Effect of prostaglandin and PMSG on prolificacy and some serum biochemical changes of Hamdani ewes synchronized with intravaginal progestagen

F. T. Juma
College of Agriculture\ University of Salahaddin-Erbil

Abstract

The study was designed to evaluate the efficiency of three different hormonal treatment of estrous synchronization in Hamdani ewes reared in northern Iraq (Kardstan). During the breeding season July 2006, 40 adult Hamdani ewes were randomly allocated to four equal groups. The first group $T_1$was treated with inravaginal progestagen sponges containing 40 mg (FGA) for 14 days. whereas second group T_2 was treated as T_1 group but injected with 500IU PMSG at the sponges removal, while the third group T_3 was treated with progestagen intravaginal sponges for 10 days, at time of sponges withdrawal prostaglandin $F_2\acute{\alpha}$ 5mg (Dinoprost) was injected, the last group left as control group C. The estrous response was comparable in treatment, T1 (90%) T_2 (100%), T_3 (100%) as compared with group C (80%) The T_2 and T_3 were the higher estrous response than the other groups. The highest and lowest number that lamed was 100% in groupT_2 and 70% in group C. Lambing rate and twin born lamb was higher in group T_2. The biochemical examined were included Aspartate transaminase (AST), Alanine transaminase (ALT), and Alkaline phosphatase (ALP), also concentrations of total protein, albumin, total cholesterol, ionized calcium, inorganic phosphor and serum glucose. Data show that there was no significant effect of hormonal treatment .There were significant ($P\leq0.05$) increased in activities of AST, ALT and ALP enzymes also concentration of total protein and concentration of total cholesterol, during late pregnancy and at parturition. The concentrations of serum glucose and inorganic phosphor were significant decreased during late stage of pregnancy and at parturition there were no significantly effects of pregnancy and parturition on the concentrations of serum glucose and ionized calcium. Its concluded from these results that the administration of an impregnate intravaginal sponges for 14 days plus a dose of 500IU PMSG was increased, lambing rate and litter size as compared to the other hormonal treatment Considered.

تأثير هرمون البروستكلاندين ومصل فرس الحامل على الخصب وبعض الصفات البيوكيميائية في مصل نعاج الحمداني الموحدة الشبق بالأسفنجات المهبلية

فاروق طيب جمعة
كلية الزراعة/ جامعة صلاح الدين–أربيل

الخلاصة

أجريت الدراسة لتقيم بعض الهرمونات المستخدمة لتوحيد الشبق في النعاج الحمداني شمال العراق (كردستان) خلال موسم التناسل (تموز) 2006. وزعت 40 نعجة بالغة عشوائيا الى أربعة مجاميع متساوية عوملت المجموعة الأولى باستخدام الأسفنجات المهبلية المشبعة 40 ملغم البروجستاجين (FGA) لمدة 14 يوم. أما المجموعة الثانية فقد عوملت نفس المجموعة الأولى و حقنت بجرعة 500 وحدة دولية PMSG عند سحب الأسفنجات المهبلية. المجموعة الثالثة استخدمت نفس الأسفنجات المهبلية لمدة 10 أيام حقنت بعد سحبها بهرمون البروستكلاندين 5 ملغم Diroprost. تركت المجموعة الرابعة بدون معاملة وعدت كمجموعة سيطرة (c).

أظهر ت النتائج أن الحيوانات التي أظهرت الشبق في المجموعة الأولى 90%، المجموعة الثانية 100% والمجموعة الثالثة 100% مقارنة مع مجموعة السيطرة التي كان 80%. لوحظ أعلى نسبة للمواليد في المجموعة الثانية (130%) وأوطاءها في مجموعة السيطرة (70%). أن معدل التوائم ومعدل الو لادات والخصب مرتفعة في المجموعة الثانية مقارنة ببقية المجاميع. أما التغيرات الكيموحيوية فلوحظ وجود فروقات معنوية ($p\leq0.05$) بزيادة تراكيز أنزيمات AST، ALT، ALP وكذلك زيادة تركيز البروتين الكلي و الكلستيرول الكلي في مصل الدم خلال المراحل الأخير ة من الحمل وعند الولادة. لوحظ انخفاض معنوي في تركيز كل من الكلوكوز و الفسفور اللأعضوي في مصل الدم خلال نفس الفترة. لم يلاحظ وجود أي فروقات معنوية في تركيز كل من الألبومين والكالسيوم المتأين خلال مراحل الحمل وعند الولادة. نستنتج من هذه الدراسة أ ن استخدام الأسفنجات المهبلية المشبعة 40 ملغم البروجستاجين (FGA) مع حقن هرمون PMSG 500 وحدة دولية أد ى إلى زيادة توحيد الشبق و معدل التوائم.

Introduction

Among the indigenous sheep breeds of Iraq, the Hamadani is the largest in size and have good mutton, milk and wools producing ability, Hamdani is vary popular among sheep keepers Erbil governorate of northern Iraq (Kurdstand). Synchronization of estrus in sheep has been achieved by using intravaginal impregnated sponges which contain synthetic progestagan as Fluorogeston acetate, FGA (1). Synchronization probably the most popular practice used to improve conception rate during season and out season breeding. Several synchronization procedure incorporate injection of pregnant mare serum gonadotropin (eCG) equine chorionic gonadotropin at the end of the progestagen treatment in order to obtain more precise and reliable synchronization of estrus.

This incorporation has proved to improve occurrence of ovulation and fertility in small ruminants (2). Estrus synchronization in small ruminant with prostaglandin (PGF_2) is reducing the length of corpus luteal phase of the estrus cycle. The synchronization of oesrous with prostaglandin in ewes is efficient when the functional corpus luteum is present on the ovary (3). The administration of PGF_2 to progestin synchronized ewes, would enhance the synchrony of ovulation, PGF_2 may be reduce the

lifes of corpus luteum (4). The aim of this study to investigate the effect of using FGA intravaginal sponges alone or combined with eCG or with $PGF_{2\&}$ for synchroning Hamdani ewes. These three hormonal treatment are carried out during normal breeding season to evaluate the efficiency of three different hormonal treatment for estrus synchronization, in addition to study the changes in some serum biochemical constituents.

Material and Method

The study was carried out at Gradarasha animal's field/college of Agriculture/ University of Salahaddin-Erbil north of Iraq, from July 2006 to January 2007, and included 40 cyclic Hamdani ewes 3-4 years of age and ranging from 45-50 kg body weight, with normal reproductive history. The ewes were randomly allocated equally into four Groups. The animals were treated as following:

1. Group1 (C) control group.
2. Group2 (T1) was treated with polyurethane vaginal sponges impregnated with 40mg Florogestone acetate (FGA)* for 14 days.
3. Group2 (T2) was treated as in T1group but was injected intramuscularly with eCG**500 I.U. at withdrawal of intravaginal sponges.
4. Group3 (T3) was treated as T1 group but the sponges were removed after 10days. It was injected intramuscularly with 5 mg eCG (Dinopsrot) at the time of sponges with drawl

The control was bred at naturally occurring estrus. Three Hamdani rams 4-5 years of age were used to inseminate ewes by handing mating during standing estrus in each group.

- **Biochemical Analysis:**

Blood samples were collected from ewes via Jugular vein-puncture using non-heparinzed vacutainer tubes. Serum was harvested and stored at freezing at (-20^0C) until analysis. Enzymes activities of aspartate aminotranferase, AST, alanine transferase, ALT and alkaline phosphates, ALP, sepectrophtometrically by using commercially available diagnostic kits (Rondox, Laborateries, U.K.). Serum total protein was determined by using Biuret method (5), while the albumin concentration was measured according to (6) method, total cholesterol concentration was analyzed by the method of Allain (7). Serum calcium concentrations were estimated using the calcium kit (BioMerieu, B.A France). Serum phosphorus was determined according to (8). Serum glucose was measured by the method of (8). Result was statistically analyzed using computer program system, SAS, 2000(9). Comparison between means of each factor was tested by Duncan's Multiple Range Test (10).

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Results

The estrus response, time from sponges withdrawal to estrus and the duration of the induce estrus period following the treatment with FGA (T1) alone or combined with eCG (T2) or $PGF_{2\acute{\alpha}}$ (T3) are shown in Table (1). The estrus observed on T1, T2, T3 and group C were 90%, 100%, 100% and 80% respectively. There were no significant difference between the treated groups and the control in one the twin rate, lambing rate and litter size. The twinning rate and litter size in T2 group were more as compared with the other groups. Cholesterol concentration in the serum was changed during pregnancy, it decline ($P \leq 0.05$) during mid pregnancy and increase ($P \leq 0.05$) during late stage of pregnancy and at parturition. There were a significant ($P < 0.05$) difference stages of pregnancy and parturition on the activities of AST, ALT enzymes in the serum especially during the mid and late stages of pregnancy (Table 2).

The level of serum glucose was significantly decrease ($P \leq 0.05$) during the last month of pregnancy and at parturition. There was no significant difference observed in the level of calcium serum between stages of pregnancy and at parturition, while there was a significant increase ($P \leq 0.05$) in the serum phosphor level at parturition comparing with the stages of pregnancy (Table 3).

Table (1) The effect of hormones on the reproductive performance of Hamadani ewes

Traits	C	T1	T2	T3	Level of Significance
No. of ewes	10	10	10	10	
Estrus response %	80	90	100	100	
Sponges with drawl to estrus (hours)	-	53.55 ± 6.91	54.4 ± 5.87	49.0 ± 7.78	NS
No. lamb born	8	10	13	11	
No. ewes born	7 (70 %)	8 (80 %)	10 (100 %)	9 (90 %)	*
No. of twins born (twin rat %)	14.28	25	30	22	*
Lambing rat %	80	100	130	110	*
Litter size	1.14	1.25	1.3	1.22	*

***-P ≤0.05**

Table (2) The effect of treatment, stages of pregnancy and parturition on serum cholesterol, total protein and albumin concentration in Hamdani ewes

Factors	No.	Total chl.mg/dI	Total Protein	Albumin
Over all mean	120	82.28 ± 1.17	7.06 ± 0.05	3.12 ± 0.03
Treatments		NS	NS	NS
C	30	81.33 ± 2.20	6.94 ± 0.08	3.08 ± 0.07
T1	30	81.97 ± 2.26	7.09 ± 0.12	3.13 ± 0.05
T2	30	81.30 ± 2.52	7.04 ± 0.10	3.18 ± 0.04
T3	30	84.50 ± 2.39	7.16 ± 0.07	3.10 ± 0.04
Periods		**	**	NS
First month	20	87.30 ± 3.64 a b	7.02 ± 0.09 b c	3.18 ± 0.07
Second month	20	74.55 ± 1.89 d	6.99 ± 0.10 b c	3.09 ± 0.07
Third month	20	75.10 ± 1.86 d	6.79 ± 0.13 c	3.13 ± 0.10
Forth month	20	84.90 ± 2.57 b c	7.18 ± 0.08 a b	3.05 ± 0.05
Fifth month	20	78.20 ± 2.19 c d	7.02 ± 0.08 b c	3.12 ± 0.05
Parturition	20	93.60 ± 2.03 a	7.37 ± 0.15 a	3.18 ± 0.03

Means ± (S. E.)
Means with different letters within the same column are different (P≤0.05).

Table (3) The effect of treatment, pregnancy stages and Parturition on serum enzymes activities in Hamdani ewes

Factors	No.	AST U/L	ALT U/L	ALP U/L

Over all mean	120	25.58 ± 0.50	11.73 ± 0.23	36.00 ± 0.58
Treatments		NS	NS	NS
C	30	25.17 ± 1.02	11.33 ± 0.40	35.67 ± 1.28
T1	30	25.60 ± 0.95	11.63 ± 0.45	36.13 ± 1.18
T2	30	25.63 ± 1.04	12.00 ± 0.48	35.27 ± 1.23
T3	30	25.90 ± 1.03	11.93 ± 0.49	36.93 ± 0.97
Periods		**	**	**
First month	20	31.85 ± 1.23 a	9.35 ± 0.37 c	26.50 ± 1.32 b
Second month	20	29.30 ± 1.14 b	9.70 ± 0.30 c	36.55 ± 1.13 a
Third month	20	20.95 ± 0.88 d	11.15 ± 0.36 b	38.15 ± 0.91 a
Forth month	20	25.70 ± 0.52 c	13.85 ± 0.42 a	39.00 ± 0.89 a
Fifth month	20	22.40 ± 0.63 d	13.00 ± 0.36 a	37.30 ± 1.19 a
Parturition	20	23.25 ± 0.63 c d	13.30 ± 0.54 a	38.50 ± 0.87 a

Means ± (S. E.)
Mean with different letters within the same column are different (P≤0.05).

Table (4) The effect of treatment, stages of pregnancy and parturition on the serum phosphor, calcium and sugar concentrations in Hamdani ewes

Factors	No.	P.mmol./IL	Ca.mmol/IL	Glucose
Over all mean	120	5.84 ± 0.14	8.82 ± 0.06	40.33 ± 1.04
Treatments		NS	NS	NS
C	30	5.68 ± 0.25	8.86 ± 0.11	38.80 ± 2.12
T1	30	5.75 ± 0.28	8.78 ± 0.12	40.57 ± 2.21
T2	30	5.99 ± 0.30	8.76 ±0.14	40.60 ± 1.98
T3	30	5.93 ± 0.28	8.89 ± 0.14	41.37 ± 2.09
Periods		**	NS	**
First month	20	6.62 ± 0.18 a	8.65 ± 0.17	52.40 ± 3.83 a
Second month	20	7.00 ± 0.28 a	8.66 ± 0.17	40.60 ± 1.61b
Third month	20	6.61 ± 0.26 a	9.02 ± 0.15	40.75 ± 1.92 b
Forth month	20	3.52 ± 0.11 c	8.76 ± 0.17	39.05 ± 1.71 bc
Fifth month	20	5.89 ± 0.2 b	8.97 ± 0.16	32.90 ± 1.79c
Parturition	20	5.41 ± 0.23b	8.89 ± 0.11	36.30 ± 1.47 bc

Means ± (S. E.)
Mean with different letters within the same column are different (P≤ 0.05).

Discussion

The increase in the response of induced estrous in the treated groups as compared with the control group might be due to the effects of the used hormones on the activity of ovaries (1) .These results are comparable to the finding of Sleeker *etal.* (12) using MAP plus 300 IU eCG, and reported a 100% oestrus response in Merino ewes. Precise interval period between sponge withdrawal to the appearance of oestrus 53.55±6.91, 54.4±5.87, 49.0±7.78, hours in T1, T2, T3 respectively were similar to the observation of Karagiannidis (12) in Greek ewes, who recorded an interval of 54-60h. The short period of the onset of oestrous observed in T_3 group might be attributed to lutolytic effect of $PGF_2\alpha$ that bring animal into estrus and has a depressed effects on progesterone (4). The highest (130%) and lowest (80%) lambing rate in the present study which recorded in group T2 and control group respectively. This is in agreement with Safdarian etal., (13). McNaity *etal.* (14) previously suggested that the use of intravaginal sponges fluorogestone with eCG have an important role in the growth of follicles. Those group using eCG in the treatment had a higher litter size compared to those groups that had not used eCG (13). In the present study the litter size in group T2 was 1.3 which is in agreement with the finding of (15,16). However Ackermann (17) reported a lower Litter size (1.08) using combination of MAP sponges and eCG for estrus synchronization in Dorper ewes. The treated ewes with PGF_2 had less affinity to litter size than eCG. These finding are similar to those perviously reported by Fairinie

etal (18). It may be noticed here that the biochemical parameters namely, AST, ALT, ALP enzymes, serum total protein, total cholesterol, serum ionized Calcium concentration, serum inorganic Phosphorous concentration and serum glucose concentration were not affected by the hormonal treatment which given support to the using of these hormones for synchronization. The pregnancy have an effect on concentration of AST, ALT, ALP enzymes (19) which decreased during the second period of gestation then increased at the late stage of gestation and after parturition (20). The significant increased in the serum enzymes activities could be resulted from high level of AST and ALT after delivery. Kupczynski, *etal* (21) showed high levels of AST, ALT, after delivery and related might be to the increase metabolic efforts caused by lactation. However Vihan and Rao (22) explained attributed the increase AST, ALT enzymes to increased requirement for amino acids in milk production. The increased ALP enzyme activity in this experiment at third and forth month of pregnancy and its reduction at fifth month of pregnancy is due to formation of skeletal bone tissue of fetus (23). The result here supported the founding of Criste etal., (24) who found an increase in activity of ALP in cow after delivery. The total cholesterol had an increased tendency to the end of the observed period the ranged from 78.2±2.03 to 93.6±203.The dynamic of change in the decrease in the total cholesterol concentration before parturition and increase in its level after parturition was recorded by (25) in cow. The cholesterol has a role of fatty acid carrier of milk synthesis (26).The serum total protein was decreased in mid gestation and significantly increased ($P \leq 0.05$) during pre-parturition and increased in post-parturition. This might be due required of more transfer of protein into colostrums. This result in agreement with Okab, etal., (20)in Rahmani ewes. Boyoumi, *etal*(27) observed increased the total protein in mid gestation which needed for embryo growth and post-parturition, More, *etal* (28) showed influence of pregnancy on total protein in Malpura ewes had a declining trend with advance of pregnancy and mainly towards the lambing period. The glucose level largely declined with approach of pregnancy, indicating an increasing demand for glucose being stored as glycogen, by the developing placenta and uterine, by the fetus (28), while Bulent, *etal* (29) reported exponent decrease in serum glucose concentration during the late pregnancy and parturition due to rapid utilized of glucose to wards the fag end of pregnancy. The plasma inorganic phosphoras was significant decrease in late pregnancy and after parturition. This decrease was due to the required mint for milk production (30).

It was concluded from this study that Hamdani ewes can be effectively synchronized intravaginal sponges and 500 IU eCG during breeding season with high success rate.

Acknowledgment

Author is thankful to Dr. Salah Mustafa Bakir, Director General of Vano Group for supplying the hormones for this study.

References

1. Gordon, I. (1997). Controlled Reproduction in Sheep and Goats. (1st ed.) Cab International, Wallingford, UK.
2. Evans, G. & Robinson, T. J. (1980). The control of fertility in sheep: Endocrine and Ovarian responses to progestagen- PMSG treatment in the breeding season and in anoestrus. J. Agric. Sci. Cambridge., 94: 69-88.
3. Whymam, D. & Moore, R. W. (1980). Effect of PMSG and prostaglandin $F_2\acute{\alpha}$ Analogue, Cloprostenol on superovulation, fertilization and egg transport in Ewe. J. Reprod. Fert., 60: 267-272.

4. Shutt, D. A.; Smith, L. D. & Sherman, R. P. (1976). Prostaglandin and lutelysis in the goat and human. Theriogenology, 6:610.
5. Henry, J. B. (1974). Clinical Diagnosis and Management by Laboratory Method. W. B. Saunders and Co. Philadelphia.
6. Doumas, B. T.; Waston, A. & Briggs, H. G. (1971). Albumin standers and the measurements Of serum creatnine with bromcresol green. Clinical Chemistry Acta, 31:87-90.
7. Allain, C.; Poon, I. S.; Chan, C. S. G. & Richmond, W. F. U. P. (1974). Ezymatic determination of total chlosterol. Clinical Chemistry Acta, 20:470-475.
8. Hawk, P. B.; Oser, B. L. & Summero, W. H. (1974). Practical Physiological Chemistry13thed. McGraw-Hill Book Co. N. Y.
9. SAS, (2005). Statistics guide for personal computers. Version 8 release 8.2 SAS Int. Cary NC. USA.
10. Duncan, D. B. (1955). Multiple range and multiple F test. Biometrics, 11:1-27.
11. Maxwell, W. M. C. (1986). Induction of estrus in ewes using a controlled internal release device and PMSG. J. Agric. Sci. (Comb.), 16:201-203.
12. Karagionnidis, A.; Varsakeli, S.; Karatz, G. & Brozos, C. (2001). Effect of time of artificial insemination on fertility of progestagen and PMSG treated indigenous Greek ewe, during non-breeding season. Small Rum. Res., 39:67-71.
13. Safdarian, M.; Kafi, M. & Hashemi, M. (2006). Reproductive performance of Karakul Ewes following different oestrous synchronization treatment outside the natural Breeding season. S. Afr. J. Anim. Sci., 36:229-234.
14. McNatty, K. P.; Hudson, N. I.; Ball, M.; Henderson, K.; Heath, K. M.; Lun, D. A. & Kieboom, I. E. (1985). FSH influences follicles viability, estradiol biosynthesis and ovulation rate in Rommey ewes. J. Reprod. Fert., 75:121-125.
15. Erasmus, J. A.; Faure, A. S. & Smith, J. M. (1994). Production and reproduction Performance following progestagen–PMSG treatment of Dorper ewes on veld. Proce. 33rd Conger. S. Afri. Soc. Anim. Sc. (Abstr.)
16. Zeleke, M. J.; Grcyling, P. C.; Schwalbach, L. M. J.; Muller, T. & Erasmus, J. A. (2005). Effect of progestagen and PMSG on estrus synchronization and fertility in Dorper ewes during transition period. Small Anim. Res., 56:47-53.
17. Ackermann, J. J. (1993). Study of Dorper sheep in the Dorper land area. M.Sc. Thesis University of Stellenbosch, South Africa.
18. Fainie, I. J.; Wales, R. S. & Gheradi, P. B. (1977). Time of ovulation, fertilization rate and biastocyst formation in ewes following treatment with prostaglandin Analogue (ICI 80996). Theriogenology, 8:183.
19. Juma, F. T. & Marzani, (2007). Effect of some hormonal treatment on reproductive Performance and biochemical changes in blood serum of Hamdani hoggets in Erbil. Kirkuk Uni. J., 2: 2-9.
20. Okab A. B.; Mekkawy, M. Y.; Elbnna, I. M.; Hassan, G. A.; El-Nouty, E. D. & Salem, M. H. (1992). Season changes in plasma volume, adrenocortical hormones, Osmolality and electrolytes during pregnancy and at parturition in Bahi and Rahmani ewes. Indian J. Anim. Sci., 62: 302-306.
21. Viha, V. S. & Rai, P. (1987). Certain hematological and biochemical attributes during Pregnancy, parturition and post parturition period in sheep and goats. Ind. J. Anim. Sci., 57: 1200-1204.

22. Kupczynski, R. & Chudoba-Drozdowska, B. (2002). Values of selected biochemical Parameters of cows blood during their drying-off and the beginning of lactation. Electronic J. Polish Agri. Uni., 5:1-10.
23. Healy, P. J. (1971). Serum alkaline phosphatase in sheep. Clin. Chem. Acta, 33:431-436.
24. Criste, W. L.; Ludwiek, T. M.; Drum, E. W. & Davis, D. R. (1967). Effect of season, stage of lactation, stage of gestation and milk production on serum transaminase Activity (Abstract) J. Dairy Sci., 50:998.
25. Bekeova E.; Elecko, J.; Hendrichovsky, V.; Choma, J. & Karjnicakova, M. (1987). The effect of beta-carotene on the changes in T_4 and cholesterol concentration in Calving of heifers before and after parturition. Vet. Med., 32:459-468.
26. Parkash, B. S. & Tndon, R. N. (1979). A note on the effect of late pregnancy and early Lactation on blood serum cholesterol and total lipids of Holstein x Tharparkar first Lactation cows. Indian J. Anim. Sci., 49:47-61.
27. Bauyoum, M. T.; Assad, F.; Nassar, A. M. & Abd El-Baky, S. M. (1986). Serum protein electrophoriss in Different physiological in ewes. World. Anim. Prod., 22:55-58.
28. More, T.; Munshi, S. K. & Chattopadhyay, S. K. (1973). A note on the effects of Pregnancy on some organic constituents of blood in various breeds of sheep. Indian J. Anim. Sci., 43: 552-555.
29. Bulent, E.; Kabu, M. & Elitok, O. M. (2006). Evaluation of liver function tests in cows during periparturitent period. F.u. Saglik Bil. Dergisi., 20:205-209.
30. Sansom, B. F.; Bunch, K. J. & Dew, S. M. (1982). Change in plasma calcium, magnesium, phosphorus, hydroxylproline concentration in ewes from 12 weeks before till 3 weeks after lambing. Br. Vet. J., 18: 391-401.