Effect of hormonal injection on bacterial content in uterus of postpartum in Iraqi ewes

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Abstract

The present study was carried on 21 ewes divided in to three groups: 7 ewes injected with prostaglandin F2α, 7 ewes injected with oxytocin and 7 ewes injected with distal water used as control group. Bacterial swabs taken from the female uterus. The result revealed that there were 6 different types of bacteria, the predominant bacteria in the uterus ewes were *Staphylococcus aureus* 32.8%, and *Escherichia coli* 22% while *Pseudomonas aeruginosa, Streptococcus spp., Klebsilla spp.* and *Proteus vulgaris*, were less frequent isolates (15, 12.4, 12.4, 5.4)% respectively. Also the result showed that the number of all species of bacteria in PGF2α group less than other groups. The observation that injected ewes with PGF2α caused a significant reduction of bacterial load in uterus compared with control group while number of uterus bacteria did not affect with injection of oxytocin which might have not stimulatory effect on immune system to resistance the bacterial infections.

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Key words: Bacterial infection, postpartum ewes, uterus, PGF2α, oxytocin.

تأثير حقن الهرمونات على المحتوى الجرثومي في رحم النعاج العراقية بعد الولادة

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الخلاصة

أجريت الدراسة الحالية على 21 من النعاج قسمت إلى ثلاث مجاميع: 7 نعاج محقونة بالهرمون PGF2α، 7 من النعاج محقونة بالهرمون الاوكسيتوسين و7 من النعاج محقونة بالماء المقطر استخدمت كمجموعة سيطرة. أخذت المسحات الجرثومية من أرحام الإناث. أكدت النتائج ان هناك 6 أنواع مختلفة من البكتريا هي السائدة في أرحام النعاج وهي *Staphylococcus aureus* (32.8%) و *Escherichia coli* (22%) بينما *Klebsilla spp., Streptococcus spp., Pseudomonas aeruginosa* و *Proteus vulgaris* اقل العزلات تكراراً (15، 12.4، 12.4، 5.4)% على التوالي. أيضا أظهرت النتائج ان أعداد جميع أنواع البكتريا في مجموعة النعاج العواسية المحقونة بالهرمون PGF2α كانت اقل من المجاميع الأخرى. من الملاحظ ان حقن النعاج بالهرمون PGF2α اختزل أعداد البكتريا مقارنةً بمجموعة السيطرة بينما لم تتأثر أعداد البكتريا المتواجدة في أرحام النعاج المحقونة بهرمون الاوكسيتوسين والذي ربما لم يكن له تأثير تحفيزي للنظام المناعي في مقاومة الإصابات البكتيرية.

الكلمات المفتاحية: التأثير الجرثومي، النعاج بعد الولادة، الرحم، البروستكلاندين، الاوكسيتوسين.

Introduction

Ovarian activity during the early postpartum period exerts an important influence on the ability of the uterus to resist or eliminate bacterial infections. The (1) have stated that, the postpartum of ruminants can resist uterine infection during the estrogenic phase but very susceptible during the progesterone phase, which is due to decreased leucocytic activity. The (2) reported an increased concentration of immunosuppressant protein in the uterine lumen under progesterone dominance, which inhibits lymphocyte proliferation, making the uterus more susceptible to infection. Postpartum uterine

infections, which are usually nonspecific, can reduce reproductive efficiency of ruminant livestock (3). Who is also reported that the Progesterone makes the postpartum uterus in ewes susceptible to infection, but ovariectomy allows ewes to remain resistant; uterine prostaglandins may mediate this change. A variety of bacteria have been isolated from female genitalia in ewes including *E. coli, Staphylococcus spp., Streptococcus spp., Proteus and Bacillus* (4) and the researcher (5) have been isolate *Escherichia coli, Salmonella, Klebsiella, Staphylococcus aureus, Streptococcus Spp., Proteus, Micrococcus Staphylococcus epidermid* and *Pseudomonas* from the ewe genital tract. These bacteria may cause genital infection that usually leads to reproductive failure in ewes (6). However, circumstances associated with increased risk of genital infections in dairy cattle, such as dystocia, assisted births, retained fetal membranes, and unsanitary conditions at parturition, are common in sheep and predispose them to uterine infections In most cattle and sheep, the uterus seems to be able to prevent bacteria that typically reside in the postpartum uterus from proliferating and creating infections A short exposure to luteal or exogenous progesterone will down regulate immune functions (3). Similarly, PGF2α is known for uterotonic effect and stimulation of phagocytosis by uterine leukocytes (7). Progesterone seems to be the primary ovarian steroid that governs the ability of the uterus to resist infections. The progesterone typically down-regulates immune functions and makes the uterus susceptible to infections (8), and reported that it is important to determine how uterine PGF2α is able to stimulate the uterus to resolve infections, even when progestrone concentrations are increased, should be important to scientists and clinicians working to understand the underlying causes of uterine infections. information from that line of research should yield important new prevention and treatment strategies for uterine infections that do not rely on antibiotic and antimicrobial compounds. Hence, the present experiment was conducted to study prevalence of uterus bacteria in postpartum Awassi ewes injected with prostaglandin F2α and oxytocin.

Materials and Methods

- **Samples:** Twenty-one ewes were divided in to three groups: 7 ewes injected with 7.5 mg I.M of prostaglandin F2α, 7 ewes were injected with 20 IU of oxytosin postpartum and 7 ewes were injected with distal water used as control does were examined and uterus swabs were taken for bacteriological studies during 2015. The swabs taken postpartum of (0, 7, 21, 28) days from uterus injected of ewes with PGF2α group and uterus injected of ewes with oxytosin group, for four times.
- **Bacteriological materials and methods and reagent:** In this study used different types of media consisted of 7% Sheep blood agar, Nutrient agar, MacConkey agar, Brain heart infusion agar and Eosin methylene blue agar for identification of *Escherichia coli*. Media were prepared according to manufacturer's instructions.
- **Inoculation of culture:** Uterus swabs were used to inoculate the cultures media and the material was streaked with a bacteriological loop for 5 dilutions of the inoculums. These plates were incubated in aerobic condition at 37C$^{\circ}$ for 24 hours after incubation period the findings were recorded.
- **Identification of bacteria:** The bacteria isolate were identified their culture, morphological and biochemical characters. To study the cultural characteristics, discrete the colonies on the agar surface were observed: the shape, size, cosistenty, 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, gelatein liquefaction, hemolysis test on sheep blood agar and different carbohydrates utilization. The isolation and identification of bacteria was done by using analysis to the methods of (9).

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 included ewes injected with PGF2α group, ewes injected with oxytosin group and ewes injected with distal water used as control group. The result of the bacteriological examination for 28 ewes of each group shown in table (1).

Table (1) Numbers of uterus swabs of ewes which gave positive results for bacterial inoculation on different media

Animals	Total No. of specimens	Positive specimens	Prevalence
Ewes injected with PGF2α	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* 32.8%, and *Escherichia coli* 22% while *Pseudomonas aeruginosa*, *Streptococcus spp.*, *Klebsilla spp.* and *Proteus vulgaris*, were less frequent isolates (15, 12.4, 12.4, 5.4)% respectively. Table (2) show the bacteria species which isolated from uterus of ewes and the proportion of their presence in all groups of the study, these results did not agree with (9, 10) concluded that *Actinomyces, klebsiella*, *Staphylococcus* were more bacteria presence in sheep (24.1, 19, 13.8)% respectively while its agree with (10) who is reported that the *Staphylococcus aureus* was more bacteria presence in ewes and also agree with (11) which recorded that the *Staphylococcus aureus*, *Escherichia coli* and *Klebsiella spp* were the most common genital bacterial isolates observed in ewes.

Table (2) Bacterial species isolated from ewes uterus swaps and its percentage

Bacteria	PGF2α group		Oxytocin group		Control group		Total	(%)
	No.	(%)	No.	(%)	No.	(%)		
Staphylococcus aureus	6	33.3	10	37.1	8	28.6	24	32.8
Escherichia coli	4	22.3	5	18.5	7	25	16	22
Streptococcus spp.	2	11.1	3	11.1	4	14.3	9	12.4
Klebsilla spp.	3	16.7	3	11.1	3	10.7	9	12.4
Protus vulgaris	0	0	2	7.4	2	7.1	4	5.4
Pseudomonas aeruginosa	3	16.6	4	14.8	4	14.3	11	15
Total	18	100	27	100	28	100	73	100

The study inducted that there were several bacterial types presented in the uterus of ewes which has no effect on the reproductive function. Similar findings were reported in ewes (4), sheep (10,12), Cows (13) and camels (14). However, (15) demonstrated that, gram positive bacteria were the most frequently recovered in infected female genital tract. The differences may be attributed to different case history such as previous parturition or bad hygienic condition (16). In table (2) showed the number of all species of bacteria in PGF2α group less than other groups. The observation that injected ewes with PGF2α caused a significant reduction of bacterial load in uterus is in agreement with finding of (17). In fact, the exogenous PGF2α removes the suppressive effect of progesterone on the genital tract defence mechanism or, alternatively stimulate it through estrogen (18). additionally, PGF2α may have stimulatory effect on phagocytic activity of polymorphonuclear leukocytes (PMN cells) (19). In conclusion, the uterus in postpartum ewes can prevent the development of infections in response to intrauterine inoculation with infectious bacteria, unless it has been exposed to some threshold amount of progesterone from an endogenous or exogenous source (3). This is consistent with another study with postpartum ewes (20) and it seems to indicate that resistance is

the default setting for the postpartum uterus. In the present study, the relationships among PGF2α, susceptibility of ewes to intrauterine inoculation with infectious bacteria seem to indicate that PGF2α may be involved in mediating the immunosuppressive effects of progesterone and its role in transforming the postpartum uterus from resistant to susceptible. Those relationships were clearly delineated in this study, and they are evident, although somewhat less so, in a previous report (20). Based on the relationships among the variables measured in this study, one may Speculate that a method for enhancing uterine PGF2α production may enhance the ability of the uterus to resist infections. In table (2), have been observed that, number of uterus bacteria did not affect with injection of oxytocin; therefore, the oxytocin might have not stimulatory effect on immune system to resistance the bacterial infection.

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