

## PLACENTAL RETENTION IN COWS AND ITS EFFECT ON SUBSEQUENT FERTILITY

**Mansour, N.\*; Montasser, A. E.\*\*; Bedier, W.\*\* and Hegab, A. O.\*\***

\*Dept of Theriogenology, Fac. Vet. Med., Kafr El-Sheikh University.

\*\*Dept of Theriogenology, Fac. Vet. Med., Mansoura University.

### ABSTRACT

*This work aimed to study the effect of retained placenta on future fertility of Friesian cows.*

*In a prevention trial, 101 pregnant Friesian cows between 7-9 months with a history of retained placenta were divided into six groups G1, 18 cows received no medications (control group), G2, 19 cows infused I/V, 2 months before parturition, with Ca, G3, 15 cows injected with vit B2 30 days before parturition, G4, 15 cows injected with vitselen 30 days before parturition, G5, 19 cows injected i/m with oxytocin immediately after parturition and G6, 15 cows received PGF<sub>2α</sub> immediately after parturition.*

*In a treatment trial, 41 Friesian cows were divided into two groups. The first group, 20 cows received no medicaments (control group) and the second group including 21 cows treated by manual removal of the placenta followed by i/u insertion of oxytetracycline tablets.*

*The highest incidence of RFM (33.33%) was observed in cows over the 4<sup>th</sup> parity. Among cows showed retained placenta, 61.90% suffered from dystocia and 57.14% delivered male calves.*

*Retained placenta was 21.05, 6.67, 6.67, 26.35, 20 and 33.33% in Ca, vitamin B<sub>2</sub>, vitselen, oxytocin, PGF<sub>2α</sub> and control groups, respectively. A significant variation (P<0.001) in both uterine involution and first postpartum heat after treatment trials of retained placenta in comparison to control group was observed. On the other hand, there was a non significant increase in both days open and number of services / conception in treated group than in control one.*

*It could be concluded that, injection of Ca, vit B<sub>2</sub> or vitselen in dry period, and injection of oxytocin or PGF<sub>2α</sub> immediately after parturition, significantly reduced the incidence of RFM in Friesian cows. Manual removal of RFM plus intrauterine insertion of oxytetracycline tablets was not the most suitable treatment of RFM which could negatively affect the subsequent fertility.*

*Key words :Retained placenta, cows, vitamin B<sub>2</sub>, vitamin E with Selenium, Ca, oxytocin and PGF<sub>2α</sub>*

## INTRODUCTION

The early postpartum period, puerperium, is stressful on the dairy cows and accompanied by, multitude of metabolic and infectious disorders. Retention of fetal membranes (RFM) and metritis are the two commonly encountered postpartum problems in dairy cows (Konigsson et al., 2001).

RFM in dairy cows is defined as a failure to expel fetal membranes by 12 to 24 hours after calving (Fourichon et al., 2000).

Although RFM is an individual cow problem, the herd becomes the unit surveillance when the incidence of RFM is elevated and designs were made to undertake preventive measures (Olson et al., 1987).

The incidence of retained placenta showed great variation between different farms. In brucellosis-free area and after normal parturition, the incidence ranged from 3 to 12% while it was higher and ranged from 30 to 50% or more after abnormal calving and in brucellosis - infected herds (Grunert, 1986). Economic losses caused by RFM in dairy cattle can be classified as direct and indirect losses, the direct losses are cost of treatment, death due to severe complications and loss of milk production (Abdelhameed et al., 2009). The indirect losses include cost of treatment of chronic metritis or endometritis, reduction in the fertility, reduction in pregnancy rate, and increase number of services per conception, longer calving interval and consequently culling of cows (Bell and Roberts, 2007).

There was an increased days open with an average of  $31 \pm 9$  days in cows having RFM

than normal ones (Dubois and Williams, 1980).

Several trials were applied during the last month of pregnancy or immediately after calving to reduce the incidence of RFM, depend on optimization of prepartum immune function through injection of dry cows by immunostimulant (Vitamin B2, Vitaselin) and Calcium preparations. (Cook and Nordland, 2004).

This work aimed to study the factors affecting retention of placenta in Frisian cows, prevention trials and treatment regime of this problem with evaluation of the future fertility of these cows.

## MATERIAL AND METHODS

This study was conducted at two farms; Farskor farm, Damietta Provenance and Gamsa farm, Dakahilia Provenance, Egypt.

### Animals :

A total of 142 7 - 9 months pregnant Frisian cows were used in this study. These cows were milked twice daily (7.00 am & 7.00 pm) using a milking machine (the average daily milk yield was 20 to 27 kg/head/day). These cows were dried off 8 weeks before parturition. After complete dryness, the cows received about 2.5-3.0 kg concentrates/head/day, in addition to 25-30 kg barseem during the green season or darawa during the dry season with suitable amount of rice straw. At the beginning of the 9<sup>th</sup> m of pregnancy, concentrates were increased up to 5 kg/head/day. All animals were periodically dewormed against external and internal parasites with vaccination against endemic diseases.

**Experimental design:**

**I - Prevention trial:**

101 cows were confirmed to be pregnant between 7 - 9 months by rectal palpation. These cows had a history of retained placenta in one of the previous parturitions and were divided into six groups.

The 1<sup>st</sup> group (G1):-

Including 18 cows received no medications (control group).

The 2<sup>nd</sup> group (G2):-

In which 19 cows were i/v infused with 500ml of Ca borogluconate (Cal. D. Mag®. Phizer, Egypt) 2 months before parturition.

The 3<sup>rd</sup> group (G3):-

Including 15 cows that were injected i.m. 30 days before parturition with 40 ml of vitamin B2 (Wako. Japan).

The 4<sup>th</sup> group (G4):-

Including 15 cows that were injected i/m 30 days before parturition with 20 ml of vitamin E and selenium (VITSELEN® 15; Adwia, 10 Ramdan City, Egypt).

The 5<sup>th</sup> group (G5):-

Including 19 cows which were injected i.m. immediately after parturition with 50 i.u. (5ml) of oxytocin (Oxytocin®; Adwia, 10 Ramdan City, Egypt).

The 6<sup>th</sup> group (G6):-

Including 15 cows that were injected i.m immediately after parturition with 2.5ml PGF2\_ (Cloprostenol Sodium, Estrumate®; Schering- Plough Animal Health Coopers, Germany).

**II-Treatment trial:**

This study was conducted on 41 Friesian cows which were divided into two groups.

The 1<sup>st</sup> group :

Including 20 cows that received no treatment on retention of the placenta (control group).

The 2nd group:

Including 21 cows showed retained placenta which was removed manually followed by i/u insertion of 4 gm oxytetracycline Hcl tablets (Terramycin®; Phizer, Egypt).

Assessment of reproductive performance

All cows were rectally examined daily after parturition to assess uterine involution according to **Lowder (1993)**.

The first postpartum heat, number of services per conception., days open and conception rate were recorded for all animals.

Cases of endometritis and pyometra were recorded.

**Statistical analysis**

Data were statistically analyzed according to **Norman and Baily (1997)**.

**RESULTS**

Among 21 cows with retained placenta the cows over 4th parity represented the highest incidence of RFM (33.33%) followed by the third parity (28.57%) while the lowest incidence was observed in 4<sup>th</sup> parity cows (14.28%) as shown in Fig.(1).

Out of 142 cows, 21 showed retained pla-

centa with 13 cows (61.90%) suffered from dystocia as shown in Fig. (2).

The incidence of retained placenta was 57.14% among cows delivered male calves and 42.85% among those delivered female calves as shown in Fig.(3).

The incidence of retained placenta and the subsequent reproductive performance studied in different prevention groups were shown in table (1).

The highest incidence of retained placenta was observed in control group (33.3%), while the lowest one (6.7%) was detected in vitamin B2 and vitselen groups as shown in Fig.(4).

The highest incidence of endometritis was detected in control group (55.55%), while the lowest one was detected in vit B2 group (13.33%) as shown in Fig.(5). No cases of pyometra were recorded in all prevention groups.

The time required for complete uterine involution was significantly ( $P<0.01$ ) higher in control group ( $31.00\pm 7.69$  d) than other prevention groups ( $26.42\pm 5.64$ ,  $24.60\pm 4.70$ ,  $23.80\pm 4.48$ ,  $26.11\pm 4.91$  and  $23.00\pm 3.59$  for Ca, vit B2, Vitselen, oxytocin and PGF2<sub>2</sub> groups, respectively).

A significantly ( $P<0.01$ ) increased days to first postpartum heat was observed in control ( $58.67\pm 21.23$ ) and Ca ( $58.58\pm 22.73$ ) than other prevention groups ( $39.53\pm 15.80$ ,  $40.07\pm 15.08$ ,  $42.95\pm 18.62$  and  $41.53\pm 15.61$  in vit B2, Vitselen, oxytocin and PGF2<sub>2</sub> groups, respectively).

On the other hand, the average days open was significantly ( $P<0.01$ ) increased for control ( $151.22\pm 58.57$ ) when compared to other prevention groups ( $111.16\pm 35.57$ ,  $107.47\pm 47.99$ ,  $96.80\pm 22.00$ ,  $103.63\pm 28.66$  and  $107.93\pm 30.01$  in Ca, vit B2, Vitselen, oxytocin and PGF2<sub>2</sub> groups respectively),

A significantly ( $P<0.05$ ) higher number of service per conception was recorded in control ( $3.33\pm 1.57$ ) and vit B2 ( $3.13\pm 2.17$ ) groups than other prevention groups ( $2.00\pm 1.00$ ,  $1.93\pm 1.16$ ,  $2.11\pm 1.15$ ,  $2.20\pm 1.42$ , respectively in Ca, Vitselen, oxytocin and PGF2<sub>2</sub> groups).

Table (2) revealed a significantly ( $P<0.001$ ) more days to uterine involution ( $31.57\pm 5.78$ ) and first postpartum heat ( $58.57\pm 15.01$ ) after treatment of retained placenta in comparison to control group ( $24.00\pm 3.66$  and  $36.25\pm 15.44$ , respectively for uterine involution and first postpartum heat). On the other hand, there was a non significant increase in both days open and number of services / conception in treated group than control one.

## **DISCUSSION**

Results of the present study revealed that, the overall incidence of RFM among 142 Friesian cows was 14.79%. This result was nearly similar to that obtained by **Nasr et al., (1993)**, who found that, the average incidence of retained placenta in Friesian cows was 12.37%. Lower incidence 7.5% was reported by **Grunert (1980)**, while, higher incidence (25%, 20.6% and 17.7%) were reported by **Chassagne and Brochart (1985) and Mutiga et al. (1993)** respectively. Differences in the incidence of RFM might be attributed to the

breed (**Roberts, 1971**) as well as nutritional, environmental and managemental factors (**Grunert, 1980**).

The present study revealed that, the incidence of RFM was 33.33% in cows with parity over fourth. This result was confirmed by that obtained by **Arthur et al., (1989)** who mentioned that, the higher tendency of RFM in the old cows may be attributed to uterine inertia as a result of decreased serum calcium concentration.

The obtained data of the present study indicated that the incidence of RFM among cows producing male calves was higher than that producing female ones. This result agreed with that obtained by **Siani et al. (1988)** that might be attributed to the increased birth weight of male calves. However, **Mutiga et al. (1993)** found no correlation between fetal sex and the incidence of RFM.

Results of the present study indicated that, the incidence of retained placenta in cows suffered from dystocia was (61.9%). These findings came in accordance with those obtained by **Han-Yeonkyung and Kim-Ilhwa (2005) and Lombard et al. (2007)**. Higher incidence of retained placenta after dystocia might be attributed to the bad interference during the birth help which delay the release of the placenta (**Nasr et al., 1993**). Trauma during dystocia to myometrial tissue, or inadequate release of PGE, PGF<sub>2</sub>, oxytocin and relaxin act as multifactorial agents (**EchlernKamp and Gregory, 1999**).

In our study, the prepartum injection of se-

lenium and vitamin E during the last month of pregnancy significantly reduced the incidence of retained placenta to 6.67% in cows compared to the control (33.33%). These results agreed with those obtained by **Hattab and Abdel Moghney (1994)** who reported that, only the lowest dose of selenium (23mg) produced significant reduction in the incidence of retained placenta.

The reduction in the incidence of RFM due to vitamin E supplementation might be attributed to enhancement of immune system, thus encourage tissue separation as vitamin E enhances migration and chemotaxis of polymorphonuclear cells (**Ndiweni and Finch, 1996**) and increased chemotaxis responsiveness by blood neutrophils (**Politis et al., 1996**). At the same time, vitamin E play an important role in increasing activity of glutathione peroxidase and enhance immunity in the uterus and placenta so aid in separation of the placenta (**Kolb and Seehawer, 2002**). Moreover vitamin E enhanced the antioxidant function (**Wilde, 2006**).

Selenium and vitamin E injection during the last month of pregnancy significantly affect the subsequent fertility, where it reduced the period to the 1<sup>st</sup> P.P heat and the number of services/conception. This result came in accordance with that obtained by **Bourne et al. (2006)**.

The present study revealed that, injection of 5mg/kg of vitamin B2 15-30 days prior to the expected date of calving resulted in a significant reduction in placental retention. This result agreed with **Zaabel (2003)**.

Vitamin B2 injection during the last month of pregnancy significantly improved the subsequent fertility as the time required to uterine involution, 1<sup>st</sup> P. P. heat and days open in comparison to the control group. These results agreed with those obtained by **Zaabel (2003)**. The improvement in fertility might be attributed to that vitamin B2 enhances the non specific host defense mechanism against the variety of bacterial infections in cattle by stimulating the generation of neutrophils leading to increased uterine immunity (**Osman et al., 1995**).

Also, Ca administration during last two months of pregnancy significantly reduced the incidence of retained placenta compared to control ones. Similar results were obtained by **Cho-JinHaeng et al. (2005)**. Our study revealed that calcium infusion as a preventive therapy for RFM lead to a significant decrease in time required for uterine involution ( $26.42 \pm 5.65$ ) days, days open ( $111.16 \pm 33.57$ ) and number of services /conception ( $2.00 \pm 1.00$ ) in comparison to control one ( $31.00 \pm 7.69$ ,  $151.22 \pm 58.75$  and  $3.33 \pm 1.57$ , respectively). This was supported by **Twafik and El-Bakhmy (2008)**, who emphasized that cows with RFM have lower Ca concentration in their blood resulting in reduction in myometrial activity during parturition which induce placental retention.

Significant decrease in the incidence of RFM after oxytocin injection. came in accordance with **Mollo et al. (1997)**. oxytocin stimulates the myometrial activity and enhances the expulsion of the fetus and placenta (**Russe, 1979**).

The reduction in the incidence of RFM after injection of PGF2<sub>α</sub> agreed with **Santos et al. (2002)** and **Hamali and Rezazadeh (2008)**. In contrast, **Stevens and Dinsmore (1997)** reported that, there was a non significant effect of PGF2<sub>α</sub> on subsequent fertility and the reduction of placental retention in cows.

The significant reduction in the incidence of RFM in cows might be attributed to PGF2<sub>α</sub> play a facilitator role in the dropping of the placenta though enhancing uterine contraction and/or may have stimulatory effect on phagocytosis by uterine leukocytes (**El-Azab et al., 1988**).

In our study, manual removal of placenta followed by intrauterine insertion of oxytetracycline lead to impaired fertility. This result agreed with **Laven and Peters (1996)**. However, **Drillich et al. (2003)**; **Mordak (2007)**; **Hamali and Karimi (2008)** reported that, manual removal of retained fetal membranes was advisable, effective and didn't impair fertility.

The significant increase of days to uterine involution and days to first P.P. heat after manual removal of retained fetal membranes might be attributed to intrauterine trauma and interference with uterine defense mechanism followed by bacterial invasion which might result in endometritis with subsequent septicemia or toxemia. Also there was a direct effect of uterine environment on ovarian activity (**Bolinder et al., 1988**).

It could be concluded that, prepartum injection of Ca, vit B2 and vit selen or postpartum injection of oxytocin or PGF2<sub>α</sub> might

reduce the incidence of RFM and improve the reproductive performance in cows. Manual removal of the placenta with intrauterine inser-

tion of antibiotics might not be the most suitable treatment of RFM with a negative effect on subsequent fertility.

**Table (1):** Postpartum reproductive performance of prophylactic groups in Friesian cows.

Parameter Groups	No. of cows with R.F.M. (%)	No. of cows with endometritis (%)	Uterine involution by days (mean±SD)	1st p.p heat by days (mean±SD)	Days open (mean±SD)	No. Service/conception (mean±SD)
Control group (n= 18)	6 (33.33%)	10 (55.55%)	31.00±7.69 <sup>a</sup>	58.67±21.23 <sup>a</sup>	151.22±58.75 <sup>a</sup>	3.33±1.57 <sup>A</sup>
Calcium group (n=19)	4 (21.05%)	7 (36.84%)	26.42±5.64 <sup>b</sup>	58.58±22.73 <sup>a</sup>	111.16±35.57 <sup>b</sup>	2.00±1.00 <sup>B</sup>
Vitamin B2 group (n=15)	1 (6.67%)	2 (13.33%)	24.60±4.70 <sup>b</sup>	39.53±15.80 <sup>b</sup>	107.47±47.99 <sup>b</sup>	3.13±2.17 <sup>A</sup>
Vitamin E & Se group (n=15)	1 (6.67%)	4 (26.67%)	23.80±4.48 <sup>b</sup>	40.07±15.08 <sup>b</sup>	96.80±22.00 <sup>b</sup>	1.93±1.16 <sup>B</sup>
Oxytocin group ( n=19)	5 (26.35%)	5 (26.31%)	26.11±4.91 <sup>b</sup>	42.95±18.62 <sup>b</sup>	103.63±28.66 <sup>b</sup>	2.11±1.15 <sup>B</sup>
PGF2 $\alpha$ group (n=15)	3 (20%)	4 (26.67%)	23.00±3.59 <sup>b</sup>	41.53±15.61 <sup>b</sup>	107.93±30.01 <sup>b</sup>	2.20±1.42 <sup>B</sup>

Different small letters within columns denote significant variation at  $P < 0.01$ .

Different capital letters within columns denote significant variation at  $P < 0.05$ .



**Table (2):** Reproductive performance after treatment trials of retained placenta in Frisian cows.

<b>Group</b>	<b>Uterine involution (Mean ± SD)</b>	<b>1<sup>st</sup> P.P. heat (Mean ± SD)</b>	<b>Days open (Mean ± SD)</b>	<b>Service/conception (Mean ± SD)</b>
<b>Control group (n=20)</b>	<b>24.00±3.66<sup>a</sup></b>	<b>36.25±15.44<sup>a</sup></b>	<b>107.75±47.75<sup>a</sup></b>	<b>2.55±1.50<sup>a</sup></b>
<b>Treatment group (n=21)</b>	<b>31.57±5.78<sup>b</sup></b>	<b>58.57±15.01<sup>b</sup></b>	<b>135.38±43.70<sup>a</sup></b>	<b>3.10±1.26<sup>a</sup></b>

Different letters within column denote significant variation at  $P < 0.001$

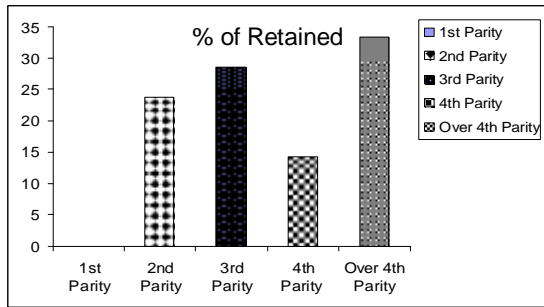


Fig (1): Effect of parity on the incidence of retained placenta in cows.

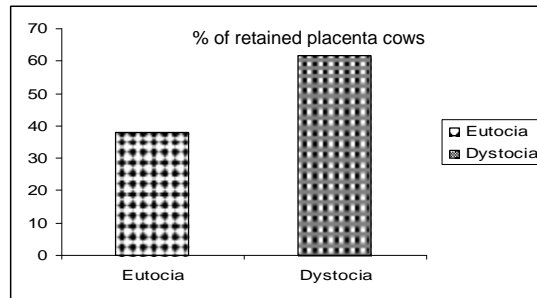


Fig (2): Effect of nature of parturition on the incidence of retained placenta in cows.

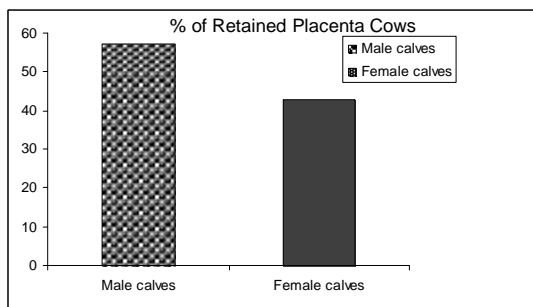


Fig (3): Effect of sex of calves on the incidence of retained placenta in cows.

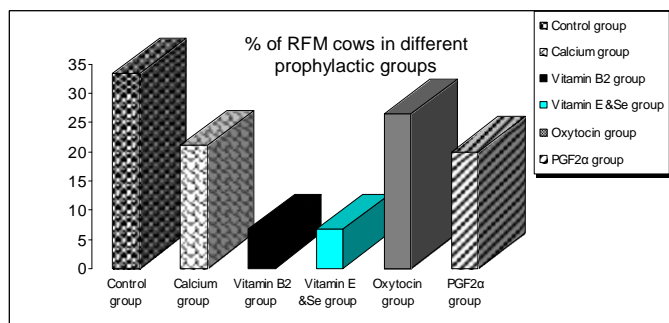


Fig (4) : Percent of retained placenta in different prevention groups.

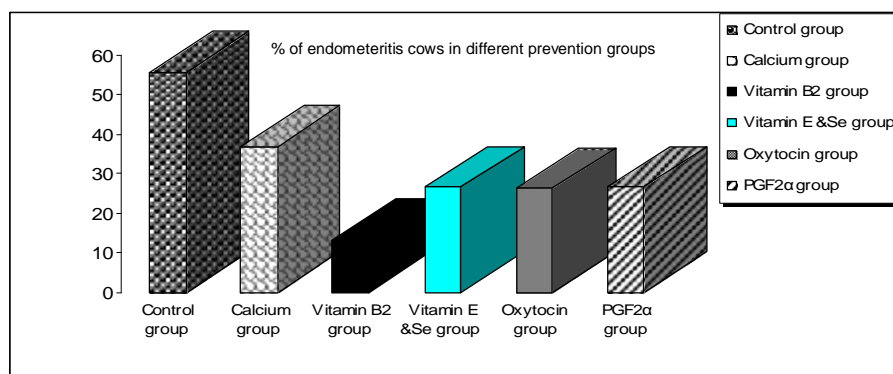


Fig (5): Percent of endometritis in different prevention groups.

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## الملخص العربى

### الاحتباس المشيمى فى الأبقار وتأثيره على الخصوبة اللاحقة

د/نبيل عبد الحميد منصور\* د/عبد المنعم منتصر محمود\*\*

د/وائل السيد بدير\*\* د/عبدالرؤف عثمان حجاب\*\*

قسم التوليد والتناسل والتلقيح الاصطناعى - كلية الطب البيطرى - جامعة كفر الشيخ\*

قسم التوليد والتناسل والتلقيح الاصطناعى - كلية الطب البيطرى - جامعة المنصورة\*\*

هذه الدراسة تهدف إلى دراسة ظاهرة الاحتباس المشيمى وتأثيرها على الخصوبة اللاحقة فى الأبقار الفريزيان، وقسمت هذه الدراسة إلى جزئين :-

#### أولاً : دراسة طرق الوقاية :

أجريت هذه الدراسة على (101) مائة وواحد بقرة فريزيان عشار ما بين 7 إلى 9 أشهر، وكانت هذه الأبقار تعاني من احتباس مشيمى فى واحدة من الولادات السابقة وقد تم تقسيم الحيوانات فى هذه الدراسة إلى 6 مجموعات :

- ١- المجموعة الأولى : وتتضمنت 18 بقرة فريزيان لم يحقنوا أى دواء واستخدمت كمجموعة ضابطة.
- ٢- المجموعة الثانية : وتتضمنت 19 بقرة فريزيان تم حقن كل حيوان 500 مل كالسيوم وريد قبل الولادة بشهرين.
- ٣- المجموعة الثالثة : وتتضمنت 15 بقرة فريزيان تم حقن كل حيوان 40 مل فيتامين ب<sub>2</sub> فى العضل قبل الولادة ب30 يوم.
- ٤- المجموعة الرابعة : وتتضمنت 15 بقرة فريزيان تم حقن كل حيوان 40 مل فيتامين هـ وسيلينيوم فى العضل قبل الولادة ب15 يوم.
- ٥- المجموعة الخامسة : وتتضمنت 15 بقرة فريزيان تم حقن كل حيوان 5 مل أوكسيتوسين فى العضل بعد الولادة مباشرة بساعتين.
- ٦- المجموعة السادسة : وتتضمنت 15 بقرة فريزيان تم حقن كل حيوان 2.5 مل البروستاجلاندين اف<sub>2</sub> ألفا فى العضل بعد الولادة مباشرة بساعتين.

#### ثانياً : دراسة طرق العلاج :

أجريت هذه الدراسة على 41 بقرة فريزيان وقد قسمت إلى مجموعتين :

- ١- المجموعة الأولى : تضمنت الأبقار التى لاتعاني من احتباس المشيمة وقد استخدمت كمجموعة ضابطة وتشمل 20 بقرة فريزيان.
- ٢- المجموعة الثانية : تضمنت 21 بقرة فريزيان تعاني من احتباس المشيمة والتى تم إزالة المشيمة فيها يدوياً مع وضع 4 جم من أقراص الأوكسى تتراسيكلين داخل رحم الأبقار وتشمل 21 بقرة فريزيان.

وكان أعلى معدل حدوث الاحتباس المشيمي فى الأبقار التى ولدت أكثر من (4) أربع مرات 33.33%. وكانت نسبة احتباس المشيمة فى الأبقار التى عانت من عسر ولادة (61.90%)، وأيضاً أدت ذكور الأجنة إلى ارتفاع معدل احتباس المشيمة فى الأبقار إلى (57.14%). احتباس المشيمة كان 21.05, 6.67, 6.67, 26.35, 20, 33.33% فى الكالسيوم، فيتامين ب<sub>2</sub>، فيتاسيلين، أوكسيتوسين، البروستاجلاندين أف<sub>2</sub> ألفا والمجموعة الضابطة على التوالى، كان هناك فارق معنوى فى كل من التنفاف الرحم والشبق الأول بعد الولادة فى المجموعة المعالجة مقارنة بالمجموعة الضابطة، من ناحية أخرى، كانت هناك زيادة غير معنوية فى الفترة من الولادة حتى حدوث الحمل وكذلك فى عدد التلقيحات اللازمة حتى حدوث الحمل فى مجموعات العلاج مقارنة بالمجموعة الضابطة. ويمكن أن نخلص إلى أن حقن الكالسيوم، فيتامين ب<sub>2</sub>، فيتاسيلين فى فترة الجفاف، وحقن هرمون أوكسيتوسين، البروستاجلاندين أف<sub>2</sub> ألفا مباشرة بعد الولادة أدى إلى تقليل معنوى فى معدل الاحتباس المشيمي فى الأبقار الفريزيان، إزالة المشيمة يدوياً ثم وضع أقراص الأوكسى تتراسكلين فى الرحم ليست هى العلاج الأنسب كما أنها تؤثر سلباً على الخصوبة اللاحقة.

**الكلمات المفتاحية :** احتباس المشيمي - الأبقار - فيتامين ب<sub>2</sub> - فيتامين هـ وسيلينيوم - كالسيوم - بروستاجلاندين أف<sub>2</sub> ألفا - أوكستوسين.