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RESEARCH ARTICLE

Pathogenesis and Immunohistochemical Studies of Caprine Pleuropneumonia in Experimentally Infected Goats

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ABSTRACT

This study was designed to evaluate the pathogenesis of caprine pleuropneumonia (CPP) in the experimentally inoculated goats with *Mycoplasma mycoides* subspecies *Capri* (*Mmc*). For this purpose, 12 goats (Group B) were inoculated with bacterial isolates of *Mmc* while four goats were kept as untreated control (Group A). Clinical signs of the disease were recorded twice daily. Two goats from group B were sacrificed on weekly basis to demonstrate gross pathological lesions in different organs. Tissue samples from lungs, trachea, liver, heart, kidney, spleen, and small intestines were preserved for histopathological studies. The lungs and lymph nodes were preserved to demonstrate the antigen in tissue by using immuno-histochemical technique. The disease was successfully reproduced in all infected goats with severe manifestation. The clinical signs and gross lesions of the disease were mild at the beginning and became severe at the third and fourth weeks and then progressed to moderate and chronic forms. The histopathological lesions characteristic of CPP were found in all the organs. Antigen of *Mmc* was detected in tissue sections of lungs and lymph nodes. In conclusion, the disease was efficiently reproduced in experimental animals that showed acute septicemic form with lethal outcome.

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INTRODUCTION

Among the various diseases of caprine, caprine pleuropneumonia (CPP) is a major threat to the goat population with a widespread distribution in Pakistan and results in heavy economic losses to the goat farmers every year (Rahman *et al.*, 2003; Awan *et al.*, 2009). This disease is common in Africa (Regassa *et al*., 2010), Middle East (Arif *et al*., 2007), Europe (Pettersson *et al.*, 1996) and subcontinent (Mondal *et al.*, 2004). It is enlisted by the Office of International Epizootic as a list B disease (OIE, 2008).

The causative agents of CPP in goats are bacteria of the genus *Mycoplasma*, which consists of a group of species and subspecies called the *M. mycoides* clusters that includes *M. Capricolum* subspecies *Capricolum, M. capricolum* subspecies *Capripneumoniae, M. mycoides subspecies capri, M. mycoides* subspecies *mycoides* large colony, *M. mycoides* subspecies *mycoides* small colony and *Mycoplasma bovine* group 7. The acute septicemic disease is caused by *Mycoplasma mycoides* subspecies *capri* (*Mmc*) (Laura *et al*., 2006). In the "subcontinent" the disease is caused by *Mycoplasma mycoides* subspecies *capri*, which is one of the major causes of contagious caprine pleuropneumonia (Thiaucourt and Bölske, 1996; Srivastava *et al*., 2010).

Under experimental infection the morbidity and mortality is 100% (Hasso *et al*, 1993). Typical cases of CPP are characterized by pyrexia (41-43°C) with respiratory symptoms. Respiration is painful, because of violent and frequent coughing. Other clinical symptoms may include lameness, diarrhea, inability to move, stand with abducted front legs, stiff neck and prostration on ground with lateral recumbancy (OIE, 2008).

Gross lesions observed are inflamed lungs with a marble appearance, fibrinous pleuropneumonia, lungs

hepatization and accumulation of a straw-colored fluid in the pleural cavity. Unilateral or bilateral pneumonia are commonly present (Ettorre *et al*., 2007; Goncalves *et al*., 2010). Bronchial and mediastinal lymph nodes are swollen and edematous. In some cases pericardial sacs are filled with a serosenguinous fluid. The liver and kidneys are enlarged with hemorrhages and diffused necrotic foci (Gelagay *et al.*, 2007). Histopathologically, the disease is characterized by fibrinopurulent pleuropneumonia and catarrhal bronchopneumonia with thickening of the interlobular septa (Laura *et al.*, 2006).

In most of the previous studies focus has been made on isolation and identification of causative agent of CPP. Here is an obvious need of further studies to monitor features of the course of infection, to provide adequate information about the pathogenesis of disease in experimental infected animals. The present study was planned to give insight into accurate and timely identification of CPP in experimentally infected goats on the basis of histopathological changes and antigen detection in infected tissues through immunohisto-chemical technique.

MATERIALS AND METHODS

Pathogenesis of CPP under experimental conditions

Sixteen goats, approximately 2-3 months of age, apparently free from any infection and non vaccinated against CPP were procured from a local market. The animals were kept at Animal House of University of Veterinary and Animal Sciences, Lahore, Pakistan under standard management conditions. They were reared on hay, green fodder and concentrate and offered clean drinking water *ad libitum*. Prior to the start of the experiment, animals were dewormed with Nilzan Plus (ICI, Karachi, Pakistan).

Experimental design

These animals were divided into two groups, group A which served as untreated control, having 4 animals and group B having 12 animals, which were inoculated with *Mmc* at dose rate of 1×10^7 CFU dissolved in 3 ml distilled water via intratracheal route by the method described by Wesonga *et al*. (2004). The course of the disease was monitored by clinical examination of infected goats according to the standard clinical protocol. Clinical examination was done at 12 hours intervals from the day of inoculation until death. The data regarding clinical parameters, like rectal temperature, cough and respiration rate were documented daily throughout the length of experiment.

Necropsy: Complete necropsies were performed on all goats of group A and B. Two goats from group B were slaughtered at weekly intervals after appearance of clinical sign of disease. Gross lesions in organs like lungs, pleura, liver, heart, kidney, spleen, trachea and small intestine were recorded.

Histopathology: Tissue samples collected were fixed in 10% buffered formalin and stored for histopathological examination. The preserved samples of lungs, trachea, liver, heart, kidney, spleen and small intestine were processed for tissue sectioning and detailed histopathological examination according to the standard procedure (Ahmad *et al*., 2011). Histological lesions in each organ were carefully observed and recorded.

Immunohistochemistry: To demonstrate *Mycoplasma* antigens in tissues, a labeled streptavidin biotin (LSAB) method was used as described by Rodriguez *et al*. (1996). Hyperimmune sera were raised in rabbits against *Mmc* by using the standard procedure of OIE (2008). The lungs and lymph nodes were processed by routine paraffin embedding technique and sections of 5 µm thickness were used for immunohistochemical processing. For recording detailed immunohistochemical reactions, the prepared slides were examined under microscope at 10X, 40X and 100X. Fisher Exact test was applied by using SPSS for analysis of data.

RESULTS

Clinical signs: All animals in group B that were infected via the intratracheal route with *Mmc* antigen exhibited elevation in their body temperatures until twelve hours post-inoculation. Temperatures subsided on day 3rd post-inoculation. During the initial pyrexic episode, animals were depressed and slightly anorexic. Body temperatures were raised again on day 6th post-inoculation with lacremation and serous nasal discharge in four goats. On day 9th post-inoculation, almost all the animals exhibited signs of high fever ranging from 41.1-41.6^{0}C (Table 1). On day 11th post-inoculation, the nasal discharges became thick and purulent; signs of pneumonia were more pronounced accompanied by rapid and abdominal breathing. On day 15th, classical signs of CPP were observed in three goats in the form of abduction of the forelimbs. The animals were reluctant to move and were slightly anorexic. The neck was extended with moderate pyrexia and excessive mucopurulent nasal discharge. There was excessive lacremation with pus drooling out from the eyes with conjunctivitis. Marked signs of dyspnea with diarrhea in two animals were observed. Two animals were found lying on the ground showing nervous signs of abnormal movements with bellowing. Goats were anorexic and unable to stand. On day 16th postinoculation, two animals were found dead. There were marked signs of pleuropneumonia with extended stiff neck and mild nervous symptoms.

Gross lesions: The goat slaughtered at the end of Week 1 showed consolidations of lungs, at the apical and intermediate lobes (Table 2). Bronchial and medistinal lymph nodes were slightly enlarged. On incising lungs, frothy and fibrinous exudates oozed out. Trachea of both goats was slightly congested with thin fibrinous mucous plug in lumens. Pleural surfaces were slightly congested with fibrinous depositions. Livers were pale in color with streaks of hemorrhages. Mild congestions of intestinal mucosa with enlarged mesenteric lymph nodes were found.

At the end of 2nd Week, the thoracic cavities of both slaughtered animals were filled with straw-colored fluid. The lungs were highly congested and consolidated. The mediastinal and bronchial lymph nodes were also enlarged. Fibrinous coats were present on the surface of

Table 1: Clinical signs and symptom of CPP in experimentally inoculated kids with *Mycoplasma mycoides capri*

Goat No	Nasal discharge	Anorexia	Lacremation	Cough	Pneumonia		pyrexia	Diarrhea	Nervous signs	Abduction of legs	Arthritis
					Bilateral	Unilateral					
1	+	+	+	+	-	L +	+	+	+	+	-
2	+	+	+	+	-	L +	+	+	+	-	-
3	+	-	+	+	-	L +	+	-	-	-	-
4	+	-	+	+	-	R +	+	+	-	+	-
5	+	+	+	+	-	L +	+	-	-	-	-
6	+	+	+	+	B +	-	+	+	+	-	-
7	+	+	+	+	-	R +	+	+	-	+	-
8	+	+	+	+	-	L +	+	-	-	-	-
9	+	+	+	+	B +	-	+	-	-	-	-
10	+	+	+	+	-	R +	+	+	-	-	-
11	+	+	+	+	B +	+	+	+	-	-	-
12	+	+	+	+	-	L +	+	+	+	+	-

= Absent, + = Present, B = Bilateral, L = Left, R = Right

Table 2: Characterization of gross lesions in goat kids inoculated with *Mycoplasma mycoides sub species capri*

Goat No	Lung involved	Consolidation	Pleural fibrin deposit	Pleural adhesions	Distribution of gross changes	Congestion in trachea	Enlarged lymph nodes	Hydro pericardium	Pus in pelvis of kidney	Intes.hea-morrhages
1	L	+	+	+	Ext	+	+	+	+	+
2	L	+	+	-	Ext	+	+	+	+	+
3	L	+	-	-	-	+	+	+	+	-
4	R	+	+	+	MF	+	+	+	+	+
5	L	+	-	-	-	+	+	-	-	-
6	RL	+	+	+	MF	+	+	+	+	-
7	R	+	+	+	Ext	+	+	+	+	+
8	L	+	-	-	-	+	+	-	-	-
9	RL	+	+	+	MF	+	+	+	+	+
10	R	+	+	+	MF	+	+	+	+	+
11	RL	+	+	+	MF	+	+	+	+	+
12	L	+	+	+	Ext	+	+	+	+	+

L = Left, R = Right, RL = Right and Left, + = Presence of lesions, - = Absence of lesions, EXT = Extensive involvement, MF = Multi Focal,

the lungs. Pale-colored pericardial fluid was accumulated in the pericardial sac of both goats. Kidneys were slightly congested and few necrotic foci were present on its surface. On incision, pus was present in the pelvis of the kidneys in all the animals. Livers were slightly pale in color with hemorrhages and multiple necrotic foci. Mesenteric lymph nodes were enlarged and the mucosa of intestine showed mild hemorrhages.

At the end of 3rd Week, postmortem of two slaughtered goats showed extensive involvement of the lungs, with consolidation and focal abscesses (Fig. 1). A thick fibrinous coat covered the surface of the lungs. A slight pleural adhesion was also present. Viscous straw-colored fluid was found in pleural cavities. Pericardial fluid was also accumulated in the pericardial sac. Kidneys were enlarged and congested and necrotic foci were present on the surfaces. The liver was pale in color having multifocal necrotic foci. The intestines were hemorrhagic with enlarged mesenteric lymph nodes.

The animals killed at the end of weeks 4th and 5th exhibited similar lesions as noted at the end of 3rd week but with more severity.

Histopathology: All the animals of group B showed significant histological changes in different organs including trachea, lungs, heart, liver, spleen, kidney and intestine (Table 3). In trachea, the microscopic lesions included sloughing of epithelial lining, hemorrhages, hypertrophy of mediastinal lymph nodes and leukocytic infiltration. The microscopic lesions revealed atelactasis, emphysema, abscesses, thickening of alveolar septae, micro-thrombi, leukocytic infiltration and fibrosis in the lungs (Fig. 2). Congestion and leukocytic infiltration was also observed in the sections of hearts from group B. In liver hemorrhages, necrotic foci and infiltrations of leukocytes were found. The sections of kidneys also showed infiltration of leukocytes along with casts in tubules. In the intestinal mucosae, epithelial sloughing, hemorrhages and infiltration of leukocytes was observed.

Immunohistochemistry: Out of 12 goats infected with *Mmc* only seven (58.3%) were positive on immunohisto-chemical examination. The antigen was successfully detected in sections of lungs and lymph nodes. The positive immunohistochemical reaction was detected as red colored spots at the site of reaction. This red color of immune reaction was due to the substrate 3-amino-9-ethylecarbazol used in the reaction. In lungs, *Mmc* antigen was detected in the alveolar macrophages and in the walls of alveoli (Fig. 3). The immune reaction was observed in extracellular spaces and in the cytoplasm of macrophages. In lymph nodes, extensive immune reaction was detected around the germinal center and also scattered throughout the parenchyma.

DISCUSSION

Clinical signs observed in this study were similar but slightly severe as compared to the field outbreaks. Disease started with increased body temperature followed by nasal discharge initially catarrhal but later on mucopurulent in nature. In the severe form, animals became weak, reluctant to move and adopted a specific posture with abducted front legs. Eight animals suffered from diarrhea and only four animals exhibited nervous signs in the form of convulsions and bellowing. The results of the present study are justified by the findings of the previous studies (Gutierrez *et al.*, 1999; Laura *et al.*, 2006). In the present study, disease produced an acute septicemic clinico-pathological picture with lethal outcomes. However, these findings were not matched with the finding of Wesonga *et*

al. (2004). They observed that during the experimental trial of infection the disease showed a mild and chronic course. These differences in results might be due to the difference in age of experimental goats (12-15 months). Secondly, Wesonga *et al*. (2004) infected the animals with different species of *Mycoplasma*, i.e., *M. capricolum* subspecies *capripneumoniae*, for experimental production of disease. In the present study, the animals were at the age of 2-3 months and the species was *Mmc*. Another unique finding of the present study was the absence of joint inflammation. Similar findings were reported by Laura *et al.* (2006) working with CPP caused by *Mmc*. However, these findings were contrary to the study conducted in the same country by Awan *et al*. (2009) who reported lameness and arthritis in the forelimb of infected goats. The reason may be due to the difference in species of *Mycoplasma*.

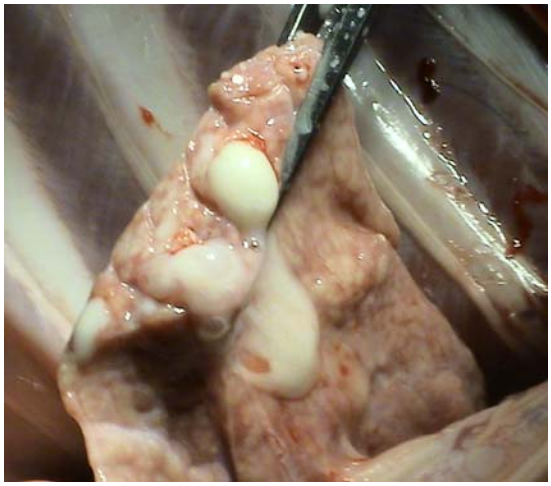

Fig. 1: Lungs of goat infected with *Mycoplasma mycoides capri* showing gross abscessation.

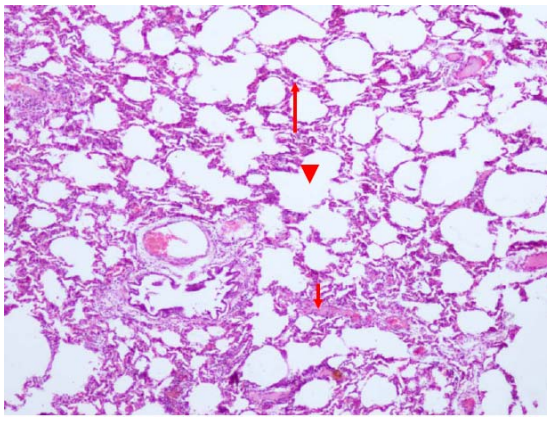

Fig. 2: Lungs section of kid infected with *Mycoplasma mycoides capri* showing Pulmonary emphysema (arrow head), mucous in the alveoli (short arrow) and thickened inter alveolar septa (long arrow). (H & E Stain 400X).

On postmortem examination, all the goats exhibited the classical lesions of CCP. Both unilateral and bilateral involvement of lungs was the striking finding of the present study. The same findings were reported by Gutierrez *et al*. (1999) and Wesonga *et al.* (2004). The lungs were marbled in appearance and in some kids hemorrhages were found on the surfaces. On incision, frothy to purulent exudates oozed out from the surface of the lungs. Hemorrhages were frequently observed on the surface of the trachea. The most classical finding of the present study was the presence of serosenguinous fluid in the pericardial sacs of inoculated animals that was slightly turbid in color. Similar findings were reported by Mondal *et al.* (2004) and Nicholas *et al.* (2008) working on the same species.

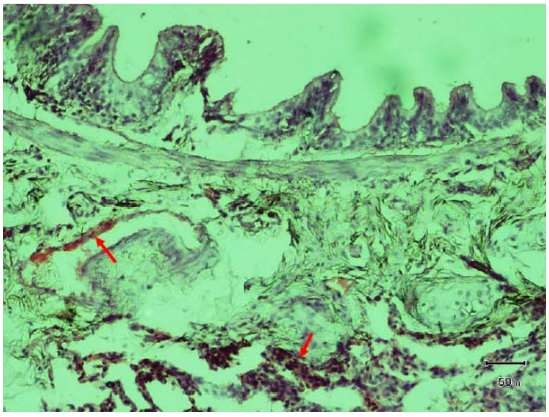

Fig. 3: Lungs of kid with induced pleuropneumonia showing the Positive immune reaction (arrow). (Streptavidin-biotin peroxidase complex method, 1000X).

Table 3: Clinical scoring* of microscopic lesions in experimentally infected goat kids with *Mycoplasma mycoides capri*

Organ/Lesion	Experimental Weeks				
	1	2	3	4	5
Trachea					
Sloughing of Epithelium	2	3	4	4	3
Hemorrhages	2	3	3	3	2
Hypertrophy of glands	2	3	3	3	2
Leukocytic infiltration	2	3	4	4	4
Lungs					
Atelactasis	2	3	3	3	3
Emphysema	2	3	3	3	3
Abscess	1	2	3	3	3
Thickening of Alveolar septa	2	3	3	4	3
Micro thrombi	1	2	3	4	2
Leukocytic infiltration	3	4	4	4	3
Fibrosis	1	2	3	2	4
Heart					
Congestion	1	2	3	4	2
Leukocytic infiltration	1	2	3	3	3
Liver					
Hemorrhages	1	2	3	3	3
Focal necrosis	1	2	3	4	3
Leukocytic infiltration	1	2	2	4	3
Kidney					
Cost in tubules	1	2	3	4	3
Leukocytic infiltration	1	3	3	3	3
Intestine					
Sloughing of Epithelium	1	2	3	3	3
Hemorrhages	1	2	4	4	3
Leukocytic infiltration	1	3	4	4	3
Cumulative lesions score	30	53	68	73	62

*Score: 1 = Normal, 2= Mild, 3 = Moderate, 4 = Severe

Livers in most of the kids were pale in color with necrotic foci and hemorrhages on the surfaces. The kidneys were slightly enlarged, congested and on incision pus was present in the pelvis region. The mesenteric lymph nodes were enlarged and the intestine showed

hemorrhages in animals suffering from diarrhea. These observations were matched with the findings of Laura *et al*. (2006) and Nicholas *et al*. (2008). This widespread distribution of lesions suggested the septicemic nature of the disease. This statement is justified by the findings of Nayak and Bhowmik, (1991) that reported erythro-cytopenia and disseminated intravascular coagulation caused by *Mmc* induced infection. The lesions observed in experimentally inoculated kids were consistent with those reported in pathogenic mycoplasmosis in goats (Hussain *et al*., 2012). The respiratory tract lesions were characterized by fibbrino-purulent pneumonia with dilation of the interlobular septa and fibrinous pericarditis in *Mmc* infection (DaMassa *et al*., 1992). The presence of micro-thrombi with heamorrhages represented septicemic nature of disease which ultimately led to disseminated intravascular coagulation.

This was a first attempt in Pakistan to use the immunohistochemical technique to detect *Mmc* antigen in the tissue sections. Immunohistochemical labeling reaction was positively demonstrated in lungs and lymph nodes. The antigen was found both in free forms in alveolar spaces and in the alveolar macrophages. Similar findings were reported by Wesonga *et al*. (2004). Positive immunohistochemical reaction was abundantly demonstrated in areas surrounding the germinal center in sections of mediastinal lymph nodes. This positive reaction strongly suggests that *Mmc* was the etiological agent of infection. The consistent presence of *Mmc* antigen in alveolar macrophages highlighted the role of these cells in the host defense mechanism.

In conclusion, the *Mmc* as the causative agent of the disease in the study areas was successfully isolated from a natural outbreak by using modified hay flick media and was identified through immunohistochemistry. The isolated species of *Mycoplasma* produced an acute septicemic form of disease in experimentally infected kids. The disease adopted similar patterns of pathological manifestation and course of disease as in the natural outbreak, but more severe with lethal outcome.

REFERENCES

Ahmad L, A Khan and MZ Khan, 2011. Cypermethrin induced biochemical and hepato-renal pathological changes in rabbits. Int J Agric Biol, 13: 865-872.

Arif A, J Schulz, F Thiaucourt, A Taha and S Hammer, 2007. Contagious caprine pleuropneumonia outbreak in captive wild ungulates at Al Wabra Wildlife Preservation, State of Qatar. J Zoo Wildl Med, 38: 93-96.

Awan MA, F Abbas, M Yasinzai, RAJ Nicholas, S Babar, RD Ayling, MA Attique and Z Ahmed, 2009. Prevalence of *Mycoplasma capricolum* subspecies *capricolum* and *Mycoplasma putrefaciens* in goats in Pishin district of Balochistan. Pak Vet J, 29: 179-185.

DaMassa AJ, PS Wakenell and DL Brooks, 1992. *Mycoplasma*s of goats and sheep. Review article. J Vet Diag Invest, 4:101-113.

Ettorre C, Sacchini F, Scacchia M. and Salda LD, 2007. Pneumonia of lambs in the Abruzzo region of Italy: anatomopathological and histopathological studies and localisation of *Mycoplasma ovipneumoniae*, Vet Ital, 43, 149-155

Gelagay A, S Teshale, W Amsaluc and G Esayas, 2007. Prevalence of contagious caprine pleuropneumonia in the Borana pastoral areas of Ethiopia, Small Rumin Res, 70: 131-135.

Goncalves R, Mariano I, Nunez A, Branco S, Fairfoul G and Nicholas R, 2010. Atypical non-progressive pneumonia in goats. Vet J, 183, 219-221.

Gutierrez C, JL Rodriguez, JA Montoya and A Fernandez, 1999. Clinico-pathological and haematological findings in goat kids experimentally infected simultaneously with *Mycoplasma mycoides subsp. capri* and *Mycoplasma mycoides* subsp. *mycoides* (large colony-type). Small Rumin Res, 31: 187-192.

Hasso SA, JM Aiaubaidi and AM Aidaraji, 1993. Contagious agalactia in goats, its severity as related to the route of infection and pregnancy. Small Rumin Res, 10: 263-275.

Hussain R, M Auon, A Khan, MZ Khan, F Mahmood and SU Rehman, 2012. Contagious caprine pleuropneumonia in Beetal goats. Trop Anim Health Prod, 44: 477-481.

Laura H, J Lopez, M St-Jacques, L Ontiveros, J Acosta and K Handel, 2006. *Mycoplasma mycoides* subsp. *capri* associated with goat respiratory disease and high flock mortality. Can Vet J 47: 366-369.

Mondal D, AK Pramanik and DK Basak, 2004. Clinico- Haematology and pathology of caprine *Mycoplasma* pneumonia in rain fed tropics of West Bengal. Small Rumin Res, 51: 285-295.

Nayak NC and MK Bhowmik, 1991. *Mycoplasmal* polyarthritis associated with septicaemia in kids: Clinico-haematological and biochemical studies. Indian J Vet Pathol, 12: 13-17.

Nicholas R, R Ailing and MCA Laura, 2008. *Mycoplasma* disease of small ruminants. CAB International, pp: 114-131.

OIE, 2008. Contagious Caprine Pleuropneumonia. In Terrestrial manual. Chapter 2.7.6: pp 1000-1012.

Pettersson B, T Leitner, M Ronaghi, G Bolske, M Uhlem and KE Johansson, 1996. Phylogeny of *Mycoplasma mycoides* cluster as determined by sequence analysis of 16 S rRNA genes from the two rRNA operons. J Bacteriol, 178: 4131- 4142.

Rahman SU, M Siddique, I Hussain, K Muhammad and MH Rasool, 2003. Standardization of indirect haemagglutination test for monitoring *Mycoplasma mycoides* subspecies *capri* antibodies raised in rabbits and goats. Int J Agri Biol, 5: 295-297.

Regassa F, M Netsere and T Tsertse, 2010. Sero-prevalence of contagious caprine pleuropneumonia in goat at selected Woredas of Afar Region. Ethiop Vet J, 14: 83-89.

Rodriguez JL, J Oros, F Rodriguez, JB Poveda, A Ramirez, A Fernandez, 1996. A Pathological and immunohistochemical study of caprine pleuropneumonia induced by subspecies of *Myeoplasma rnyeoides*. J Comp Path, 114: 373-384.

Srivastava AK, Meenowa D, Barden G, Salguero FJ, Churchward C and Nicholas, RAJ, 2010. Contagious caprine pleuropneumonia in Mauritius, Vet Rec, 167, 304-305.

Thiaucourt F and G Bölske, 1996. Contagious caprine pleuropneumonia and other pulmonary mycoplasmoses of sheep and goats. *Rev Sci Tech Off Int Epiz,* 15: 1397-1414.

Wesonga HO, G Bölske, F Thiaucourt, C Wanjohi and R Lindberg, 2004. Experimental contagious caprine pleuropneumonia: A long term study on the course of infection and pathology in a flock of goats infected with *Mycoplasma capricolum* subsp. *capripneumoniae*. Acta Vet Scand, 45: 167-179.