

Original Research Article

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Seroprevalence of Leptospirosis among Aborted Goats in Kerala

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ABSTRACT

A total of 62 serum samples were obtained from goats with history of abortion from Teaching Veterinary Clinical Complex (TVCC) and University Sheep and Goat Farm, College of Veterinary and Animal Sciences (CVAS), Mannuthy as well as various private farms and households from villages in Thrissur, Malappuram and Kannur districts of Kerala. The samples were examined for antileptospiral antibodies by microscopic agglutination test (MAT) using 12 serovars viz., Australis, Autumnalis, Bataviae, Canicola, Grippotyphosa, Hebdomadis, Icterohaemorrhagiae, Javanica, Pomona, Pyrogenes, Sejroe and Tarassovi maintained. It was found that 16 samples (25.81 per cent) were positive. Seroreactivity was observed against the serovars Australis, Autumnalis, Bataviae, Canicola, Grippotyphosa, Hebdomadis, Javanica, Pomona, Pyrogenes and Sejroe. Serovar Pomona (23.08 per cent) was found to be the most prevalent one. The other serovars were Australis, Sejroe (15.38 per cent each), Bataviae, Canicola, Hebdomadis, Javanica, Pyrogenes (7.69 per cent each), Autumnalis and Grippotyphosa (3.84 per cent each). Among the seropositive animals, six were found to have mixed infections with more than one serovar. Seropositivity was found higher in goats above three years of age. Present study reveals the seroprevalence of leptospirosis in aborted goats and also indicates the circulation of multiple serovars among goats in Thrissur, Malappuram and Kannur districts of Kerala.

Keywords

Goats,
Leptospirosis,
MAT, Kerala,
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Introduction

Leptospirosis, a transmissible disease of animals and human beings, is caused by pathogenic members of the genus *Leptospira*. It is a neglected zoonotic disease with worldwide distribution and is endemic in Kerala. More than 300 serovars of *Leptospira* have been identified (Picardeau, 2017). Rats, mice and moles serve as the primary hosts for

transmission, while other mammals including dog, cat, cattle, goats, sheep and pig act as secondary hosts. It affects almost all domestic and wild mammals causing severe renal and hepatic damage leading to fatal conditions, if untreated. The disease is of considerable economic importance in livestock due to manifestations like abortion, infertility and decreased production (Faine *et al.*, 2000). Goats are vital part of rural farming to landless

and marginal farmers. They are reared mainly by grazing thus making them susceptible to several infectious diseases including leptospirosis. In India, the seroprevalance of caprine leptospirosis has been reported previously (Verma *et al.*, 2001; Sivaseelan *et al.*, 2003; Balakrishnan *et al.*, 2008; Meenakshisundaram and Chellapandian, 2010; Krishna *et al.*, 2012; Vihol *et al.*, 2016; Vihol *et al.*, 2017). In goats, leptospirosis manifests as a sub-clinical infection and most often goes undiagnosed. The disease occurs as both acute and chronic infections with clinical manifestations like anorexia, pyrexia, jaundice and haemorrhagic syndrome in acute condition (Faine *et al.*, 2000), whereas in chronic form symptoms like impaired fertility, neonatal deaths, abortions and decreased milk production are noticed (Lilenbaum *et al.*, 2007). Affected goats develop chronic renal infection and act as carriers, transmitting the leptospires to other animals through urine and contaminates the surroundings (Lilenbaum *et al.*, 2008). In India, particularly in Kerala, the studies on leptospirosis in goats are limited. Hence, the present study was undertaken to identify the seroprevalance and serovar distribution of *Leptospira* among goats with history of abortion from Thrissur, Malappuram and Kannur districts of Kerala.

Materials and Methods

Sample collection

A total of 62 serum samples were collected from aborted goats belonging to different age groups from private farms and households in different villages of Thrissur, Malappuram and Kannur districts of Kerala and also from Teaching Veterinary Clinical Complex (TVCC), University Sheep and Goat Farm, College of Veterinary and Animal Sciences (CVAS), Mannuthy. The abortions were reported during early to late stages of pregnancy.

Microscopic Agglutination Test

Microscopic Agglutination Test (MAT) was carried out using a battery of 12 live *Leptospira* antigens (serovars Australis, Autumnalis, Bataviae, Canicola, Grippotyphosa, Hebdomadis, Icterohaemorrhagiae, Javanica, Pomona, Pyrogenes, Sejroe and Tarassovi) as described by Faine (1982). A 1:100 serum dilution was prepared in PBS, 50 µL of which was taken in 96 well microtitre plates and mixed with 50 µL of each of the five to ten day-old live leptospiral serovars separately.

Antigen controls were set with 50 µL PBS and 50 µL of different live leptospiral serovars and the plates were incubated at 37°C for two to three hours. After incubation, the results were read by examining a drop of serum-antigen mixture from each well under low power objective of a dark field microscope for agglutination of leptospires.

Further, quantitative assay was carried out in 96 well microtitre plates against the reacting serovars of leptospires. All the 96 wells were filled with 50 µL PBS. In the first well of each row, 50 µL of 1 in 50 diluted serum samples were added and mixed well. Then, serial double fold dilutions were made up to eight wells. From the eighth well, 50 µL was discarded.

A constant volume of 50 µL of a particular serovar with a density of 2×10^8 cells per mL was added in each row and incubated at 37°C for two to four hours. All the final dilution mixtures (100, 200, 400, 800, 1600, 3200, 6400, 12800) were observed under dark field microscope and the results were recorded. The reciprocal of the highest dilution of the serum which showed 50 per cent agglutination or 50 per cent reduction in the number of free leptospires in comparison to control was considered as the respective titre.

Statistical analysis

Chi-square test was performed to determine the association between age-wise and district wise seroprevalence rate using SPSS software version 24.0.

Results and Discussion

Out of the 62 samples, antileptospiral antibodies were detected in 16 samples revealing a seropositivity of 25.81 per cent. The findings were in accordance with earlier observations made in goats (Faine *et al.*, 2000; Lilenbaum *et al.*, 2007; Lilenbaum *et al.*, 2008; Balakrishnan 2012; Vihol *et al.*, 2017). They recorded higher prevalence in aborted animals. In goats, reproductive problems caused by leptospirosis might be due to colonisation of leptospire in the reproductive tract of the goats which impairs fertility (Dehkordi *et al.*, 2011). The serovars prevalent in seropositive goats are summarised in table 1.

Serovar Pomona (23.08 per cent) was found to be the most prevalent one. The other serovars were Australis, Sejroe (15.38 per cent each),

Bataviae, Canicola, Hebdomadis, Javanica, Pyrogenes (7.69 per cent each), Autumnalis and Grippotyphosa (3.84 per cent each). Leon-Vizcaino *et al.*, (1987) and Vihol *et al.*, (2017) reported the involvement of serovar Pomona in aborted goats. This study revealed the presence of mixed infections. Tripathy *et al.*, (1985), Balakrishnan (2012) and Vihol *et al.*, (2017) also reported the association of more than one serovars in aborted goats. Balakrishnan (2012) reported the involvement of five serovars *viz.*, Australis, Hardjo, Hebdomadis, Icterohaemorrhagiae and Pomona in goats.

The highest seroprevalence was noticed in goats above three years of age. Out of 35 samples, 11 were positive in the above-mentioned age group. This was followed by goats in the age group of one to three years. The age-wise seroprevalence is given in table 2. On statistical analysis, no significant difference could be noticed among different age groups and seropositivity. However, Talebkhan *et al.*, (2003), Hassanpour *et al.*, (2008), Balakrishnan (2012) and Vihol *et al.*, (2017) reported that the age and incidence of leptospirosis are highly correlated.

Table.1 Seroprevalance of leptospirosis by MAT

Serovar	No. of Positive samples	Per cent Positivity (%)
Australis	4	15.38
Autumnalis	1	3.84
Bataviae	2	7.69
Canicola	2	7.69
Grippotyphosa	1	3.84
Hebdomadis	2	7.69
Icterohaemorrhagiae	0	0.00
Javanica	2	7.69
Pomona	6	23.08
Pyrogenes	2	7.69
Sejroe	4	15.38
Tarassovi	0	0.00

Table.2 Age wise distribution

Age	No. of samples collected	No. of positive samples	Per cent positivity
1-3 years	27	5	18.51
> 3 years	35	11	31.42
Total	62	16	25.81
			X²
			1.327^{NS} (P>0.05)

NS - Non-significant

Table.3 District wise distribution

Age	No. of samples collected	No. of positive samples	Per cent positivity
Thrissur	41	14	34.15
Malappuram	10	2	20.00
Kannur	11	0	0
Total	62	16	25.81
			X²
			5.495^{NS} (P>0.05)

NS - Non-significant

On statistical analysis, no significant difference could be noticed between district wise seroprevalence (P<0.05) (Table 3).

From this study, the seroprevalence of leptospirosis in aborted goats in Thrissur, Malappuram and Kannur districts of Kerala was studied. The overall seropositivity was found to be 25.81 per cent and Pomona was found to be the prevalent serovar. Although, seropositivity was noticed more in goats above three years of age, the statistical analysis revealed that the age and seropositivity are not associated significantly. This study also revealed the circulation of various leptospiral serovars in the area under study. Leptospirosis plays a major role in reproductive disturbances in goats and it should be considered while diagnosing any reproductive problems where infection is presumed. The seropositive goats may contaminate the surroundings by excreting leptospiral organisms through urine, act as a threat to other animals and human beings and should be subjected to detailed study.

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