimulation of LH secretion in comparison with progesterone alone or progesterone in combination with hCG. As a result, there is a lot of study on procedures for stimulating LH secretion, such as GnRH (26,31), or possessing LH activity in a similar way to human chorionic gonadotrophin (hCG). The high similarity between the hormones hCG and LH causes their binding to the same LH receptor (23). Treatment with GnRH induces

earlier return to cyclic ovarian activity by increasing LH concentration. the pituitary of postpartum ewes regains the ability for maximal release of LH in response to GnRH challenge within 7 days postpartum (16). Increasing LH concentration may be related to the effect of estrogen hormone which known to promote the ovulation through LH surge as estrogen shows a positive feedback effect toward the pituitary (22). It is known that hormones have a rhythmic variation among the various reproductive phases; in the present work, the significant level of serum LH is present in the estrus period(5.230) mIU/ml with significant differences($p \leq 0.01$) among

the three stages (0.257,0.121 and 0.062) mIU /ml respectively. This result agrees with those reported on the effect of the period (41,30,19). Increasing LH concentration in the estrus period is related to the estrogen hormone's effect, as estrogen has a positive feedback impact on the pituitary, it is known to stimulate ovulation through an increase in LH (22). The type of breed didn't significantly affect LH concentration in general; together, the karadi surpassed the Arabi in all periods. This difference may be attributed to the variation in the genetic abilities of the individuals of the two breeds (19, 8).

Table 4. Effect of treatment, breed, and their interaction on Serum LH levels (mIU /ml) during different periods

Periods	Breed	Treatments			Effect of Breeds	Effect of Periods
		T1	T2	T3		
Luteal	Karadi	0.254 ± 0.005a	0.260 ± 0.007a	0.266 ± 0.005a	0.260 ± 0.00 a	0.257 ± 0.002 b
	Arabi	0.248 ± 0.004a	0.254 ± 0.005a	0.260 ± 0.004a	0.254 ± 0.003a	
	Effect of T.	0.251± 0.003a	0.257 ± 0.004a	0.263 ± 0.003a		
Estrus	Karadi	5.160 ± 0.07a	5.260 ± 0.05a	5.320 ± 0.06a	5.247 ± 0.04 a	5.230 ± 0.023 a
	Arabi	5.140 ± 0.05a	5.220 ± 0.04a	5.280 ± 0.04a	5.213 ± 0.03 a	
	Effect of T.	5.150 ± 0.04a	5.240 ± 0.03a	5.300 ± 0.03a		
Pregnancy	Karadi	0.118 ± 0.006 a	0.122 ± 0.004a	0.128 ± 0.006a	0.123 ± 0.003a	0.121 ± 0.002 c
	Arabi	0.114 ± 0.005a	0.118 ± 0.004a	0.126 ± 0.005a	0.119 ± 0.003a	
	Effect of T.	0.116 ± 0.004 a	0.120 ± 0.003a	0.127 ± 0.004a		
After Parturition	Karadi	0.058 ± 0.004 a	0.062± 0.009a	0.070 ± 0.007a	0.063 ± 0.004a	0.062 ± 0.003 d
	Arabi	0.054 ± 0.005a	0.060 ± 0.007a	0.066 ± 0.008a	0.060 ± 0.004a	
	Effect of T.	0.056 ± 0.003a	0.061 ± 0.005a	0.068 ± 0.005a		

a, b, c, d, Means of each factor followed by different letters are significantly different ($p \leq 0.01$).

CONCLUSION

Estrus synchronization by using hCG and in a higher magnitude GnRH in combination with progesterone may be effective in inducing reproductive hormones in order to induce follicular growth and ovulation and finally may increase pregnancy rate and production.

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