



ORIGINAL RESEARCH ARTICLE

Seroprevalence of leptospirosis in Chennai city, India.

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Abstract: Leptospirosis is a zoonotic disease, it occurs worldwide but is most common in tropical and subtropical countries. The magnitude of the problem varies from community to community. The serovars which cause the disease in a community change with adaptation to a new maintenance host and is epidemiologically important. Aims and Objectives: To determine the seroprevalence of leptospirosis in Chennai city. To identify the prevalent serovar in Chennai causing leptospirosis by using gold standard test Microscopic Agglutination Test (MAT). Methods: 1209 patients from Chennai, with fever of one-week duration and with signs and symptoms of Leptospirosis were the study population and were screened for Leptospirosis by Macroscopic Slide Agglutination Test (MSAT). The positive samples by MSAT were tested for the prevalent serovar and for confirmation by MAT and the results were analyzed statistically. Result: The seroprevalence of Leptospirosis is 17.8%. The prevalent serovar circulating in Chennai is *L. grippityphosa*. Conclusion: Laboratory diagnosis is essential to confirm the current illness, and for epidemiological and public health reasons, namely to determine which serovar caused the infection, the likely source of infection and the potential reservoir and its location. This helps in control strategies.

Key words: Leptospirosis; MSAT; MAT; seroprevalence; serovar.

Introduction

Leptospirosis is a zoonotic disease, occurs worldwide, but is most common in tropical and subtropical areas with high rainfall. Leptospirosis is becoming an increasingly significant public health problem, particularly in tropical developing countries. The disease is found mainly in places where humans come into contact with the urine of infected animals or a urine polluted environment.

It presents with signs and symptoms similar to other diseases like influenza, dengue fever, hepatitis, meningitis and mimic them, so laboratory diagnosis of leptospirosis is essential. Leptospirosis is potentially lethal but it is a treatable disease. Case fatality rate for severe forms of leptospirosis is 5 to 40%, respectively. (Bharti *et al.*, 2003; Faine *et al.*, 1999; Farr 1995 & Mc Bride 2005). Antibiotic therapy has proved useful when administered in the early stage of the disease, thus prompt identification of the disease is important (Faine *et al.*, 1999; WHO., 2003). An understanding of the transmission cycle of the disease in the community is important.

Even though the basic modes of transmission of leptospirosis i.e., presence of carrier animals, contaminated environment which helps in the survival of leptospires and the people's behaviour and their occupation are common, the magnitude and nature of these factors differs from community to community. Therefore, knowledge of the risk factors, and the factors which can be modified is essential to plan control strategies. Chennai being endemic for leptospirosis, studying the above factors becomes essential.

Finding out the prevalent serovar of leptospirosis causing the disease in an endemic area is important in epidemiological point of view, as different serovars may develop a mild pathogenic relationship with a certain animal host species (1). Because of the development of new maintenance host, the prevalence of different serovars also changes accordingly (2).

Serology becomes an important diagnostic tool from the moment antibodies appear in detectable quantities in blood (3). MSAT is a valuable screening test, and MAT is the gold standard laboratory test in the diagnosis of leptospirosis, and can determine the infecting serovar. The study was conducted with an objective of assessing the seroprevalence of leptospirosis in Chennai city which is endemic for the disease, and hence control strategies could be more effective.

Materials and Methods

The study was conducted in Leptospirosis Research Cell, Madras Medical College and hospital over a period of one-year duration from November 2012 to October 2013 after getting Ethical clearance. 1209 patients and who attended Rajiv Gandhi Government General Hospital, with fever of one-week duration, having signs and symptoms of leptospirosis were included in the study. Our hospital received samples from patients from nearby Government and private hospitals in Chennai.

The demographic and clinical profile of the patients involved in the study was obtained by standard questionnaire. The serovars used for MSAT and MAT are *australis*, *autumnalis*, *canicola*, *grippityphosa*,

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hebdomadis, icterohaemorrhagiae, louisiana and *semaranga*. The above standard strains were obtained from Royal Tropical Institute, Netherlands and sub cultured in LRC, Institute of Microbiology, M.M.C.

Under aseptic precautions 5ml of venous blood sample was collected in a clean dry test tube. Blood was centrifuged serum separated and was subjected to serological test MSAT. MSAT was done using a dense suspension of killed Leptospire was mixed with a drop of serum on a slide and rotated on a rotator (120 rpm) for 4 min. It was then examined by naked eye for presence of agglutination. Clumps of agglutination with complete clearing of leptospiral antigen was considered significant. The 237 patients who were positive by screening test MSAT were subjected to MAT for confirmation and to find out the prevalent serovar. The cut-off point for a positive MAT was a titre of $\geq 1:80$, giving a total number of 216 positive MAT cases.

Results

1209 samples were screened for leptospirosis were confirmed by MAT in 216 patients which gives a seroprevalence of 17.8%. 129 patients were males and 87 patients were females. More than 50% of the affected patients were outdoor manual workers.

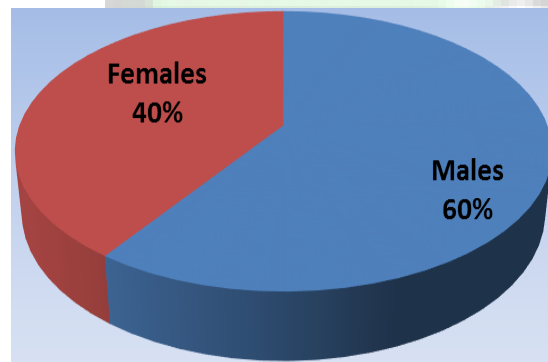


Figure 1: Gender distribution among positive cases (n=216)

Males were found to be more commonly affected by leptospirosis than females.

Age In Years	No Of Patients	Percentage
<20	26	12.0
21-30	64	29.6
31-40	56	25.9
41-50	34	15.7
51-60	25	11.5
61-70	7	3.2
>70	4	1.85

The leptospiral infection was relatively higher in the age group of 20-40 years.

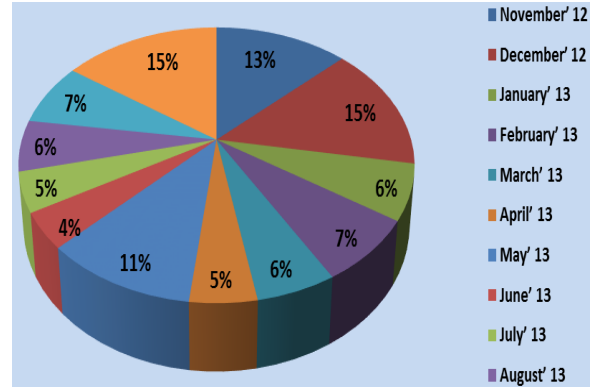


Figure 2: Month wise distribution of infected cases (n= 216)

There was clustering of cases seen during the monsoon period and during the month of May.

Table 3: Zone wise distribution of leptospirosis cases (n=216), There are 15 zones in Chennai.

Zone division	Zone	No of patients	Percentage
1, 2, 4, 5, 6, 7	Chennai north	83	38.4
3	Thiruvallur	51	23.6
8, 9	Chennai central	35	16.2
10, 11, 12, 13	Chennai south	29	13.4
14, 15	Sholinganallur	18	8.3

38.4% of patients affected with leptospirosis were from Chennai North.

Table 4: MSAT result among screened cases (n=1209)

Total Samples	Positive	Negative
1209	237	972

237 cases were positive by the screening test MSAT.

Table 5: correlation of results of MSAT and MAT

MSAT Positive Samples	MAT Results In Titre			Total (Confirmed Cases)	Prevalence
	1/80	1/160	1/320		
237	155	61	-	216	17.8%

The seroprevalence of leptospirosis in Chennai is 17.8%.

Table: 6: MAT titre and Serovar distribution in confirmed cases (N=216)

Serovar	MAT Titre		Total	%
	1:80	1:160		
<i>L. grippityphosa</i>	54	19	73	34%
<i>L. icterohaemorrhagiae</i>	33	21	54	25%
<i>L. hebdomadis</i>	16	12	28	13%
<i>L. australis</i>	15	6	21	9.7%
<i>L. autumnalis</i>	12	6	19	8.6%
<i>L. semaranga</i>	9	5	14	6.45%
<i>L. canicola</i>	4	1	5	2.1%
<i>L. louisiana</i>	-	2	2	1%
TOTAL			216	

L. grippityphosa was the predominant serovar causing infection in leptospirosis confirmed patients.

Discussion

Chennai is one of the most populous metropolitan cities in India. Chennai has a long coastline and is the home of several millions of people. Due to rapid changes in the ecological system many zoonotic diseases have caused epidemics. Leptospirosis is one of them and is endemic in Chennai. The infection is of public health importance due to its morbidity and mortality. Finding out its seroprevalence in Chennai and the zone most affected, and the category of people most affected would help take preventive measures to control the spread of the disease.

Leptospirosis mimics many other diseases in its atypical presenting symptoms and clinical features and thus laboratory diagnosis of the disease is of utmost importance. Serological test is the test most commonly used to diagnose leptospirosis due to difficulty in its isolation, lack of sensitivity and specificity in dark field microscopy in laboratory diagnosis of leptospirosis (4).

The present study was conducted among 1209 patients in Chennai city with clinical features suggestive of leptospirosis, 216 cases were positive by MSAT. Usefulness of the test as a screening test was evaluated by Sumathi *et al.*, (5, 6). Out of 216 patients 129 (59.7%) patients were males which indicate that male preponderance in leptospirosis due their outdoor activities and occupation. This correlates with the Pappachan *et al.*, (7) study in which 58.9% patients were males. In a study conducted by Shivakumar *et al.*, 88% of the patients were males, this is in contrary to the present study.

The age categories of the patients most affected are of 20–40 years. The mean ages of the affected patients were 36.4 years who are young adults and are the active population involved in outdoor activities and recreational activities and hence an increased chance of exposure to infection. In a study conducted by Margarita R *et al.*, (8), the mean age of the patients affected with leptospirosis was 36 years which correlates with the present study. Study conducted by Shivakumar *et al.*, recorded the mean age of the affected patients to be 39.6 years.

30% of the patients were affected during Oct, Nov and Dec which proves the maximum spread of leptospirosis during the monsoon period and this correlates with the study of Sharma K. K. *et al.*, (9) study that highest incidence of leptospirosis occurred during rainy season, may be due to polluted environment. In another study conducted in Chennai by Ganesan Arumugam *et al.*, the more number of positive cases were recorded during the monsoon period, this correlated with the present study. There was clustering of cases during the month of May, the reason may be due to more number of people involving in leisurely activities

and recreational activities during the holidays like swimming in pools and water sport (10).

More than 50% of the patients affected by leptospirosis were outdoor manual workers. Most of them were agricultural labourers, masons, sewage workers and plumbers. This establishes the fact that persons who are more exposed to contaminated environment are at a higher risk of contracting the disease (11). In a study conducted by Shivakumar *et al.*, in 1993, 49% percentages of the patients affected were outdoor manual workers. In a study conducted by S Sharma *et al.*, (12) in Andaman during 2006 the percentage of people affected was 59.6% which correlates with the present study.

The most predominant symptom was fever which was seen in 100% of the patients followed by headache (81.4%), myalgia (74%), vomiting (28.07%), conjunctival suffusion (25.9%), high coloured urine (25%). The other symptoms which were present among patients in the study are abdominal pain (27.3%), diarrhoea (20.08%), jaundice (12.03%), joint pain (9.7%), dysuria (4.16%), skin rashes (2.7%) and seizures (0.4%).

In De A *et al.*, study fever was present in 100% of cases, myalgia in 51.35% of cases, jaundice and conjunctival suffusion in 32.43% of the patients. In a study conducted by Linda Anderson *et al.*, in Andaman Islands in 2006, fever formed 100% of the cases, body ache 80%, headache and chills 40%, vomiting 33%, pain abdomen 27%, diarrhoea 27% and icterus 13% which correlates with the present study.

The difference in varying symptoms and signs in our study may be due to the fact that, through years the signs and symptoms of the disease keep changing due the change in the serovar pattern causing the disease in a particular locality. In the previous years the severity of the disease was high and there were more complications like pulmonary haemorrhage and renal failure (13,14). The complications of the disease have considerably decreased over years due to increased clinical suspicion, awareness about the disease, early diagnosis and institution of appropriate antibiotics. There was no mortality observed in the present study.

North Chennai is mostly affected and 50% of the cases are from this region which is endemic for leptospirosis which belongs to the zone 1, 2,4, 5, 6 and 7. In a study conducted by Muruganath, Shivakumar *et al.*, in North Chennai in 2009, the percentage of patients affected due to leptospirosis was 28.6% which is contrary to the present study. This is due to the fact that most of the urban slums are in North Chennai and people live under unhygienic environmental conditions prevailing in

the area which an important epidemiological factor in the spread of leptospirosis.

The seroprevalence of leptospirosis in the present study is 17.8%. The seroprevalence in Chennai was 32.9% in 1993 (Shivakumar *et al.*). In a study conducted by Chinari Pradeep *et al.*, (15) during 1995-1997 the prevalence rate was 31%. Leptospirosis has also declined since 1995. In a recent study conducted in Chennai in 2012 the prevalence rate was 19.5%. Prevalence of Leptospirosis shows wide variation. (16) and there is a consistent decrease in the prevalence of leptospirosis and its severity may be due to the fact that increasing suspicion about the disease in clinicians and increasing awareness among public. The persistence of mild Leptospirosis suggests that the environmental risk factors (Infected rodents and domestic animals, contaminated environment and rainfall) play an important role in the continuous occurrence and the spread of the disease.

In the present study the predominant serovar was *L. grippityphosa* (34%) followed by *L. icterohaemorrhagiae* (25%), *L. hebdomadis* (13%), *L. australis* (9.7%) and *L. autumnalis* (8.6%). Along with these serovars, serovar *L. semaranga*, *L. canicola* and *L. louisiana* were also found in few cases. Sumathi *et al.*, study during 2004 – 2006 reported the predominant serovar was *L. icterohaemorrhagiae* (48%), which was followed by *L. australis* (37%) and *L. grippityphosa* (26%). In a study conducted by Shivakumar *et al.*, during 1997 the predominant serovar was *L. autumnalis* (59.9%) followed by *L. icterohaemorrhagiae* (15.5%). This reveals that serovar group keeps changing, and different serovars become predominant infecting serovar during different periods.

Conclusion

The serovar concept is of epidemiological significance. The prevalence of the different serovars changes as a result of new maintenance host. Seroprevalence of leptospirosis in Chennai is 17.8%. The predominant serovar which caused leptospirosis in Chennai in the present study is serovar grippityphosa which is mostly spread by rodents which would have contaminated water sources during monsoon period responsible for maximum spread of the disease during that period. Early recognition and appropriate treatment leads to favourable outcome of this potentially life threatening condition.

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