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TREATMENT OF OVARIAN INACTIVITY IN POST PARTUM BUFFALO-COWS WITH SPECIAL REFERENCE TO ITS ECONOMIC EVALUATION

(With 5 Tables)

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علاج خمول المبايض بعد الـولادة فى الجاموس وتقييمه الاقتصـادى

كمـــال كمـــال متولى

استهدفت هذه الدراسة تقييم تأثير محاولات علاجية مختلفة لخمول المبايض بعد الولادة فى الجاموس وتقييمها الاقتصادى. اجريت هذه الدراسة على عدد ٧٥ جاموسة و التى لم يظهر عليها الشبق طيلة الثلاثة اشهر التالية للولادة وظهر فى الفحص الاكلينيكى وتحليل هرمون البروجستيرون أنها تعانى من خمول المبايض. قسمت الحيوانات الـى خمـس مجموعـات متساوية (١٥جاموسة فى كل منها). المجموعة الاولى: (ضابطة) تم حقنها بـ ٥ مل محلول ملح فسيولوجى. المجموعة الثانية : تم حقنها بـ٥ مل ريسبتال (الهرمـون الحـاث للغـدة النخامية). المجموعة الثالثة: تم حقنها بـ ٤٠ مجم بروجستيرون لمدة ثمانية أيـام متتاليـه و٥ مل ريسبتال فى اليوم الثامن. المجموعة الرابعة: تم حقنها بـ ٤٠ مجم بروجسـتيرون لمدة ثمانية ايام متتالية و٢ مجم و١مجم من بنزوات الاستراديول فى اليـوم الاول و الثـامن على الترتيب. المجموعة الخامسة: تم حقنها بـ ١٠ مل تونوفوسفان لمدة يـومين متتـالين بالاضافة الى ٢٠٠ جرام ثنائى فوسفات الصوديوم و ٢٥٠ جـرام مـن مخلـوط المعـادن والاملاح أعطيت مقسمة على خمسة أيام مخلوطة بالردة. تمت ملاحظة الحيوانـات لرصـد الشبق بواقع ٣مرات يوميا (فجراً – ظهراً – مساءاً) تم تلقيح الحيوانـات التـى أظهـرت الشبق طبيعياً كما جمعت عينات البلازما بعد ثمانية أيام من التلقيح لتحليل البروجسـتيرون. تم فحص الحيوانات التى لم تظهر عليها علامات الشبق أسبوعياً. تم تشخيص الحمـل عـن طريق الجس المستقيمى ٤٢ يوم بعد التلقيح. أظهرت النتائج ان نسبة الحيوانات التى أظهرت الشبـق كانت ٤٠% و٧٣,٣ % و ٨٠% و٦٠% و٥٣,٣ % فى المجموعات من الاولى الى الخامسة على الترتيب. أظهرت المجموعة الثالثة أقصر مدة مابين العلاج و ظهور الشبق وما بين العلاج حتى الحمل وما بين الـولادة حتـى الحمـل (٨,١ ± ١,٧ و ٢٠,٩ ± ٤,٧ و١١٦,٤ ± ٦,١ يوم على الترتيب). تلتها المجموعة الرابعة (١٨,٣ ± ٥,٧ و٣١,٩ ± ٥,٧ و١٢٧,٧ ± ٦,١ يوم على الترتيب). بينما أظهرت المجموعة الاولى الضابطة أطـول مـدة مابين العلاج و ظهور الشبق (٥٢,٦ ± ٤,٦ يوم) وما بين العـلاج والحمـل (٦٦,٥± ٦,٢ يوم) وما بين الولادة و الحمل (١٦١,٦±٦,٩ يوم). كان اقل معدل للتلقيحات بالنسـبة لحدوث الحمل (١,٦± ٠,٢) فى المجموعة الثالثة وكان اعلاهـا فـى المجموعـة الرابعـة

(١,٨٦±٠,٣) وكان أعلى معدل للحمل بعد التلقيحه الاولى هو ما سجلته المجموعة الثالثة (٦٠%) بينما تساوت باقى المجموعات فى هذا المعدل (٤٦,٧ %). أظهر تحليل هرمون البروجستيرون فى بلازما الدم الى زيادة مستواه عند اليوم الثامن بعد التلقيح فى اشارة الى فعالية الجسم الاصفر فى هذه المرحلة. كما أظهرت دراسات الربح والتكلفة الى أن أعلى قيم الربحية كانت فى المجموعة الثالثة تلتها المجموعة الرابعة. مما سبق يمكن ان نستخلص أن المعاملة الهرمونية لحمول المبايض فى الجاموس بعد الولادة أدت الى أفضل النتائج وأدت الى تحسن الخصوبة وتقصير الفترة ما بين الولادتين.

SUMMARY

The objective of this study was to evaluate the effects of different trials for treatment of post partum (p.p.) ovarian inactivity in buffaloes and their economic values. 75 buffalo-cows not observed in estrus 3 months followed parturition, were used in this study. Clinical examinations and progesterone assay revealed ovarian inactivity. The animals were randomly allocated to 5 treatment groups. Group I (GI) was kept as control (injected by 5 ml saline). Group II (GII) was injected by 0.02 mg buserelin (5 ml Receptal, GnRH analogue). Group III (GIII) was injected by 40mg progesterone for successive 8 days and 0.02 mg buserelin at 8th day. Group IV (GIV) was injected by 40mg progesterone for successive 8 days and 2mg and 1mg estradiol benzoate at 1st and 8th day respectively. Group V (GV) was injected by 10 ml tonophosphane daily for 2 days and received 200 gm disodium phosphate and 250 gm mineral mixture divided into 5 days with bran mash. All buffalo-cows were observed for estrus detection three times daily (at dawn, afternoon and evening). Animals came in estrus were naturally mated using fertile buffalo-bulls and their plasma were tested for progesterone level 8 days after mating. While animals did not came in estrus were weekly examined. Pregnancy diagnosis was done using rectal palpation 42 days after mating. The results indicated that, buffalo-cows came in estrus were 40%, 73.3%, 80%, 60% and 53.3% in GI, GII, GIII, GIV and GV respectively. GIII showed the shortest treatment-estrus interval (TEI), treatment conception interval (TCI) and days open (DO) (8.1±1.7, 20.9±4.7 and 116.4±6.1 days, respectively), followed by GIV (18.3±5.7, 31.9±5.7 and 127.7±6.1 days, respectively). While, the longest TEI, TCI and DO (52.6±4.6, 66.5±6.2 and 161.6±6.9 days, respectively) was obtained in GI. The lowest service per conception (S/C) (1.60±0.2) was obtained in GIII while, the highest was obtained in GV (1.86±0.3). The highest 1st service conception rate was obtained in GIII (60%) while, it was equal in other groups (46.7%). Plasma progesterone assay revealed

highly significant increase in progesterone levels at day 8 post service (>1ng /ml) indicating functioning corpora lutea. The obtained net profit, profit/cost ratio, net profit/return and net profit/cost in GIII were the highest followed by that of GIV. In conclusion, hormonal treatment of buffalo-cows proved useful in reducing the calving interval and to increase fertility.

Key words: *Ovarian inactivity, infertility, estrus, parturation, buffalo-cows.*

INTRODUCTION

In recent decades, buffalo farming has expanded widely in Mediterranean areas and in Latin America, but also in Central/Northern Europe where several herds were introduced (Barile, 2005). Reproductive efficiency is the primary factor affecting productivity and is hampered, in female buffalo, by the late attainment of puberty, seasonality of calving, long postpartum anestrous and subsequent calving interval (Barile, 2005). Moreover, he added that, artificial insemination (AI), which is normal practice in cattle, is seldom performed in buffalo, because of the weakness of estrus symptoms and the variability of estrus length, which make estrus detection very difficult. Resumption of estrous cyclicity after calving which is a critical factor in obtaining a satisfactory reproductive performance, remains an important problem specially for buffaloes. Ovarian hypofunction tended the females to show long anestrous periods after calving which could extend to 11 months for buffaloes (Barr, 1963). Excessively long calving conception interval in the buffalo results in substantial economic losses, as well as creating managerial problems (Singh, *et al.,* 1979). One of the most important factors responsible for ovarian inactivity during the postpartum period is believed to be suppressed pituitary function (Gordon, 1996). The ability of the pituitary to respond to GnRH is restored by day 20 postpartum in Murrah buffaloes (Palta and Madan, 1995). In addition, ovarian inactivity affected 30-40% of lactating buffaloes and persist until the calves are weaned naturally or separated from their dams (Gordon, 1996) for this reasons many treatment have been used to treat such cases.

This study aimed to evaluate some trials of treatments of post partum ovarian inactivity in Egyptian buffalo-cows and the economic evaluation of theses trials.

MATERIALS and METHODS

Animals:

This study was performed using buffalo-cows in their first, second or third lactation period. Seventy five buffalo-cows without a corpus luteum (CL) or follicles on their ovaries and without uterine or ovarian pathology upon rectal palpation and not detected in estrus at least for 3 months postpartum were used in this study. The animals were present in private farms in Kafr-El-Sheikh Province, Egypt. They were fed on balanced ration and free from internal and external parasites. Blood plasma progesterone assay by RIA (El-Banna *et al.*, 1985) revealed that, the progesterone levels were < 1ng/ml. The clinical examinations and progesterone assay indicated ovarian inactivity. Average daily milk yield was recorded in all animals.

Treatments:

The animals were divided randomly into five equal groups (15 buffalo-cows per each):

Group I (GI) was kept as control and injected i.m. by 5 ml saline. Each case costed zero LE.

Group II (GII) was injected i.m. by 0.02 mg buserelin (5 ml Receptal, GnRH analogue, Intervet International) given by two shots 1 hr interval (3 ml in first shot and 2 ml in second shot.). Each case costed 25 LE.

Group III (GIII) was injected i.m. by 40 mg progesterone (Lutone, Misr Co. for Pharm. Ind. S.A.E., Egypt) for successive 8 days and 0.02 mg buserelin at 8th day at two shots 1 hr interval. (0.012 mg in first shot and 0.008 mg in second shot.). Each case costed 40 LE.

Group IV (GIV) was injected by 40mg progesterone for successive 8 days and 2mg and 1mg estradiol benzoate (Folone, Misr Co. for Pharm. Ind. S.A.E., Egypt) at 1st and 8th day respectively. Each case costed 20 LE.

Group V (GV) was injected by 10 ml tonophosphane (Intervet International) daily for 2 days and received 200 gm disodium phosphate (El-NasrPharm. Co., Egypt) and 250 gm mineral mixture (Agri-Vet, Egypt) divided on 5 days on bran mash. Each case costed 15 LE.

All buffalo-cows were observed for estrus detection three times daily (at dawn, afternoon and evening). Animals came in estrus were naturally mated using fertile buffalo-bulls (each mating costed 10 LE) and their plasma were tested for progesterone level 8 days after mating. While animals did not came in estrus were weekly examined. Pregnancy diagnosis was done using rectal palpation 42 days after mating.

Reproductive efficiency measures of buffalo-cows were calculated:
1. Treatment estrus interval (TEI).
2. Treatment conception interval (TCI).
3. Calving conception interval (Days Open "DO").
4. Number of services per conception (S/C).
5. First service conception rate.

N.B. - The price of milk = 2.5 LE/kg

Statistical analysis:

The obtained results were statistically analyzed using Statistical Analysis System (SAS, 1987).

RESULTS

The results are presented in tables 1-5. The effects of different treatments of ovarian inactivity in buffalo-cows (Table 1) indicating that, the buffalo-cows came in estrus were 40%, 73.3%, 80%, 60% and 53.3% in GI, GII, GIII, GIV and GV respectively. The shortest TEI, TCI and DO (8.1±1.7, 20.9±4.7 and 116.4±6.1 days, respectively) were obtained in Group III followed by that in Group IV (18.3±5.7, 31.9±5.7 and 127.7±6.1 days, respectively). While, the longest TEI, TCI and DO (52.6±4.6, 66.5±6.2 and 161.6±6.9 days, respectively) were obtained in Group I (control group). The lowest S/C (1.60±0.2) was obtained in Group III while, the highest was obtained in Group V (1.86±0.3). The highest 1st service conception rate was obtained in Group III (60%) while in other groups; it was equal (46.7%).

Blood plasma progesterone assay revealed that, in all Groups, there were highly significant ($P < 0.01$) increase in progesterone levels at day 8 post service (>1ng /ml) indicating functioning corpora lutea (Table 2).

Group III had DO less than Group IV, II, V and I by 11.3, 15.5, 40.9 and 45.2 days respectively. Group IV had DO less than Group II, V and I by 4.2, 29.6 and 33.9 days respectively. Group II had DO less than Group V and I by 25.4 and 29.7 days respectively. Group V had DO less than Group I by 4.3 days (Table 3).

Returns, cost and net profit of different treated groups of ovarian inactivity indicated that, Group III gave the highest net profit and the lowest cost followed by Group IV. However, the lowest net profit and highest cost were obtained in Group I (Table 4).

In the same manner, economic efficiency measures of drugs used in different treated groups of ovarian inactivity indicated that, the

highest profit/cost ratio, net profit/return and net profit/cost were obtained in Group III followed by that of Group IV. While the lowest were obtained in Group I (Table 5).

Table 1: Effects of different treatments of ovarian inactivity in buffalo-cows on their reproductive efficiency parameters (means±SE)

Groups	Animals No.	Animals respond to treatments	TEI (days)	TCI (days)	S/C	1st service conception rate (%)	DO (days)
Group I	15	6 (40.0%)^{c}	52.6±4.6^{a}	66.5±6.2^{a}	1.66±0.2^{a}	46.7^{b}	161.6±6.9^{a}
Group II	15	11 (73.3%)^{a}	20.5±2.9^{b}	35.9±6.2^{b}	1.73±0.2^{a}	46.7^{b}	131.9±6.5^{b}
Group III	15	12 (80.0%)^{a}	08.1±1.7^{b}	20.9±4.7^{b}	1.60±0.2^{a}	60.0^{a}	116.4±6.1^{b}
Group IV	15	9 (60.0%)^{b}	18.3±5.7^{b}	31.9±5.7^{b}	1.80±0.2^{a}	46.7^{b}	127.7±6.1^{b}
Group V	15	8 (53.3%)^{b}	41.8±5.4^{a}	60.1±7.0^{a}	1.86±0.3^{a}	46.7^{b}	157.3±7.1^{a}

Means in the same column carry different letters are significantly different ($P < 0.05$)

Table 2: Levels of plasma progesterone before treatment of buffalo-cows with ovarian inactivity and 8 days after mating (means±SE)

Groups	Animals No.	Progesterone (ng/ml)	
		Before treatment	8 days after mating
Group I	15	0.31±0.07*	3.22±0.46**
Group II	15	0.29±0.06*	2.91±0.32**
Group III	15	0.52±0.04*	2.88±0.72**
Group IV	15	0.34±0.03*	3.11±0.55**
Group V	15	0.48±0.09*	2.49±0.33**

Means in the same raw carry different * are significantly different ($P< 0.01$).

Table 3: More (+) or less (-) days open in groups of buffalo-cows treated for ovarian inactivity (The groups were in ascending order)

	Group III	Group IV	Group II	Group V	Group I
Group III	0.0	-11.3	-15.5	-40.9	-45.2
Group IV	+11.3	0.0	-4.2	-29.6	-33.9
Group II	+15.5	+4.2	0.0	-25.4	-29.7
Group V	+40.9	+29.6	+25.4	0.0	-4.3
Group I	+45.2	+33.9	+29.7	+4.3	0.0

Table 4: Returns, costs and net profit of different treatments of ovarian inactivity in buffalo-cows per each animal (By LE)

Groups	Drug cost /animal	Cost of services	Increased days open	Decreased milk production (kg)	Costs of decreased milk	Total cost	Return from life calves (at 40 days)	Net profit
Group I	00.0	16.6	45.2	361.6	904.0	920.6	1500.0	579.4
Group II	25.0	17.3	15.5	124.0	310.0	352.3	1500.0	1147.7
Group III	40.0	16.0	00.0	000.0	000.0	056.0	1500.0	1444.0
Group IV	20.0	18.0	11.3	090.4	226.0	264.0	1500.0	1236.0
Group V	15.0	18.6	40.9	327.2	818.0	851.6	1500.0	648.4

* Values represented compared to the group of shortest DO (Group III).

Table 5: Economic efficiency measures of different treatments of ovarian inactivity in buffalo-cows per each animal

Groups	Profit/cost ratio (%)	Net profit /return (%)	Net profit /cost(%)
Group I	0162.9±32.4^{c}	38.6±4.2^{c}	0062.9±005.6^{c}
Group II	0425.8±43.2^{b}	76.5±4.9^{b}	0325.8±022.1^{b}
Group III	2678.6±162.4^{a}	96.3±9.7^{a}	2578.6±134.2^{a}
Group IV	0568.2±55.6^{b}	82.4±9.5^{b}	0468.2±046.2^{b}
Group V	0176.1±23.5^{c}	43.2±6.2^{c}	0076.2±008.2^{c}

Means in the same column carrying different letters are significantly different ($P< 0.01$).

DISCUSSION

Buffaloes and cattle are considered to be the most important animals among livestock in Egypt, but infertility is widespread among these two species of animals. This infertility incurs large economic losses to animals husbandry (Afiefy, 1966). The long calving interval is one of the major problems in buffalo breeding where the interval from calving to resumption of ovarian function is longer in buffalo when compared with cattle (Barile, 2005). Egyptian National Committees (1987) observed that, low fertility rates can be attributed to many reasons which would be faced as low births rate and low milk output leading to great losses in national incomes resulting from imbalance between production and consumption.

To maintain a calving interval of 13-14 months in buffaloes, successful breeding must take place within 85-115 days after calving (Usmani, *et al.,* 2001). They added that, disturbances during this period due to delay of uterine involution or resumption of estrous activity are likely to prolong the calving interval and reduce the lifetime reproductive and productive efficiency. Also, they concluded that,

prolonged postpartum acyclicity (absence of ovarian cyclic activity) and anestrum (absence of overt estrus signs) are major sources of economic loss to buffaloes breeders.

Great efforts have been made to face the postpartum ovarian inactivity in buffaloes. As many factors interfere with the problem, treatments take many different ways, the hormonal treatment using GnRH is one important way which was represented in the present study by Group II (GnRH treated group) and Group III (progesterone and GnRH treated group).

In this study, 80% (GIII) and 73.3% (GII) of buffaloes came in estrus and became cycling while in GI (control) only 40% of buffaloes came in estrus. These results are agree with that of Borghese *et al.* (1993) who concluded that, 84.8% of anestrous buffaloes treated by progesterone and GnRH became cycling and 70.6% of that treated by GnRH only became cycling with no significant differences while none of control animals were found to be cyclic in the same period.

Group III accomplished the shortest TEI and TCI (8.1±1.7 and 20.9±4.7 days respectively). Such results assured by Singal *et al.* (1988) as they reported the role of exogenous administration of GnRH in solving anestrous problem in buffaloes. Also with that of Shah *et al.* (1990) and Ramoun *et al.* (2006) as they reported the effect of injection of GnRH in buffalo-cows in fasten the uterine involution, resumption of ovarian inactivity, shorter calving interval and high 1st service conception rate that improve the reproductive performance of buffaloes. Borghese *et al.* (1993); Uma *et al.* (1999) and Hattab and Osman (2000) recorded that, the more useful and efficacious method to induce estrus in buffaloes have been the treatment with progesterone in association with GnRH. Also, Barile (2005) concluded that, induction of estrus in buffaloes by hormonal treatments (progesterone and GnRH) proved useful in reducing the calving interval and to increase fertility. On other hand, Fateh *et al.* (1999) and Takkar *et al.* (1999) reported that, the use of GnRH given by multiple injections or in microencapsulated form, was not efficacious to induce estrus in buffaloes.

In the present work, treatment of ovarian inactivity by progesterone and estradiol benzoate (GIV) resulted in satisfactory results (60% showed estrus with 46.7% 1st service conception rate). These results are in agreement with that obtained by McDougall (2001, 2003) and Rhodes *et al.* (2003). They found that, treatment of anestrous cows by 5-6 days intravaginal progesterone and 1-2 days later by injection of 0.5-1 mg estradiol benzoate resulted in approximately 90% of cows

undergoing estrus and ovulation and 45% of inseminated cows conceiving to the first service. Also, McDougall *et al.* (2005) showed that, treatment of anestrous cows by 6-8 days intravaginal progesterone and injection of 2 mg estradiol benzoate at device insertion and 1mg estradiol benzoate 24 hrs after device removal resulted in 59.6% of inseminated cows conceiving to the first service. Xu *et al.* (2000) reported that, treatment of anestrous cows with either estradiol benzoate or GnRH at the time of insertion of progesterone device for 7-8 days resulted in an increase in conception rate compared to no estradiol benzoate or GnRH.

Complete ovarian inactivity had been attributed to a variety of external factors among them the prolonged insufficient mineral intake (Roberts, 1986). He showed that, phosphorus deficiency adversely affects fertility. In the present study, the fertility was improved after supplying the animals by phosphorus sources (Group V that treated by tonophosphane, disodium phosphate and mineral mixture). These results are in agreement with Zaki and Gohar (1961) who reported that, ovarian inactivity in buffalo-cows was successfully treated by adding phosphate in ration. Also, Ali (1992) revealed the importance of phosphorus in treatment of the ovarian inactivity in buffaloes. On the other hand, Noseir *et al.* (1987) concluded that, there was no relationship between phosphorus and postpartum ovarian inactivity in buffalo-cows. Also, El-Amrawi (1990) reported that, phosphorus had negligible effect on treatment of postpartum ovarian inactivity in buffaloes even though he successfully treated ovarian inactivity by glucose and tonophosphane.

In the present work, the lowest S/C (1.6 ± 0.2) and the highest 1st service conception rate (60%) were obtained in GIII. These results come in agreement with that obtained by Barile *et al.* (2001) who concluded that, total conception rate was 56.5% after estrus induction in buffaloes by progesterone and GnRH.

Group III had DO less than Group IV, II, V and I by 11.3, 15.5, 40.9 and 45.2 days respectively. The most important factors influencing milk production and their profitability were calving interval and persistence of milk production. In abroad studies, the total losses due to reproductive failure were reported by Dijkuizen *et al.* (1985) to be about 2% of gross production value or 10% of average farmer income. Importance of regular calving intervals in the herd is emphasized in another abroad paper by the fact that, in most herds it costs $1.25 to 1.95 per day in lost income for each day calving interval prolonged beyond 365 days in cattle (Lafi *et al.*, 1992).

Returns, cost, net profit and efficiency measures of drugs used in different treated groups of ovarian inactivity indicated that, G III gave the highest net profit per head (1444.0 L.E) and the lowest cost (56.0 L.E.) followed by G IV (1236.0 L.E. and 264.0 L.E. respectively). While, the lowest net profit and highest cost per head were obtained in G I (Table 4).

It could be concluded that, Treatment of buffalo-cows with ovarian inactivity by progesterone and GnRH gave short TEI, TCI and high net profit and economic efficiency measures.

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