

## SEROEPIDEMIOLOGICAL STUDY OF LEPTOSPIROSIS IN BUFFALOES OF SOUTH GUJARAT, INDIA

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### ABSTRACT

A total of 102 serum samples were collected randomly from buffaloes exposed with different clinical conditions (abortion, repeat breeding, fever, mastitis, anorexia) suspected for leptospirosis and apparently healthy. These serum samples were subjected to seroepidemiological study using microscopic agglutination test (MAT) having different serovars of *Leptospira* spp. The seroprevalence of leptospirosis among buffaloes was noted to be 15.69% (16/102). All the three districts of South Gujarat showed the presence of leptospiral antibodies without any significant difference ( $P \leq 0.05$ ) with the highest rate in Tapi (50.00%) followed by Navsari (14.89%) and Surat (13.72%). Jafrabadi breed showed 50.00% seropositivity followed by Surati (16.67%), Mehsana (15.00%) and Non-Descript (5.55%). In female buffaloes the seroprevalence positivity was noted in 16.49% cases. However, none of male exhibited seropositivity. In respect of age groups the highest rate of seropositivity (19.23%) was observed in age group of 1 to 4 years followed by above 4 years (15.71%) and below 1 year

(00%) without significant difference ( $P \leq 0.05$ ). In buffaloes out of 102 sera screened, 16 were positive with one or more serovars. The highest number of seropositivity was recorded against serovar Kaup (17.39%).

**Keywords:** buffaloes, Leptospirosis, seroepidemiology, zoonosis, MAT

### INTRODUCTION

Leptospirosis is an economically important widespread zoonotic disease caused by pathogenic species of *Leptospira interrogans* occurs in man and different species of animals like cattle, buffalo, sheep, goat, deer, pig, rodents, camel, horse, sealion, shank, raccoon (Cutler *et al.*, 2005; Cheema *et al.*, 2007). The causative agent is frequently excreted through urine of infected individual and contaminate the environment, there by exposing human being especially farmers, agriculture labour and animal holders.

The state of Gujarat, Maharashtra, Tamil Nadu, Kerala and Andaman and Nicobar Islands

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are endemic in India and incidences in humans are perennially reported from these states during monsoon season (Maskey *et al.*, 2006). Hence the present seroepidemiological study was conducted to determine the magnitude of occurrence pattern of leptospirosis in buffaloes, reared in rural areas of various district i.e. Navsari, Surat and Tapi of South Gujarat.

## MATERIALS AND METHODS

### Animal sera

A total of 102 serum samples were collected randomly from different age groups and breeds of buffaloes (n=102) of either sex reared in villages of various districts (Navsari, Surat, Tapi) of south Gujarat (Table 1). Whole blood samples were collected from jugular vein directly or during slaughter of buffaloes in sterile 9.0 ml plain vacutainers. To obtain serum, whole blood was kept in slanting position in 9.0 ml plain vacutainers until serum extracted out of the whole blood. Then these 9.0 ml plain vacutainers were centrifuged at 7000 rpm for 10 minutes. The straw coloured serum was collected into 1.5 ml sterile cryo vials and aliquoted and stored at -20°C for carrying out MAT.

### MICROSCOPIC AGGUTINATION TEST (MAT)

All the sera were tested for antibodies against live antigens of *Leptospira* sp. serovars Pyrogenes, Australis, Bankinang, Grippotyphosa, Patoc, Pomona, Icterohaemorrhagiae, Hebdomadis, Canicola, Hardjo, Bellum, Bataviae, Tarassovi, Shermani, Kaup, Hurstbridge and Javanica by

Microscopic agglutination test at Leptospirosis Reference Laboratory, Government Medical College, Surat (Vijayachari *et al.*, 2001) and Project Directorate on Animal Disease Monitoring and Surveillance (PD-ADMAS), Bangalore using standard procedure (WHO-OIE, 2013; Faine, 1982).

## STATISTICAL ANALYSIS

Chi-square test was used according to WEB AGRI STAT PACKAGE software developed by Jangam and Wadekar, ICAR research complex, Goa for statistical analysis of data (Jangam and Wadekar, 2012).

## RESULTS AND DISCUSSION

In the present study a total 102 sera were screened from three different districts of South Gujarat (Navsari, Tapi, Surat) for leptospiral antibodies. The details of district, breed, sex and age wise seroprevalence results are depicted in Table 1.

The seroprevalence of leptospirosis among buffalo was noted to be 15.69% (16/102) and was comparable to reported prevalence of 14.55% in Gujarat (Savalia, 2001), 14.7% in Uttaranchal (Agrawal *et al.*, 2005) and 15% in Uttaranchal, Tamil Nadu and Uttar Pradesh (Mariya *et al.*, 2007). In contrast to above findings higher seroprevalence was 54.14% in Gujarat (Balakrishnan *et al.*, 2011), 26.66% in Andaman and Nicobar Islands (Varma *et al.*, 2001) and 88.8% (125/111) in Chennai buffaloes (Selvaraj *et al.*, 2010) have been reported.

All the three districts of South Gujarat showed the presence of leptospiral antibodies

without any significant difference ( $P \leq 0.05$ ) with the highest rate in Tapi (50.00%) followed by Navsari (14.89%) and Surat (13.72%). Comparable findings of leptospirosis seroprevalence were reported by Savalia (2001) from Valsad (17.24%), Navsari (19.35%) and Surat (22.50%) districts.

No significant difference in seroprevalence was observed among samples tested from the different breeds of buffalo. Jafrabadi breed showed 50.00% seropositivity followed by Surati (16.67%), Mehsana (15.00%) and Non-Descript (5.55%). Contrary to this Balakrishnan *et al.* (2011) reported Murrah breed to be most susceptible (58.25%) followed by Pandharpuri (40.91%) and Jaffrabadi (37.50%). In the present study prevalence rate of leptospirosis in Jafrabadi breed of buffalo was higher (50.00%) than reported earlier by Balakrishnan *et al.* (2011). There is every likelihood that the breed susceptibility reported by various workers is influenced by the sample size in particular area where specific breed might be prevalent but need further elucidation.

In female buffaloes the seroprevalence positivity was noted in 16.49% cases. However, none of male exhibited seropositivity. Possibly because a wide gap occurred in the number of samples processed (Male-5 and Female-97).

In respect of age groups (above 4 years, 1 to 4 years and below 1 year) the highest rate of seropositivity (19.23%) was observed in age group of 1 to 4 years followed by above 4 years (15.71%) and below 1 year (00%) without significant difference ( $P \leq 0.05$ ). It was not in agreement with findings of Balakrishnan *et al.* (2011) who observed maximum seropositivity (77.05%) among buffaloes between 4 to 7 years (adult group) followed by above 7 years (75.00%, older age group) and below 4 year (26.67%) with significantly different among age group ( $P \leq 0.01$ ). Agrawal *et al.* (2005) studied

the seroprevalence in cattle, buffaloes and goats and reported higher seropositivity in buffalo above 9.0 years of age and concluded that seropositivity increases with advancing age irrespective of animal species involved.

A total of 102 sera screened, 16 were positive with one or more serovars. In the present study highest number of seropositivity was recorded against serovar Kaup (17.39%) followed by Grippytyphosa (13.04%), Pomona (13.04%), Javanica (13.04%), Patoc (8.70%), Canicola (8.70%), Hardjo (8.70%), Bataviae (8.70%), Autumnalis/Bankinang (4.35%), Hurstbridge (4.35%). As against the present observations the most common serovars in Indian buffaloes reported by earlier workers from different states included Hardjo (Agrawal *et al.*, 2005; Balakrishnan *et al.*, 2011), Grippytyphosa (Varma *et al.*, 2001) and Pomona (Selvaraj *et al.*, 2005; Selvaraj *et al.*, 2010). However serovars distribution seen in South Gujarat region in present investigation was comparable with the findings of Savalia (2001) and Balakrishnan *et al.* (2011) who reported serovars Hardjo, Grippytyphosa, Australis, Hebdomadis, Ballum and Pomona. Other serovars reported from different states of India enlisted Grippytyphosa, Pomona and Australis from Andaman and Nicobar (Varma *et al.*, 2001), Hardjo, Javanica and Australis from Uttaranchal (Agrawal *et al.*, 2005), Pomona, Hebdomadis, Tarassovi, Sejroe, Australis, Pyrogenes, Autumnalis, Grippytyphosa, Ballum, Javanica, Icterohaemorrhagiae, Canicola from Chennai (Selvaraj *et al.*, 2005; Selvaraj *et al.*, 2010).

Major areas of South Gujarat districts are used for paddy and sugarcane cultivation and are rich in natural vegetation with plenty of marshy lands and small ponds/water logging areas with almost neutral pH, suitable humidity and

Table 1. Seroprevalence of leptospirosis in buffaloes.

Attributes	No. of Tested	No. of Positive	Percent Positive
<b>Region</b>			
South Gujarat	102	16	15.69
<b>Districts</b>			
Navsari	47	07	14.89
Tapi	04	02	50.00
Surat	51	07	13.72
Total	102	16	15.69
$\chi^2 = 5.99$ <sup>NS</sup> (P<0.05)			
<b>Breed wise</b>			
Surati	60	10	16.67
Mehsani	20	03	15.00
Jafrabadi	04	02	50.00
Nondiscript	18	01	5.55
Total	102	16	15.69
$\chi^2 = 7.82$ <sup>NS</sup> (P<0.05)			
<b>Sex wise</b>			
Male	05	00	00
Female	97	16	16.49
Total	102	16	15.69
$\chi^2 = 3.84$ <sup>NS</sup> (P<0.05)			
<b>Age wise</b>			
<1 year	06	00	00
1-4 years	26	05	19.23
>4 years	70	11	15.71
Total	102	16	15.69
$\chi^2 = 5.99$ <sup>NS</sup> (P<0.05)			

Note: <sup>NS</sup>-Non significant at P<0.05

temperature which are optimum for growth/survival of leptospires and perpetuation in the environment. Cultivated fields infested with rats, a carrier for leptospires (Collares-Pereira *et al.*, 2000) also play an important role in spreading the infection. Prevalence in buffalo could be attributed to their habit of wallowing in water bodies contaminated with infected urine (Trembl *et al.*, 2002) as one of the main sources of transmission of leptospires.

The present study concludes that seroprevalence of leptospirosis among buffaloes in South Gujarat region was ranged from 14.55% (Savalia, 2001) to 54.14% (Balakrishnan *et al.*, 2011) with presently reported seroprevalence was 15.69% without significant difference between age, breed, sex and different district. The continuous presence of Leptospires in this area lead to potential zoonotic risk to slaughter house workers, meat inspectors, animal holder, agriculture labour and farmers. This study also determines the need for continuous monitoring of leptospirosis in animal and humans to combat this zoonotic infection.

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