

## Evaluation of the dry-off intramammary antibiotic therapy for controlling of ovine mastitis

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### Abstract

The study was conducted on 100 pregnant Awassi ewes, aged 2-5 years to evaluate a prepartum intramammary treatment for controlling postpartum mastitis. Four hundred mammary secretions or milk samples were collected aseptically (200 samples, 2-3 weeks prepartum and 200 samples, one week postpartum). These were subjected to bacteriological culturing and identification. California mastitis test and white side test were used for diagnosis of subclinical mastitis. Immediately after the prepartum sampling, dry-off intramammary antibiotic was administered to 87 randomly selected udder halves (42/92 infected halves and 45/108 non-infected halves), the remainder 50 and 63 halves were allowed without treatment as infected and non-infected control respectively. Results of culturing revealed 148 and isolates of 20 different bacterial species. Eighty-nine were gram positive and 59 were gram negative. Coagulase negative staphylococci were the most frequent gram positive bacterial isolate 40 (27%), while the predominant gram negative bacteria were Mannheimia haemolytica 16 (10.81%). The Prepartum clinical and subclinical mastitis were 13% and 33% respectively, compared with 5.5% and 22.5% in post partum period. Results of the prepartum treatment of infected udder halves revealed that, out of 42 treated halves, 38 (90.5%) halves were recovered of infection and only 4 treated halves experienced infection during the postpartum period, of these 2 (4.8%) halves retained the same pathogen and the other 2 (4.8%) developed new infection with diverse bacteria. While 42 (93.33%) of 45 prophylactically treated healthy halves were protected from developing postpartum infection and only 3(6.66%) halves contracted infection through the post partum period. In conclusion, Dry-off prophylactic treatment could reduce the incidence of postpartum ovine mastitis. The self-cure phenomenon has an important role in resolving mastitis in ewes.

**Key words:** Mastitis, dry-off therapy, mammary secretion or milk samples

تقييم العلاج بالمضادات الحيوية في الضرع في فترة الجفاف للسيطرة على التهاب الضرع الضائي

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### الخلاصة

أجريت الدراسة على 100 نعجة عواسية حامل، تتراوح أعمارها بين 2 - 5 سنوات، وذلك لتقييم العلاج داخل الثدي المستخدم قبل الولادة للسيطرة على التهاب الضرع بعد الولادة. تم جمع 400 عينة حليب أو إفرازات الضرع (اللبأ) بطريقة معقمة (حيث تم جمع 200 عينة خلال فترة 2 - 3 أسابيع قبل الولادة و 200 عينة أخرى خلال الأسبوع الأول بعد الولادة) وقد خضعت هذه العينات للزرع والتوصيف البكتيري كما اجري اختبار كالفورنيا لالتهاب الضرع California Mastitis Test واختبار الجانب الأبيض White side test لتشخيص التهاب الضرع تحت السريري. بعد جمع العينات ما قبل الولادة تم فوراً حقن المضادات الحيوية الخاصة بفترة الجفاف داخل الضرع في 87 غدة ثديية تم اختيارها عشوائياً (تم اختيار 42 من 92 غدة ثديية مصابة بالتهاب الضرع، في حين تم اختيار 45 من 108 غدة ثديية غير مصابة)، بينما تركت 50 و 63 غدة ثديية من الغدد المصابة والغدد غير المصابة على التوالي، بدون علاج. أظهرت نتائج الزرع البكتيري 148 عزلة من 20 نوع بكتيري مختلف. كانت 89 عزلة

موجبة لصبغة غرام، في حين كانت 59 عزلة سالبة لصبغة غرام، وكانت المكورات العنقودية السالبة لأنزيم التجلط الأكثر انتشارا حيث عزلت من 40 (27%) عينة، بينما كانت بكتريا *Mannheimia haemolytica* الأكثر انتشارا بين البكتريا السالبة لصبغة غرام، حيث عزلت من 16 (10.81%) من العينات. شخّصت حالات التهاب الضرع السريري تحت السريري في فترة ما قبل الولادة في 13 و33% على التوالي، بينما شخّصت في 5.5 و22.5% على التوالي في فترة النفاس. أظهرت نتائج العلاج قبل الولادة للغدد المصابة ان 38 من 42 (90.5%) من الغدد قد شفيت بينما بقيت 4 غدد فقط مصابة بالتهاب الضرع في فترة النفاس، وكانت غدتان منها 4.8% مصابة بنفس نوع البكتريا المعزولة قبل الولادة، بينما أصيبت الاثنتين الباقيتين بنوع آخر من البكتريا. من بين 45 غدة عولجت وقائيا من خلال العلاج في الفترة ما قبل الولادة شفيت 42 (90.5%) غدة من التهاب الضرع بعد الولادة، في حين أظهرت 3 (6.66%) غدد التهاب الضرع في فترة النفاس. نستنتج من الدراسة الحالية ان العلاج الوقائي خلال فترة الجفاف يقلل نسبة حدوث التهاب الضرع بعد الولادة وأن ظاهرة الشفاء الذاتي تلعب دورا مهما في الشفاء من التهاب الضرع في النعاج.

### Introduction

Mastitis is an inflammation of the parenchyma of mammary gland, regardless of the causative agents, but it may also be caused by injury and less commonly allergy and neoplasm (1, 2). Most of the research works conducted to study the impact of mastitis in the livestock have focused on dairy cattle. However, mastitis also exists in the sheep industry and only recently researchers have begun to evaluate the extensiveness and significance of this problem (3). The peripartum increase in the incidence of clinical mastitis observed in dairy cattle is frequent in ewes, a maximal incidence is observed at drying off or at lambing in relation to poor hygiene practices or environmental contamination (4, 5). This prevalence is influenced by several factors including breed, age, number of lactation (parity), general and milking management and environment (5, 6). One-third of the ewes developed clinical mastitis during the first week after lambing, and a second peak, although smaller was observed in the third week postpartum (7). Data on persistence of subclinical mastitis during the dry period are rare. In dairy ewes the mean prevalence of subclinical mastitis may be about 20 – 30 % ranging from 7 to > 60% per lactation(8, 9). Clinical cases around parturition might be newly acquired intramammary infection or aggravations of existing subclinical infections and are mainly of bacterial origin (10,11). In Iraq few studies have been conducted on ovine clinical and subclinical mastitis (12,13,14). The aim of this study is evaluation of dry-off intramammary antibiotic therapy for controlling ovine mastitis.

### Materials and Methods

One hundred pregnant ewes, 2-5 years old of Awassi breed were used to evaluate a dry-off therapy for controlling mastitis during a period from July 2010 to May 2011. Four hundred mammary secretion or milk samples were collected aseptically (200 samples, 2-3 weeks prepartum and 200 samples, one week postpartum) table 1. These were transported to the laboratory on ice and subjected to bacteriological culturing and identification (15, 16). California mastitis test and white side test were used for diagnosis of subclinical mastitis (17). Immediately after the first sampling a dry-off intramammary antibiotic (Siccalactin)\* was administered to randomly selected 42 of 92 infected udder halves and 45 of 108 non-infected halves. The remainder 50 and 63 halves were used as control positive and control negative respectively.

\* Siccalactin: Dry-off intramammary tubes containing long-acting antibiotics, consisting of Benzathin penicillin 200000Iu and Dihydrostreptomycin sulfate 0.4 gm. (Boehringer-Ingelheim Vetmedica As).

## Results and Discussion

Results of culturing revealed 148 isolates of 20 different bacterial species (Table 2). Coagulase negative staphylococci were the most predominant gram positive bacteria accounting for 40 (27.03%) of all isolates, followed by *S. aureus* 10 (6.8%). Similar findings suggested that CNS were the most common pathogen isolated from sheep and goat mastitis (13, 18, 19, 20, 21). Other gram positive isolates were streptococcus dysgalactiae 2(1.35%), *S. uberis* 2(1.35%), *Enterococcus faecalis* 5 (3.38%), *C. renale* 6 (4.05%), *C. pseudotuberculosis* 7 (4.73%), *A. pyogenes* 7 (4.73%), *Micrococcus* Spp. 5 (3.38%) and *Nocardia asteroides* 5 (3.38%). Similar results have been reported by (2, 22, 23). Who demonstrated that most cases of mastitis are caused by gram positive bacteria especially in dry period. Regarding gram negative bacteria, *M. haemolytica* was the more frequent isolate 16(10.81%), followed by *Pseudomonas aeruginosa* 12(8.11%), *Pasteurella multocida* 11(7.43%), *E. coli* 8 (5.40%), *Proteus mirabilis* 5 (3.38%), *Proteus vulgaris* 2(1.35%), *Histophilus ovis* 2(1.35%), *Serratia marcescens* 1(0.67%), *Klebsiella pneumoniae* 1(0.67%) and *Yersinia enterocolytica* 1(0.67%). The relatively high percentage of mastitis due to *M. hemolytica* was reported by other investigators. (22, 24, 25). Who reported that *M. hemolytica* was responsible for 5.3%, 50%, and 21% of ovine mastitis respectively. Enterobacteriaceae isolates in the current study were in agreement with results of (21, 23, 26). *Nocardia asteroides*, *Serratia marcescens* and *klebsiella pneumoniae* were isolated for the first time in Iraq from ovine mastitis. The Results of dry off therapy in treatment of prepartum udder infection revealed 38 (90.5%) out of 42 treated infected halves were recovered of mastitis and only 4 (9.5%) halves experienced postpartum infection, of these, 2 (4.8%) halves retained the same pathogen and the other 2(4.8%) developed new infection with diverse bacteria (Table 3, Fig. 1). Of the fifty non treated control positive halves, 27 (54%) were recovered spontaneously, 11 (22%) halves retained the same pathogens during the postpartum period, and 12 (24%) halves developed infection with other bacteria. It has been mentioned (2) that most dry preparations maintain a dequate minimum concentration in the udder for about 4 weeks, therefore, they protect the mammary gland from susceptible bacteria during weeks after parturition. In this study the low percentage of mastitis in postpartum period, in comparison with prepartum period, may be attributed to dry-off intramammary therapy in last term of pregnancy which may eliminate a large number of mastitis pathogens (27) and to self-cure phenomenon (11, 28). This phenomenon may be ascribed to many of defense mechanisms of the udder, including lysozymes, immunoglobulins, Leukocytes and high concentration of lactoferrin in the dry secretions (2, 29). Results of prophylactic effect of the dry-off therapy revealed that of the forty-five prophylactically treated halves, 42 (93.33%) were protected from contracting postpartum infection and only 3 (6.66%) were infected compared with 25 (39.7%) of 63 non treated halves developed infection during this period. (Table 4, Fig. 2). These results were in agreement with suggestions of many investigators, whose recommended to use prophylactic intramammary treatment during the dry period to protect the mammary glands from new infections, as bacteria become more susceptible to antimicrobial agents during the dry period (27, 30, 31). Relative proportion of mastitis was greatest during the first week post lambing and the new cases of postpartum mastitis may be attributed mainly to environmental infection with several types of bacteria such as *E. Coli*, *P. aeruginosa*, *S.uberis* and *S.dysgalactiae* due to contamination of the udder especially after parturition as well as teat wounds due to vigorous suckling of lambs (2, 32, 33). The results of the study proved that the prophylactic treatment of the udder during the last period of pregnancy is a good execution for controlling mastitis, or at least to decrease the incidence of developing new cases of bacterial mastitis in ewes as the protection rate in treated glands was (93.33%) compared with 60.32% in non treated glands.

**Table (1) Incidence of clinical and subclinical Ovine mastitis**

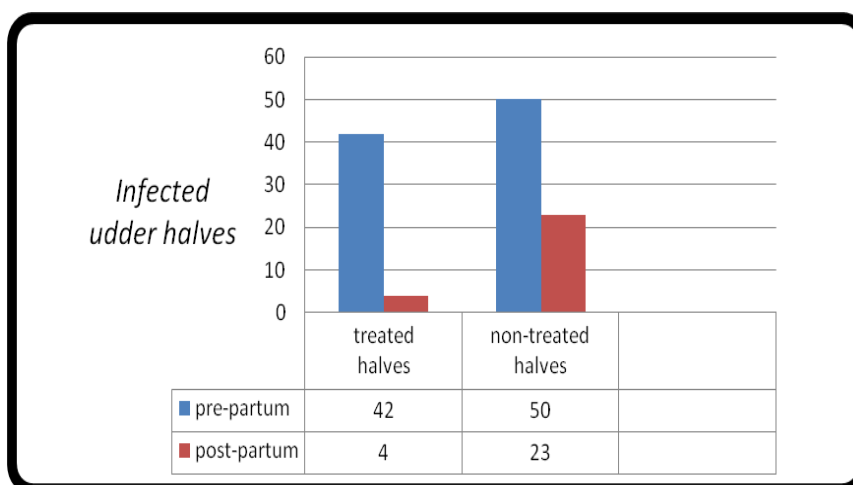
Period	Samples	Infected halves		Total	
		Clinical	Subclinical	Infected	Healthy
Prepartum	200	26 (13%)	66 (33%)	92 (46%)	108(54%)
Postpartum	200	11 (5.5%)	45 (22.5%)	56 (28%)	144(72%)
Total	400	37	111	148	252

**Table (2) Number of isolated bacteria from clinical and subclinical ovine mastitis**

Isolates	Clinical		Subclinical		Pre partum	Post partum	Total (%)
	Pre partum	Post partum	Pre partum	Post partum			
S. aureus	1	2	6	1	7	3	10 (6.8%)
C-NS	4	0	25	11	29	11	40 (27.03%)
S. dysgalactiae	1	0	0	1	1	1	2 (1.35%)
S. uberis	1	0	1	0	2	0	2 (1.35%)
E. faecalis	2	0	1	2	3	2	5 (3.38%)
C. renale	4	0	2	0	6	0	6 (4.05%)
C. pseudotuberculosis	0	0	3	4	3	4	7 (4.73%)
A. Pyogenes	0	0	6	1	6	1	7 (4.73%)
Micrococcus spp.	0	0	4	1	4	1	5 (3.38%)
N. asteroides	0	0	5	0	5	0	5 (3.38%)
M. heamoltyca	3	5	3	5	6	10	16 (10.81%)
P. multocida	2	1	2	6	4	7	11 (7.43%)
P. aeruginosa	4	2	2	4	6	6	12 (8.11%)
E. coli	2	1	1	4	3	5	8 (5.40%)
P. mirabilis	0	0	3	2	3	2	5 (3.38%)
P. vulgaris	1	0	1	0	2	0	2 (1.35%)
H. ovis	0	0	1	1	1	1	2 (1.35%)
S. marcescens	1	0	0	0	1	0	1 (0.67%)
K. pneumonia	0	0	0	1	0	1	1 (0.67%)
Y. enterocolytica	0	0	0	1	0	1	1 (0.67%)
Total	26	11	66	45	92	56	148

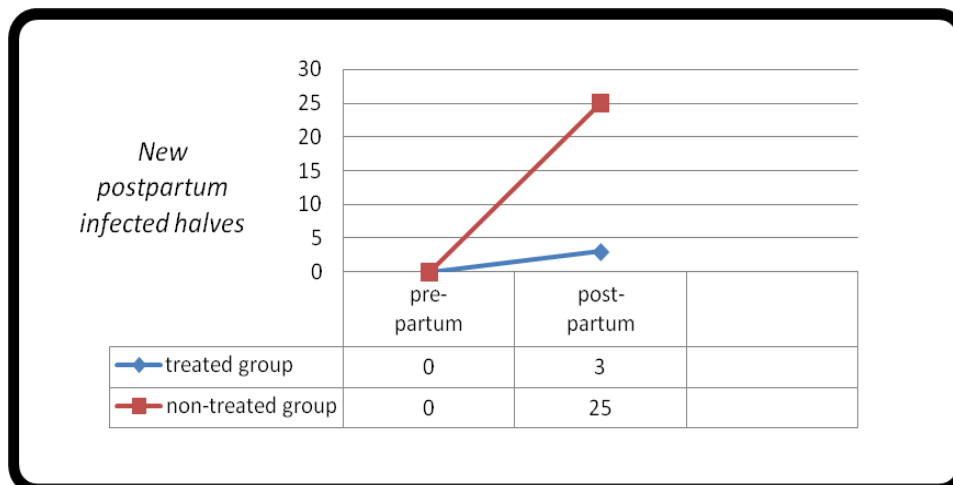
**Table (3) The efficiency of dry-off therapy intreatment of infected glands**

Udder halves	Treated halves			Non treated halves		
	Pre partum	Post partum		Pre partum	Post partum	
		Same bacteria	Other bacteria		Same bacteria	Other bacteria
Infected	42	2 (4.8%)	2 (4.8%)	50	11 (22%)	12 (24%)
Recovered	0	38 (90.5%)		0	27 (54%)	
Total	42	42		50	50	

**Fig. (1) The efficiency of dry-off therapy in cure of infected glands**

**Table (4) The efficiency of dry-off prophylactic therapy in prevention of post partum ovine mastitis**

Udder halves	Treated halves		Non- treated halves	
	Pre partum	Post partum	Pre partum	Post partum
Healthy	45	42 (93.33%)	63	38 (60.32%)
Infected	0	3 (6.66%)	0	25 (39.68%)
Total	45	45	63	63

**Fig. (2) Comparison of the cure rate between prophylactically treated and non-treated halves**

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