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Endemic Contagious Agalactia in Sheep and Goats: Clinical Evaluation, Treatment and Vaccination

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Abstract: In this study, it aimed to evaluate clinical signs of Contagious Agalactia (CA) that had been affecting small ruminants in Kirikkale Province for 1.5 years and to intervene with this endemic CA by administration of enrofloxacin 5 mg kg^{-1} for 5 days and a single dose of meloxicam (0.5 mg kg^{-1}) to acutely infected animals and by vaccination of the flocks. The study was conducted between July 2007 and December 2008 on a total of 745 animals of 5 different flocks with problems of mastitis, keratoconjunctivitis and arthritis. Clinically determined CA was also confirmed by PCR analysis of milk samples. Arthritis was the most frequently observed clinical sign with a rate of 71.84% in diseased animals. Arthritis was observed either as a single problem or concurrently with mastitis or ocular lesions. The co-existence rate of ocular lesions with mastistis was 2.91% while observing all three lesions concomitantly was the least frequent, 2.91%. Ocular lesions alone was not seen in ewes and goats but co-existed with other signs. Ewes and goats exhibited a higher frequency of mastitis alone with a rate of 45 and 44.83%, respectively compared to other two clinical signs. The treatment protocol mentioned above was successful as the general body condition of the clinically diseased animals was improved and subsequently no incidence of death was observed. Animals gradually recovered from mastititis and conjunctivitis and the severity of keratoconjunctivitis and arthritis was decreased. It also concluded that application of a live CA vaccine twice with a 6 month-interval was quite effective as no case of CA was observed in vaccinated flocks during a 1 year follow-up period.

Key words: Contagious agalactia, clinical evaluation, enrofloxacin, meloxicam, treatment, vaccination

INTRODUCTION

Known for almost two centuries, Contagious Agalactia (CA) is a highly infectious disease of dairy goats and ewes. As cited by Madanat *et al.* (2001) the disease was first defined in 1816 and later named as contagious agalactia in 1871. Agalactia is the primary clinical sings in CA affected lactating animals. Polyarthritis, keratoconjuctivitis and respiratory problems are also common (Aiello and Mays, 1998). The disease causes a significant economic loss due to reduction or complete cessation of milk production and abortion as well as due to a high mortality rate in kids and lambs (up to 40-70%) (Madanat *et al*., 2001; Aiello and Mays, 1998; Corrales *et al*., 2004). The disease was previously listed in the Office International de Epizooites (OIE) list B disease group; however, it is now categorized as a notifiable disease (Antunes *et al*., 2008). *Mycoplasma agalactiae* is the classical etiological agent of CA in goats and ewes. However, other *Mycoplasma* species including *M. putrefaciens*, *M. capricolum* subsp. *capricolum* and *M. mycoides* subsp. *capri* may cause similar pathologiacal and clinical signs (Bergonier *et al*., 1997; Aiello and Mays, 1998; Madanat *et al*., 2001; De la Fe *et al*., 2005). Infection begins with intake of contaminated milk, feces, urine, nasal and ocular discharges as well as excretion from effected joints (Madanat *et al*., 2001). Infection can also be induced via galactogenic pathway due to contaminated surroundings or milking equipment (Aiello and Mays, 1998). Some animals may carry the disease without showing any clinical signs while posing a great health risk for the flock. Such animals also reduce the success of treatment as they are erroneously considered healthy based on clinical examination (Aiello and Mays, 1998; Madanat *et al*., 2001). Tetracycline, macrolide, florfenicol, tiamulin and fluoroquinolones are currently used in treatment of CA. However, endemic agalactia can be controlled by means

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of anti-microbial treatment as well as vaccination of the flocks (Bergonier *et al*., 1997; Madanat *et al*., 2001; Assuncao *et al*., 2006). In the study, we aimed to characterize the clinical signs of CA and to control an endemic agalactia in Kirikkale region, applying enrofloxacin and meloxicam to animals with acute clinical signs and through vaccination of all animals in the affected flocks.

MATERIALS AND METHODS

The study was conducted in Kirikkale province between July 2007 and December 2008. The study materials were collected from 5 different flocks with clinical signs of mastitis, keratoconjunctivitis and arthritis. After receiving the history of the flocks, all animals belonging to 5 different flocks were clinically examined for the presence of swollen and sensitive udder and enlarged mammary lymph nodes, swollen testis, arthritis and ocular lesions. Out of 745 animals, 103 animals had one of the above findings. Out of 103 animals, 41 animals were randomly selected for clinical evaluation. The body temperature, heart rate and respiration rate were measured prior to and post-treatment in these animals.

Milk samples from the diseased ewes (n = 18), goats (n = 36) and yearling female lambs (n = 4) were collected. Milk samples centrifuged at 12000 g for 20 min and the pellet was resuspended in 300 μL of water. The bacterial suspension was then treated with 300 μL TNES buffer (20 mM Tris pH 8.0, 150 mM NaCl, 10 mM EDTA, 0.2% SDS) and proteinase K (200 μg mL^{-1}) and was kept at 56°C for 1 h.

The suspension was heated at 95°C for 10 min to inactivate proteinase K. DNA samples were tested with species specific PCR for *M. agalactiae*. The PCR was performed in a TC 512 Temperature Cycling System (Techne, Staffordshire, UK) in a reaction volume of 50 μL containing 5 μL 10×PCR buffer (750 mM Tris Hcl, pH 8.8, 200 mM $(NH4)_2SO4$, 0.1% Tween 20), 5 μL 25 mM $MgCl_2$, 250 μM of each deoxynucleotide triphosphate, 1.25 U Taq DNA Polymerase (MBI, Fermentas), 20 pmol of each primer (IDT) and 25 ng of template DNA. A primer pair specific to *ma-mp 81* gene of *M. agalactiae* (*Ma-mp 81* F 5′-AGCAGCACAAAACTCGAGA-3′; *Ma-mp81* R 5′-AACACCTGGATTGTTTGAGT-3′) was used in the PCR (Foddai *et al*., 2005).

Blood samples were collected into anti-coagulated tubes via venipuncture of vena jugularis of the diseased ewes (n = 27), goats (n = 19) and lambs (n = 23). These samples were analyzed for total blood counts using a MS9 cell counter (MS9-3-Melet Schloesing Laboratories, France). Animals with acute clinical signs were given enrofloxacin (Vilfloks, Vilsan, Turkey) subcutaneously at a dose of 5 mg kg^{-1}/5 days and a single dose of 0.5 mg kg^{-1} subcutaneous meloxicam (Maxicam, Sanovel, Turkey). The diseased animals with prominent arthritis were also given a single intra-articular injection of 1 mL of a combination of meloxicam (2.5 mg) + enrofloxacin (50 mg). Data collected prior to and post-treatment were analyzed with student t-test. Data expressed as mean±SD are shown in Table 1. All animals of 5 different flocks (255 ewes, 199 goats, 8 yearling female lambs, 163 lambs, 79 kids, 6 rams and 35 bucks) were vaccinated twice with a 6-months interval.

RESULTS

General findings: Excluding the 12 kids died of septicemia prior to beginning of the treatment protocol, the flock size varied from 28-317. The species distributions of the flocks are shown in Table 2.

Clinical findings and diagnosis: Mastitis, arthritis and ocular problems with various degrees of severity were observed in animals of each flock. As an indication of an ongoing infection, animals with acute clinical signs had increased body temperature, heart rate, respiration rate and leukocyte counts. The clinically diagnosed CA cases were further confirmed by PCR analyses for *Mycoplasma agalactiae*.

Table 1: Some clinical parameters and white blood cell profile at before and after treatment

	Ewe (X±SD)		Goat (X±SD)		Lamb (X±SD)	
Parameters	B	A	B	A	B	A
T (°C)	39.87±0.520^{a}	38.93±0.470^{b*}	40.02±0.460^{a}	39.25±0.62^{b**}	40.29±0.490^{a}	39.00±0.72^{b**}
P (min^{-1})	93.14±13.00^{a}	78.24±10.27^{b*}	88.50±10.87	79.50±9.83	99.88±8.620^{a}	80.40±3.62^{b***}
R (min^{-1})	52.00±13.00^{a}	28.24±7.180^{b***}	40.33±7.060^{a}	30.05±2.83^{b**}	61.52±16.19^{a}	40.40±3.67^{b***}
WBC ($x10^{9}$ L^{-1})	20.10±2.400^{a}	13.24±2.180^{b***}	15.52±3.060^{a}	11.05±2.13^{b**}	18.75±2.860^{a}	14.10±1.07^{b**}
LYM (%)	79.71±8.670^{a}	64.90±5.080^{b**}	19.57±9.700^{a}	40.12±7.37^{b***}	78.90±5.230^{a}	62.70±4.33^{b**}
MONO (%)	6.28±2.180^{a}	9.17±5.780^{b*}	7.42±1.840	7.60±2.18	4.38±1.750^{a}	7.08±2.39^{b**}
NEU (%)	13.75±4.430^{a}	25.77±4.210^{b**}	70.92±12.74^{a}	52.27±8.44^{b*}	18.16±4.430^{a}	29.54±3.20

A: After treatment, B: Before treatment, *p<0.05, **p<0.01, ***p<0.001

Out of the 745 animals included in this study, 103 (13.83%) animals showed at least one of the followings: ocular lesions, arthritis, or mastitis. None of the male animals showed testicular lesions such as redness, swelling, soreness or atrophy. Table 3 shows the distribution of clinical findings.

Either alone or in association with mastitis and ocular problems, arthritis was the most frequently observed clinical sign (71.84%) (74/103). The rate of co-existence of ocular lesions with mastitis was 2.91% (3/103) and the rate of co-existence of all three lesions was 2.91% (3/103). Arthritis was the only clinical finding in diseased rams and bucks.

Mastitis alone was the most frequently observed clinical sign in ewes and goats at a rate of 45% (9/20) and 44.83% (13/29), respectively. Ocular lesions in ewes and goats were always associated with mastitis or arthritis or both.

Bilateral mastitis was observed in 88.24% (15 out of 17 cases) of the diseased ewes and 72% (18 out of 25 cases) of goats (Table 4). The affected mammary lobes were sored, swollen and stiff and some of the affected mammary lobes had complete loss of milk production. Milk was greenish in color and milk clots were often present. The mammary lymph nodes were easily palpated and some of the lymph nodes were enlarged up to 3 cm in diameter.

Table 2: The species distributions in the flocks

	Flock					
Species	1	2	3	4	5	Total
Ewe	17	15	150	21	52	255
Goat	18	5	7	19	150	199
Yearling female lamb	8	0	0	0	0	8
Lamb						
Male	2	1	0	74	2	79
Female	4	3	70	1	6	84
Kid						
Male	0	1	0	1	35	37
Female	0	1	0	1	40	42
Ram	1	1	3	1	0	6
Buck	1	1	0	1	32	35
Total	51	28	230	119	317	745

A 5 months old lamb and two yearling female lambs which never became pregnant had bilateral mastitis with swollen mammary lymph nodes. In these yearling female lambs, milk secretion was observed and the milk contained greenish pus and milk clots. The affected ewes and goats of non-lactating period restarted lactating with an abnormal appearance of milk.

One lobe of the udder was missing *in toto* in two ewes (Fig. 1). The remaining lobe was sored. The intervention protocol alleviated the soreness. Arthritis was another common finding affecting either one joint as in 53 cases (71.62%) or multiple joints as in 21 cases (28.38%). While ewes, goats, yearling female lambs, male lambs, male kids and rams usually had mononarthritis, female lambs and bucks had polyarthritis (Table 4). The arthritic joints were hot, painful and swollen. Ankylosis was detected in the tarsal joint of an ewe (Fig. 1). In cases associated with arthritis, carpal (54.90%) and tarsal joints (31.37%) were most frequently affected while the stifle joint (0.98%) had the least frequency of arthritis. While the carpal joint was most frequently affected in ewes, yearling female lambs and male lambs, the tarsal joint was most frequently affected in goats and female lambs (Table 5). As far as ocular

Table 3: The distribution of clinical findings

Species	O	A	M	O+A	O+M	M+A	O+ M+A	Total(n)
Ewe	0	2	9	1	1	7	0	20
Goat	0	3	13	1	2	7	3	29
Yearling female lamb	0	2	0	1	0	2	0	5
Lamb								
Male	2	24	-	4	-	-	-	30
Female	1	2	0	5	0	1	0	9
Kid								
Male	0	2	-	0	-	-	-	2
Female	1	0	0	0	0	0	0	1
Ram	0	2	-	0	-	-	-	2
Buck	0	5	-	0	-	-	-	5
Total (%)	4 (3.88)	42 (40.78)	22 (21.36)	12 (11.65)	3 (2.91)	17 (16.50)	3 (2.91)	103 (100)

O: Ocular lesions, A: Arthritis, M: Mastitis

Table 4: The severity of clinically findings

	Ocular lesion			Arthritis			Mastitis		
Species	n	Unilateral	Bilateral	n	Monoarthritis	Poliarthritis	n	Unilateral	Bilateral
Ewe	2	0	2	10	8	2	17	2	15
Goat	6	3	3	14	12	2	25	7	18
Yearling female lamb	1	1	0	5	3	2	2	0	2
Lamb									
Male	6	6	0	28	22	6	-	-	-
Female	6	4	2	8	2	6	1	0	1
Kid									
Male	0	0	0	2	2	0	-	-	-
Female	1	1	0	0	0	0	0	0	0
Ram	0	0	0	2	2	0	-	-	-
Buck	0	0	0	5	2	3	-	-	-
Total	22	15	7	74	53	21	45	9	36

Table 5: The species distribution of arthritis with respect to affected joints

Species	Carpal joint	Tarsal joint	Metacarpophlangeal joint	Metatarsophlangeal joint	Stifle Joint	Total
Ewe	7.00	5.00	0.00	0.00	0.00	12
Goat	4.00	6.00	2.00	3.00	1.00	16
Yearling female lamb	6.00	0.00	0.00	1.00	0.00	7
Lamb						
Male	25.00	9.00	0.00	0.00	0.00	34
Female	10.00	11.00	0.00	0.00	0.00	21
Kid						
Male	0.00	0.00	0.00	2.00	0.00	2
Female	0.00	0.00	0.00	0.00	0.00	0
Ram	2.00	0.00	0.00	0.00	0.00	2
Buck	2.00	1.00	4.00	1.00	0.00	8
Total (%)	56 (54.90)	32 (31.37)	6 (5.88)	7 (6.82)	1(0.98)	102 (100)

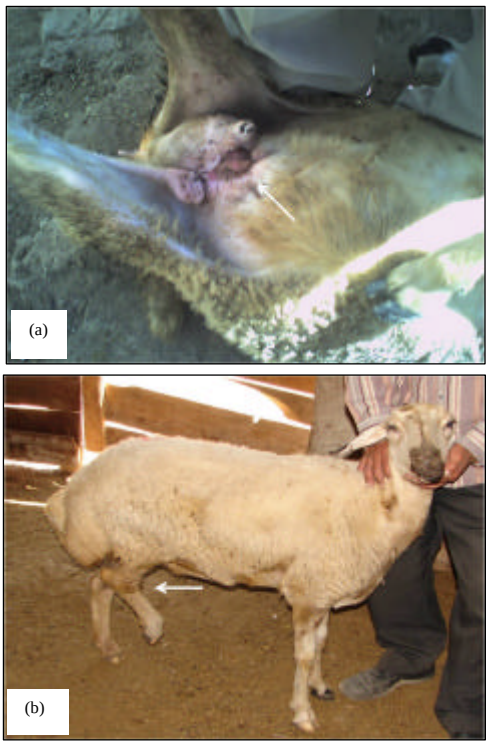

Fig. 1: (a) An ewe with a missing lobe of the udder and (b) An ewe with ankylosis in the tarsal joint

lesions, 15 cases (68.18%) had unilateral ocular lesions while 7 cases (31.82%) had bilateral ocular lesions. Two ewes had bilateral ocular problems while goats had either bilateral or unilateral ocular problem with an equal frequency.

Ocular lesions are unilateral in all male lambs with ocular problems and 4 out of 6 (66.67%) of female lambs with ocular problems (Table 4). The severity of ocular lesions varied from mild conjunctivitis to keratoconjunctivitis. female lambs, the udder lesions were regressed quickly however, it took longer time for the swollen lymph nodes However, corneal ulceration was also detected in 5 out of 22 cases of ocular lesions (22.73%).

Post-treatment observation: Following antibiotic and NSAID treatment, the general health condition of the diseased animals improved to great degree (Table 1) and recovered from the disease within 3 weeks of the first vaccination and no new cases of CA were detected.

The higher body temperature ($p<0.05$ in ewes; $p<0.01$ in goats; $p<0.01$ in lambs), heart rate ($p<0.05$ in ewes; $p<0.001$ in lambs), respiratory rates ($p<0.001$ in ewes; $p<0.01$ in goats; $p<0.001$ in lambs) and leukocyte counts ($p<0.001$ in ewes; $p<0.01$ in goats; $p<0.01$ in lambs) in diseased animals significantly dropped compared to values obtained prior to treatment (Table 1). In animals with mastitis including lamb and yearling to return their normal size. The swollen udder and lymph nodes of the affected animals returned to normal sizes within a month and animal regained to regular lactation.

In all animals received an intra-articular injection (1 ewe, 9 goats, 2 yearling female lambs, 4 lambs and 3 kids), the severity of limping was decreased within 24 h after treatment. Although, the severity of joint swelling decreased in a short period of time, it took 3-4 weeks for the arthritic joints to return normal condition. In animals with ocular lesions, conjunctivitis disappeared within a week while keratoconjunctivitis was needed at least 3-4 weeks following antibotic administration. Likewise, the severity of the corneal ulcerations was decreased but the complete healing took longer time. During a 1 year follow-up period beginning from the second vaccination given 6 months after the first one, no clinic CA cases were detected in vaccinated flocks.

DISCUSSION

Clinical evaluation: CA is a serious disease of sheep and goats that is characterised by mastitis, arthritis, keratoconjunctivitis and occasionally, abortion and respiratory disease. *Mycoplasma agalactiae* is the main cause of the diseases (Nicholas, 1995; Bergonier *et al*., 1997; Aiello and Mays, 1998; Madanat *et al*., 2001; De Azevedo *et al*., 2006, Kizil and Ozdemir, 2006; OIE, 2008). The diagnosis of CA is established on the basis of

epidemiological finding and the presence of clinical signs (Madanat *et al*., 2001; Aiello and Mays, 1998). In the study, mastitis, arthritis and ocular lesions with variable degrees of severity were detected in animals with CA. The clinical diagnosis was confirmed with determination of *Mycoplasma agalactiae* in milk samples by PCR analysis, as well.

Madanat *et al.* (2001) reported that the morbidity in CA can reach up to 30-60% and the mortality can increase up 40-70% in lambs and kids. In the study, the general morbidity was 13.83% (103/745) however, it was 7.84% (20/255)-14.57% (29/199) in ewes and goats, respectively while it was 23.93% (39/163)-3.79% (3/79) in lambs and kids. Gil *et al.* (2003) reported that the mortality rate in kids was 82%. Taking into consideration the time period past from the first observance of CA to the beginning of the study, the mortality rate in kids in the study was 80% (12/15) that supported the findings by Gil *et al*. (2003). As also mentioned by Bergonier *et al.* (1997) prominent septicemia and pneumonia were common to kids died of the disease in the study however, respiratory problems were not observed in adult diseased animals. Such an observation supported the claim by Aiello and Mays (1998) that CA causes pneumonia during early ages. The absence of dead incidence post-treatment in the study is most likely related to success of treatment protocol and refinement of animal care and feeding conditions as also previously implicated by Corrales *et al*. (2004).

Arthritis is one of the characteristics clinical findings in CA however, its incidence rate varied in different reported studies. Gil *et al.* (2003) reported the arthritis incidence rate as 30% while De Azevedo *et al.* (2006) reported 48.30% in goats. In the study, the arthritis rate among the diseased ewes (50%) (10/20) and goats (48.28%) (14/29) was similar to that of the De Azevedo *et al.* (2006) study. However, arthritis is quite frequent among the diseased male lambs, 93.33% (28/30) (Table 3). Importantly, arthritis was the only clinical sign in diseased rams and bucks. As also outlined by the present study, the carpal and tarsal joints are the most requently effected joints in CA (Madanat *et al*., 2001). There is no difference between the carpal and tarsal joints by means of arthritis incidence in ewes and goats. However, the incidence of carpal joint arthritis was the most frequent in yearling female lambs (85.71%) (6/7) and male lambs (73.53%) (25/34). Arthritis in the stifle joint was only observed in goats with an incidence rate of 0.98% (1/102) (Table 5). While a single joint was affected in 71.62% (53/74) of arthritis cases, the remaining cases were observed in more than one joint. As we also observed in the cases, the arthritic joints resulting from CA are often swollen and painful and ankylosis may occur as the case becomes chronic (Aiello and Mays, 1998; Madanat *et al*., 2001; De Azevedo *et al*., 2006). Following the treatment of enrofloxacin and meloxicam, joint swelling and pain disappeared and a periarticular thick tissue generated gradually. Ankylosis was observed only in one tarsal joint of an ewe (Fig. 1).

Lactating mammary glands are quite susceptible to ascending infection including CA such that mastitis is another commonly seen problem especially in lactating animals affected by CA (Madanat *et al*., 2001). It observed that mastitis is the most frequent clinical finding in CA affected ewes and goats such that approximately 45% of CA affected ewes (9/20) and goats (13/29) had mastitis. This rate may increase up 75% in animals with a higher milk yield such as Saanen goats (Gil *et al*., 2003; Verbisck-Bucker *et al*., 2008). As Madanat *et al.* (2001) and Aiello and Mays (1998) previously described, it found that the udder at the beginning of mastitis was hot, swollen and sored; it gradually became atrophied due to extensive fibrosis of the secretory tissues. The clinical diagnosis of CA affected lactating ewes and goats was not difficult as the mammary lymph nodes were enlarged to a size up 3 cm in diameter and the milk samples were watery and greenish in color and had clots, characteristic to CA (Aiello and Mays, 1998; Madanat *et al*., 2001).

Mastitis was more severe in two ewes by the time of the first examination such that one lobe of the mammary gland was necrotic and totally missing. As the other lobe which also has mastitis was not missing and responded to treatment protocol positively (Fig. 1). Consequently, it raised a question was raised if other concomitant ascending infections secondary to *Mycoplasma agalactiae* might have increased the severity of mastitis in the necrotic lobe. Importantly, a 5 months old lamb and 2 yearling female lambs had bilateral mastitis and enlarged mammary lymph nodes. Milk secretion with pus and clots from these young animals may show that the mammary gland is a target tissue in CA even in the absence of galactogenic pathway.

Ocular lesions are also common to CA affected animals. As also found in the present study, the ocular lesions initiating with conjunctivitis, lacrimation and photophobia may progress to corneal vascularization and later to keratoconjunctivitis (Aiello and Mays, 1998). The OIE (2008) stated that ocular lesion was seen in 5-10% of CA affected animals while Gil *et al.* (2003) reported a much higher rate, 25%. In the study, we observed ocular lesions approximately in 21% (22/103) of the diseased animals. Out of 22 cases of CA cases associated with ocular lesions, 15 (68.18%) had unilateral ocular lesions while 7 (31.82%) had bilateral ocular lesions. In addition, the ocular lesion was characterized by corneal ulceration

in 5 animals. The ocular lesions in ewes and goats were always associated with CA lesions, arthritis and mastitis (Table 3). The ocular lesions were more severe in lambs such that they often had unilateral keratoconjunctivitis (Table 4). This situation is most likely related to the fact that lambs are more susceptible to keratoconjunctivitis compared to adults.

Genital lesions may ocur especially in *M. putrefaciens* induced CA cases (Gil *et al*., 2003). However, it did not encounter genital lesions in the cases. This is evidently related to the fact that *M. agalactiae* is the etiological agent in the cases as we confirmed through PCR analyses. In addition, the mastitis and arthritis co-existence is quite higher in *M. agalactiae* induced CA cases (De Azevedo *et al*., 2006). In the study, the rate of mastitis and arthritis co-existence was 35% (7/20) in ewes and 34.48% (10/29) in goats.

Evaluation of treatment: In treatment of CA in sheep and goats, antibiotics including tetracycline, macrolide, florfenicol, tiamulin and fluoroquinolones can be used (Bergonier *et al*., 1997; Aiello and Mays, 1998; Madanat *et al*., 2001). Based on the minimum inhibitory concentration $(MIC)_{90}$ determined through an *in vitro* sensitivity assay on field isolates of *Mycoplasma agalactiae* by Antunes *et al*. (2008) and according to flow cytometric determination by Assuncao *et al*. (2006), the most effective antibiotic derivatives are fluoroquinolons. Consequently, enrofloxacin was used a fluoroquinolon derivative, at a dose of 5 mg/kg/day for 5 days. In addition to the choice of antibiotic, we administered subcutaneously a single dose of meloxicam (0.5 mg kg^{-1}), an oxicam group NSAID with anti-inflammatory, antipyretic, anti-exudative and analgesic effects (Van Bree *et al*., 1994; Rainsford *et al*., 1999). As it observed no incidence of death post-treatment, improvement in general body condition as well as return of leukocyte counts, heart rate, respiratory rate and body temperature to normal ranges (Table 1), it thought the intervention protocol was successful. Likewise, the severity of mastitis, conjunctivitis, keratoconjunctivitis and arthritis was significantly decreased. The limping in arthritic cases was improved within 24 h following intra-articular injections. Evidently, this situation indicates a strong antimicrobial activity as well as anti-inflammatory, analgesic and antiexudative effects reducing the joint effusion. In addition, meloxicam has no chondrotoxic effect (Rainsford *et al*., 1999). The treatment outcome in this study was quite comparable to those of Van Bree *et al*. (1994), Rainsford *et al*. (1999) and Peterson and Keefe (2004).

Evaluation of vaccination: Attenuated live or inactive vaccines have been used in prophylaxis of CA in sheep and goats (Leon-Vizcaino *et al*., 1995; Aiello and Mays, 1998; Madanat *et al*., 2001; Greco *et al*., 2002; Nicholas, 2002; Gil *et al*., 2003). In the study, it administered live vaccine twice with a 6 month interval and subsequently no cases of CA was observed during 1 year follow-up period.

However, live vaccines are not allowed in European countries (Chessa *et al*., 2009). Although, it has some disadvantages, live CA vaccine provide an excellent immunity with a longer period of protection (Nicholas, 2002; Madanat *et al*., 2001) and can be used in infected animals.

When applied to infected animals, it helps re-establish lactation and alleviate the severity of arthritis. These are the facts behind our choice of vaccine. Inactive vaccines have no common use since they provide a lower level of immunity so that its re-administrations are needed in short periods (Leon-Vizcaino *et al*., 1995; Madanat *et al*., 2001; Chessa *et al*., 2009). Thus, there has been an undergoing effort to develop a safer vaccine with a longer period of protection (Santona *et al*., 2002; De la Fe *et al*., 2005, 2006; Chessa *et al*., 2009; Buonavoglia *et al*., 2010).

CONCLUSION

The study concluded that arthritis either alone or in association with ocular lesions or mastitis is the most frequently observed clinical sign in small ruminant CA. Moreover, arthritis is the only clinical sign in CA affected rams and bucks as well as in 80% of male lambs. Mastitis alone is the most frequently observed clinical sign in ewes and goats, characteristic to almost half of the CA affected ewes and goats.

The treatment protocol composed of enrofloxacin 5 mg kg^{-1} for 5 days and a single dose of meloxicam (0.5 mg kg^{-1}) is effective as it alleviates the clinical signs in a short period of time. The administration of a live vaccine twice with a 6 month interval is successful in CA prophylaxis as no CA case was observed during a 1 year follow-up period.

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