

Bacterial study of vagina in Awassi ewes treated with Prostaglandin (PGF2 α) and oxytocin

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Abstract

This study was carried on 21 Awassi ewes postpartum, the age of the animals ranged between 3-5 years. The animals were divided in to 3 groups (each group contain 7 animals). The first group was treated with 7.5 mg of prostaglandin F2 α . The second group was treated with 20 IU oxytocin the third group was considered as a control group and was treated with 2 ml of normal saline. Bacterial swabs were taken from the vagina. The result revealed that there were 6 different types of bacteria, the predominant bacteria in the vagina of ewes were *E.coli*, and *Staphylococcus aureus* while *Pseudomonas aeruginosa* and *Proteus vulgaris* were less frequent isolates. The result showed that the number of all species of bacteria in PGF2 α treated group less than in other groups. There were a significant differences (p<005) between the first group and the other two groups, While the number of vaginal bacteria did not affected in oxytocin treated group. It was concluded from this study that treatment of ewes with PGF2 α postpartum reduce the bacterial flora of the vagina and metritis keeping of the ewes under non stressful condition is warranted to avoid clinical infection of reproductive tract with opportunistic bacteria.

Keyword: uterine involution, PGF2 α , vaginal flora, Awassi ewes.

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دراسة جرثومية على مهابل النعاج العواسية المحقونة بالبروستاكلاندين PGF2 α

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الخلاصة

أجريت الدراسة الحالية على 21 من النعاج العواسية. قسمت الحيوانات الى ثلاثة مجاميع كل مجموعة تتكون من (7) نعاج، عولجت المجموعة الأولى 7.5 ملغم PGF2 α بالعضل المجموعة الثانية عولجت 20 وحدة دولية بالاوكسيتوسين بالعضل اما المجموعة الثالثة اعتبرت كمجموعة سيطرة فقد تم حقنها 2 مل من محلول الملح الفسلجي. أخذت المسحات الجرثومية من مهابل الاناث. اظهرت النتائج ان هناك 6 انواع مختلفة من البكتريا هي السائدة في مهابل النعاج وهي *Staphylococcus aureus* و *E. coli* بينما *Proteus vulgaris* و *Pseudomonas aeruginosa* اقل العزلات تكررًا. وقد اظهرت النتائج ايضا ان اعداد جميع انواع البكتريا في مجموعة النعاج العواسية المعالجة PGF2 α كانت اقل من المجاميع الأخرى. وقد لوحظ ان ان حقن النعاج PGF2 α اختزل اعداد البكتريا الى ما يقارب 32% مقارنةً بمجموعة السيطرة بينما لم تتأثر اعداد البكتريا المتواجدة في مهابل النعاج المحقونة بهرمون الاوكسيتوسين. وقد استنتج من الدراسة ان المعالجة بالبروستاكلاندين F2 α بعد الولادة يختزل اعداد الجراثيم المتواجدة في المهبل ويقلل من الاصابة بالتهاب الرحم. كلمات مفتاحية: ارتداد الرحم، بروتستاكلاندين PGF2 α ، فلورا المهبل، النعاج العواسي.

Introduction

Postpartum infections, can reduce reproductive efficiency of economic animals. Both incidence and consequences of infections are documented far more extensively for dairy cattle than for, goat, or sheep. However, circumstances that increased risk of genital infections in dairy cattle, such as assisted births, dystocia, retained fetal membranes and unsanitary conditions at parturition, are common in sheep and predispose them to uterine infections in most sheep and cattle, the uterus able to prevent bacteria that typically reside in the postpartum uterus from proliferating and cause infections. A short exposure to luteal or exogenous progesterone will down regulate immune functions (1). Similarly, PGF₂ α is known for uterotonic effect and stimulation of phagocytosis by uterine leukocytes (2). Vaginal infection with pathogenic bacteria can reduce reproductive efficiency of ewes. These bacteria related to disease due to reduction of immunity of reproductive system and information regarding the use of antibiotics in the control of reproductive diseases in ewes is inadequate (3). A variety of bacteria have been isolated from female genitalia in ewes including *E. coli*, *Staphylococcus aureus*, *Streptococcus spp.*, *Staphylococcus epidermidis*, *Proteus mirabilis* (4). (5) Have been isolated *E. coli*, *Klebsiella*, *Salmonella*, *Staphylococcus aureus*, *Proteus*, *Micrococcus*, *Streptococcus Spp.*, *Staphylococcus epidermid* and *Pseudomonas* from the ewe genital tract. These bacteria may cause genital infection that leads to reproductive failure (6). Hence, the present study was aimed to know the prevalence of vaginal bacteria in postpartum awassi ewes treated with prostaglandin F₂ α and oxytocin.

Materials and Methods

- **Experimental animals and sample collection:** Twenty one Iraqi Awassi ewes were divided in to three groups after parturition. Each group contains 7 animals, the age of the animals ranged between 3-5 years. The animals were presented in the farm of collage of veterinary Medicine University of Fallujah. The first group was received with 7.5 mg of prostaglandin F₂ α . The second group was received with 20 IU of oxytocin the third group received placebo treatment of 2 ml of normal saline and considered as a control group. The swabs were taken from the vagina after 7, 14, 21 and 28 day postpartum.
- **Culture media for isolation:** Different types of media were used consisted of 7% Sheep blood agar, Nutrient agar, MacConkey agar, Brain heart infusion agar and Eosin methylene blue agar for identification of *E. coli*. Media were prepared according to manufacturer's instructions (7).
- **Inoculation of culture:** Vaginal swabs were used to inoculate the plate. Plates and the material were streaked with a bacteriological loop for 5 dilutions of the inoculums and incubated under aerobic condition at 37C for 24 hours.
- **Identification of bacteria:** The bacterial isolate were identified on their culture by biochemical and morphological characters. To study the cultural characteristics, discrete the colonies on the agar surface were observed: the shape, size, consistently, color and pigment production. The cellular morphology of isolates were observed by staining the isolates with gram stain and examined microscopically. The biochemical tests include: catalase, oxidase, IMVC test (indol production, methyl red, vogas-proskauer and citrate utilization), TSI (triple sugar iron), coagulase test, urease production, gelatine liquefaction, hemolysis test on sheep blood agar and different carbohydrates utilization. The isolation and identification of bacteria was done according to (8).

Results and Discussion

The total samples were 84 swabs, 73 (87%) were positive and 11 (13%) were negative results for bacterial culture in the three groups. The results of the bacteriological examination for 21 ewes of each group are shown in table (3).

Table (1) Showed the number of vaginal swabs isolates

Animals group	Total No. of specimens	Positive specimens	Prevalence
Ewes injected with PGF2 α	28	18	64.3%
Ewes injected with Oxytocin	28	27	96.4%
Control	28	28	100%
Total	84	73	

The result showed there were 6 different types of bacteria, the predominant bacteria in the ewes were *Staphylococcus aureus*, and *E.coli* while *Proteus vulgaris* and *Pseudomonas aeruginosa* were less frequent isolates. Table (2) showed the bacterial species which isolated from vagina of ewes and of their presence in all groups of the study. These results did not agree with other workers (8, 10) whom reported that *Actinomyces*, *klebsiella*, *Staphylococcus* were the most prevalent bacteria presence in sheep (24.1, 19, 13.8)% respectively.

Table (2) Bacterial species isolated from ewes vaginal swabs and its percentage

Bacteria	PGF2 α group		Oxytocin group		Control group	
	No.	(%)	No.	(%)	No.	(%)
<i>Staphylococcus aureus</i>	5	28%	8	30%	7	25%
<i>Escherichia coli</i>	4	22%	5	18.4%	6	21.4%
<i>Streptococcus spp.</i>	3	16.7%	3	11%	4	14.3%
<i>Klebsilla spp.</i>	2	11.1%	3	11%	3	10.7%
<i>Protus vulgaris</i>	2	11.1%	4	14.8%	4	14.3%
<i>Pseudomonas aeruginosa</i>	2	11.1%	4	14.8%	4	14.3%
Total	18	100%	27	100%	28	100%

The *Staphylococcus aureus*, *Escherichia coli* and *Streptococci*, they could have been derived from fecal contamination of the vagina, which contrast with the of study in Sudanese sheep and goats (6). Bacteria colonizing the vagina are likely to cause reproductive failure in domestic ruminants and ewes. Vaginal bacteria get access into the uterus during the peripartum period leading to endometritis and metritis and subsequent reduction in the reproductive capacities of these animals (11). Therefore, it is important to identify these bacteria with the view of providing remedial intervention that will restore fertility. The results indicated that there were several bacterial species presented in the ewe vagina. This confirms the results of previous studies to contain the reproductive tract of ewe on commensalism natural bacteria without affecting the reproductive function. This findings agreed with several investigators (8, 9, 12, 13, 14). Table (2) showed that the numbers of all species of bacteria in PGF2 α treated group less than other groups. It has been observed that ewes treated with PGF2 α caused a significant reduction in bacterial isolates of vagina and conception approximately 32%, these findings were in agreement the findings of (15). In fact, the exogenous PGF2 α removes the suppressive effect of progesterone on the uterine defense mechanism or, alternatively stimulate it through estrogen secretion (16). Additionally, PGF2 α may have stimulatory effect on phagocytic activity of polymorphonuclear leukocytes (17). It has been also, have been observed that the number of vaginal bacteria did not affect with treatment of oxytocin; therefore, the oxytocin might have not stimulatory effect on immune system to resistance the bacterial infection. It was concluded from this study that the bacteriological findings should reflect the "normal" flora of the reproductive

tract of the ewe postpartum. Treatment with PGF 2α postpartum could reduce the bacterial flora of vagina and metritis.

References

1. Lewis, G. S. (2003). Role of ovarian progesterone and potential role of prostaglandin F 2α and prostaglandin E 2 in modulating the uterine response to infection bacterial in postpartum ewes. *J. Anim. Sci.*, 81: 285-293.
2. Steefan, J.; Agric, M.; Adriamanga, S. & Thibier, M. (1984). Treatment of metritis with antibiotics or prostaglandin F 2α and influence of ovarian cyclicity in dairy cow. *Am. J. Vet.*, 45: 1090-1094.
3. Martins, G.; Figueira, L.; Penna, B.; Brandao, F.; Varges, R.; Vasconcelos, C. & Lilenbaum, W. (2009). Prevalence and antimicrobial susceptibility of vaginal bacteria from ewes treated with progestin-impregnated intravaginal sponges. *Small Rumin. Res.*, 81 (2-3): 182-184.
4. Al-Sariy, S. M. (2014). Isolation and identification of some aerobic bacterial flora from female genitalia in goats in Babylon city. *J. Agri. Vet. Sci.*, 7(10): 19-22.
5. Al-Zubaidi, S. F.; Hasson, S. O. & Ajeel, H. H. (2013). Isolation and identification of microflora species at different levels of the ewe genital tract. *J. Agri. Vet. Sci.*, 6(3): 54-57.
6. Shallali, A. A.; Hussein, A. M.; Salih, M. M. & Dafalla, E. A. (2001). A preliminary report on bacteria isolated from the female genital tract of Sudanese sheep and goats. *The Sudan J. Vet. Res.*, (17): 55-63.
7. Forbes, B. A.; Sahm, D. F. & Weissfeld, A. S. (2007). *Bailey and Scotts' Diagnostic microbiology*. 12th ed. Elsevier.
8. Sneath, P. A.; Mair, N. S.; Sharp, M. E. & Hott, J. G. (1986). *Bergey's Manual of systematic Bacteriology*. William and Wilkins, USA.
9. Aziz, D. M.; Al-Sultan, M. A. H. & Al-Jawally, E. A. K. (2000). Uterine microflora in Awassi ewes. *Iraqi J. Vet. Sci.*, 13: 201-205.
10. Al-Delemi, D. H. J. (2005). The normal bacterial flora in the vaginal cavity of Iraqi cows, sheep, goats and camels during the luteal phase. *Al-Qadisiya J. Vet. Med. Sci.*, 4: 23-29.
11. Levinson, W. E. & Jawetz, E. (1994). *Medical Microbiology and Immunology*. Third edition. Prentice- Hall Int. Inc., Englewood Cliffs, New Jersey-USA. PP. 20-23.
12. Azawi, O. I.; Ali, J. B. & Ali, D. S. (1995). Non-specific genital tract microflora of Iraqi local breed cows. *Iraqi J. of Vet. Sci.*, 8 (2): 261-265.
13. Al-Hilali, H. A. & Al-Delemi, D. H. (2001). The uterine bacterial flora of normal reproductive tract, non-pregnant Iraqi cows. *The Veterinarian*, 11(1):112-120.
14. Al-Delimi, D. H. J. (2002). The uterine bacterial flora of reproductive tract of the Iraqi she-camels. *Al-Qadisiya J. Vet. Med. Sci.*, 1 (2): 55-59.
15. Sarkar, P.; Rawat, M.; Varshney, V.; Goswami, T.; Yadav, M. & Srivastava, S. (2006). Effect of administration of garlic extract and PGF 2α on hormonal changes and recovery in endometritis cows. *Asian- Aust. J. Anim. Sci.*, 19 (7): 964-969.
16. Lindell, J. & Kindahl, H. (1983). Exogenous prostaglandin F 2α promotes uterine involution in the cow. *Acta veterinary scandinavies* 24: 269-270.
17. Paisley, L.; Mickelsen, W. & Anderson, P. (1986). Mechanisms and therapy for retained fetal membranes and uterine infection of cow. A review. *Theriogenology*, 25(3): 353-381.