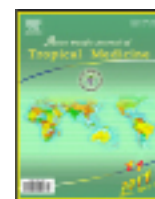




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# Serologic evidence of human leptospirosis in and around Kolkata, India: A clinico–epidemiological study

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## ABSTRACT

**Objective:** To investigate the prevalence of leptospirosis among patients from within and outside Kolkata, India, attending the Calcutta School of Tropical Medicine, for treatment during August 2002 to August 2008. **Methods:** The leptospirosis cases were determined on the basis of clinical, epidemiological, and biochemical factors, and were tested for leptospiral antibodies using IgM ELISA. Serum samples with absorbance ratio  $\geq 1.21$  were interpreted as reactive. **Results:** The commonest presentation involved fever, headache and jaundice. The male–female ratio was 61:46. A total of 65(64.20%) cases had abnormal liver and renal functions respectively, and 57.1% had both the abnormalities. The highest incidence (75, 35.04%) was recorded in September–October followed by July–August (53, 24.77%). The reactive cases had absorbance ratios between 1.21 and 8.21, and 53 showed equivocal result, while IgM non reactivity were seen in 90 patients (absorbance ratios 0.10–0.90). The patients responded to treatment with parenteral antibiotics, penicillin, ceftriaxone and cefotaxime; follow up did not reveal case fatality. **Conclusions:** The cardinal signs of leptospirosis help in making clinical diagnosis, but in any hyper–endemic situation any patient reporting with acute fever and signs of pulmonary, hepatic or renal involvement should be suspected to have leptospirosis and investigated accordingly. Increased awareness, and early diagnosis and treatment, can reduce mortality due to leptospirosis.

## 1. Introduction

Leptospirosis, a septicemic zoonosis of worldwide distribution with a much greater incidence in tropical countries, is caused by the spirochete *Leptospira*. The spectrum of illness is extremely wide, ranging from undifferentiated febrile illness to severe multisystem complications. The disease is maintained in nature by chronic renal infection of carrier animals such as rodents, cattle, sheep, goat, etc., which excrete the organism in their urine, contaminating the environment[1]. Human infection occurs by direct contact with infected urine or tissues or, more commonly by indirect exposure to the organisms in damp soil or water[2].

In Kolkata, a metropolitan city of the West Bengal state (India), there is a high average rainfall with water holding

capacity of soil; here uncontrolled increase in rodent and stray dog population serve as carriers for the disease, and the sanitary conditions of roadside slaughtering stations are poor. The natural water reservoirs in and around the city are frequently used for bathing, swimming and watering of both man and animals. All these factors predispose this area for the persistence of leptospires outside and inside the host, and thus, humans are being affected every year producing a serious health hazard[3]. During the recent times the prevalence of the disease have been reported from various parts of the country[4], but the true incidence of human cases in West Bengal state is not known because of the lack of awareness, or the lack of diagnostic techniques; though only scanty reports are available[3, 5]. Moreover, people of this part of the globe suffering from infectious jaundice are sometimes treated as a case of viral hepatitis, but, many of the cases might be due to *Leptospira* infection. Therefore, the present investigation has been undertaken to study the epidemiology of leptospirosis in and around Kolkata, India, for proper diagnosis of the disease and its treatment as well.

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## 2. Materials and methods

The present study is a retrospective review of records of leptospirosis cases diagnosed at the Calcutta School of Tropical Medicine (CSTM), Kolkata, India during the last 6 years from August 2002 to August 2008. During this period, the Bacteriology and Serology unit of the Microbiology department received 404 blood samples from suspected cases with pyrexia of unknown origin. Those patients attending medical OPD of CSTM and referred from Kolkata based Medical College as well as from B. C. Roy Children Hospital, Kolkata, who fulfilled the criteria for clinical suspicion of leptospirosis, were included in the study.

### 2.1. Case selection

Patients suspected for leptospirosis were evaluated on the basis of their clinical history, epidemiological risk factors, laboratory findings, as per modified Faine's criterion[6], shown in Table 1. A presumptive diagnosis of leptospirosis was made if score of (1) + (2)  $\geq$  26 or (1) + (2) + (3)  $\geq$  25; a score between 20 and 25 suggested the possibility of the disease. A case was defined as a person with fever, headache, and myalgia and more than two of the following symptoms: (a) renal failure due to increase in the blood urea nitrogen (BUN) or serum creatinine (>5 mg/dL) or both above normal ranges, or oliguria or anuria, and hypokalaemia, serum sodium >150 mEq/L, serum potassium >5.5 mEq/L, and platelet count >100 000/mm<sup>3</sup>; (b) hepatic dysfunction due to jaundice with serum bilirubin (>1.5 mg/dL) and elevated hepatic enzymes, hepatic encephalopathy;(c) hemorrhagic manifestations: petechiae/purpura, epistaxis, hematemesis, gastrointestinal bleeding, hematuria, intracranial bleeding, bleeding gums, subconjunctival hemorrhage; (d) neurological: meningeal irritation and convulsion; (e) respiratory: breathlessness, cough, hemoptysis, abnormal chest x-ray, pneumonia or effusion[2].

### 2.2. Serological test

The paired acute and early convalescent (10 - 15 days into illness) serum samples were tested for the specific leptospiral antibodies using IgM ELISA (Diagnostic Automation, Inc. California, USA). Serum was separated and stored at -20 °C until tested. Briefly, for ELISA, serum samples were diluted 1:64 in dilution buffer (Tween 20, BSA, PBS, final pH 7.2  $\pm$  0.2) and incubated for 10 min in antigen (*Leptospira biflexa* Patoc I, serovar patoc I)-coated microwells. After washing three times with wash buffer (PBS and Tween 20, final pH 7.2 $\pm$ 0.2), 2 drops of peroxidase-conjugated goat anti-human IgM were added for 10 min. Subsequently, the wells were washed three times, 2 drops of tetramethylbenzidine were added for 5 min, and the reaction was stopped with 2 drops of 1.0 M H<sub>3</sub>PO<sub>4</sub>. The absorbance was measured at 450 nm. The reactivity of serum samples were interpreted on the basis of absorbance ratio (index value) and IgM levels were recorded from the given absorbance values. The absorbance ratio was calculated as test absorbance/cutoff absorbance, where the cutoff absorbance was correction factor times the mean absorbance of the calibrator. A sample with an absorbance ratio  $\geq$  1.21 was

interpreted as reactive, between 0.91 and 1.20 as equivocal, and  $\leq$  0.90 as non-reactive. For samples showing equivocal results, another blood sample was drawn after a period of 10 days, the test was repeated, and was accounted for in the final result. The method had a sensitivity of 100% and specificity of 90%.

**Table 1**

Modified Faine's criteria for the diagnosis of leptospirosis.

Factors	Score
(1). Epidemiological	
a. Rain fall	5
b. Contact with contaminated environment	4
c. Contact with animal	1
(2). Clinical	
a. Jaundice	1
b. Headache	2
c. Fever	2
d. If fever, temperature 39 °C or more	2
e. Muscle pain (especially calf muscle)	4
f. Meningism	4
g. Conjunctival suffusion (bilateral)	4
h. Conjunctival suffusion+Meningism+Muscle pain	10
i. Albuminuria or nitrogen retention	2
(3). Bacteriological and laboratory findings	
a. Isolation of <i>Leptospira</i> on culture	Diagnosis certain
b. ELISA positive/SAT positive/ MAT single high titre	15
c. MAT rising titre (paired sera)	25

## 3. Results

Among 404 blood samples collected from 404 clinically suspected patients, 214 (52.97%) were found positive for leptospirosis, among which, the male-female ratio was 61:46, indicating males were more prone (122, 57.00%) than females (92, 42.99%) to the disease; there was no clustering of cases in families. Most of the patients by occupation were laborers (87, 40.65%), followed by students (70, 32.71%), housewives (39, 18.22%), indoor non-manual workers (10, 4.67%), farmers (8, 3.73%).

The commonest features include fever (214,100%), headache (214,100%) and jaundice (201, 93.92%). The hepatosplenomegaly were observed with tender and palpable hepatic and splenic enlargement of 1-4 cm and 3-13 cm, respectively in 186 patients (86.91%). Other clinical features were malaise, fatigue, cold, cough, headache, bodyache, distended abdomen, uremia, vomiting, arthritis, pedal edema, diarrhea; bleeding manifestation including petechial hemorrhage, hematuria, hematemesis, and epitaxis. Among the leptospiral positive cases, other infections were also found, such as HIV ( $n=1$ ), HEV ( $n=1$ ), meningococcus ( $n=2$ ), herpes labialis ( $n=1$ ) and septicaemia ( $n=1$ ), conjunctivitis ( $n=1$ ). Mixed infections noticed include malarial parasites (*Plasmodium vivax*, *Plasmodium falciparum*) and dengue virus in 3 cases; only one had complication of cervical lymphadenopathy. Neither the reactive nor the nonreactive leptospiral cases were positive for widal test. X-ray chest taken in 2 patients indicated tuberculosis. Deep vein

thrombosis with arthritis, and thrombocytopenia were seen, each in one case. The 65 % ( $n=139$ ) and 64% ( $n=137$ ) cases had abnormal liver and renal functions respectively, and 57% ( $n=122$ ) had both the abnormalities. No patient suffered from simple pulmonary form of leptospirosis. An overlap between hepato–renal and pulmonary involvement were observed in 5 patients. Central nervous system– involvement in combination with hepato–renal involvement was found in only 3 patients; a single patient suffered from coma.

Serum bilirubin levels among those with abnormal liver function ranged between 1.89 and 42.7 mg/dL (mean 14.61 mg/dL). Fifty four (25.23%) leptospiral cases had bilirubin levels below 1 mg/dL within a range of 0.5–0.9 mg/dL; the SGOT and SGPT values for them ranged 99–410 IU/L and 57–887 IU/L, respectively with a mean of 158.09 IU/L and 198.23 IU/L, respectively. Of the 214 positive cases, 17 (7.94%) and 41 (19.15%) had normal SGOT and SGPT values of 20–30 IU/L and 30–53 IU/L, respectively. The 3 patients had severe renal failure with serum creatinine > 5 mg/dL. Serum creatinine values ranged 0.5 to 6.8 mg/dL (mean 3.78 mg/dL) for the 214 cases, while 42.05 % patients had levels between 0.6 and 1.2 mg/dL. Blood urea levels in patients with abnormal renal function ranged between 53 and 202 mg/dL (mean 114.2 mg/dL), while 54.5% cases indicated serum urea level below the normal range, 10–50 mg/dL. Bleeding manifestations like petechiae, hematuria, hematemesis and epistaxis were presented in 10.5%, 5.14%, 4.7% and 1.2% cases, respectively. Analysis of platelet counts revealed the range of 15 000–28 000/mm<sup>3</sup> (mean 21 000/mm<sup>3</sup>) among the cases studied, the normal range being 1.5–4 lacs/mm<sup>3</sup> (average 2.59 lacs/mm<sup>3</sup>).

During the six years of current study period, 214 cases of leptospirosis were detected by IgM ELISA. In all, maximum number of samples ( $n=164$ ) were collected in 2007, out of which 99 (60.36 %) were positive for leptospirosis (Figure 1). The commonest age group among the positive cases was 5–10 year for both male (31, 14.48%) and female (28, 13.08%). The highest incidence (75, 35.04%) has been recorded in September–October followed by July–August (53, 24.77%). The incidence rate was 14%–15% in May–June and November–December, of the 214 cases; nine cases were obtained in the months of January and February. Out of 214 cases, 126 were from Kolkata and the remaining 88 were from rural suburban areas of ten various adjoining districts of the West Bengal state; three cases, one each from 3 different districts Burdwan, Dinajpur and Murshidabad were recorded.

The reactivity of antileptospiral antibodies in the serum samples of total study cases were interpreted from the absorbance ratios. The absorbance ratios for the reactive cases were between 1.21 and 8.21, with maximum number ( $n=39$ ) of patients having the values 2.21–3.21. The equivocal result was found among 53 patients with absorbance ratios 0.91–1.2, while IgM non reactivity was seen among 90 patients (absorbance ratios 0.10–0.90), of which maximum ( $n=27$ ) number of patients having absorbance ratios 0.60–0.70. The IgM levels in the serum samples in response to the antigen *Leptospira biflexa* Patoc I (serovar patoc I) were interpreted from the absorbance values and expressed in units/ml (U/mL), as shown in Figure 2; the figure showed a

plateau ( $n = 121$ ) in the range 0.5–1.5 U/ml and a minimum value ( $n = 4$ ) in the range 2.0–2.5 U/mL IgM values.

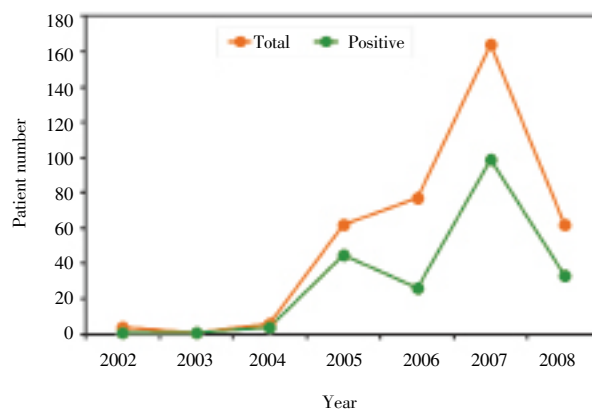


Figure 1. Year-wise incidence of leptospirosis cases, 2002–2008.

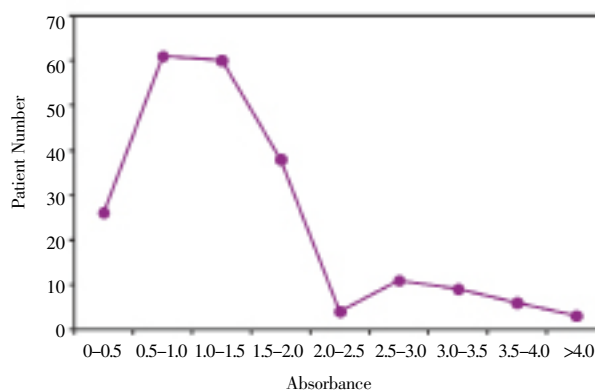


Figure 2. Number of leptospirosis patients showing IgM values.

#### 4. Discussion

The present study has been undertaken as a pilot study and it places on record the serological evidence of human leptospirosis in and around Kolkata by using IgM ELISA.

Several earlier authors reported infection and outbreaks of leptospirosis from different parts of the country[7]. Sethi *et al*[4] in a retrospective study (2004 – 2008) in north India reported an increased incidence of leptospirosis from 11.7% in 2004 to 20.5% in 2008. A total of 215 leptospira cases have been reported during 2007–2009 from Coimbatore[8], while 6 (75%) cases were reported from the same place during 2007–2008[9]. Ninety two of 130 patients suffered from leptospirosis in 2005 in Trivandrum, Kerela, with a case fatality of 15%[2]. The overall incidence of the disease from the same state (Kerela) has been reported high during 1999–2009 with mortality rate 4.01%–8.48%[10]. In Orissa, in 2002, an attack rate of 5.95% with the case fatality ratio of 7.69% has been recorded[11]. Between the years 2001 and 2009, 70%–80% leptospira infection were recorded in Gujrat[12]. Andaman Islands, one of the most endemic areas of the world, has a documented incidence rate of 50/lac[13]. The disease has also been reported from many other parts of the world. Among 263 subjects, 69 positive cases were reported from the Western Pacific Islands (2003–2005)[14]. The incidence of leptospirosis in Sri Lanka has been estimated to be 5.4/lac, the highest rates being recorded from the south and north–central provinces, particularly in Pollonaruwa (24.7/

lac)<sup>[13]</sup>. In Loei Province, Thailand, the incidence rate reached 46.8/lac<sup>[13]</sup>. The national incidence was 5.2/lac in Thailand<sup>[15]</sup>, and 0.5/lakh in Indonesia<sup>[16]</sup>, in 2007, as has been reported by the Ministry of Health. From New Zealand, a disease rate of 2.0/lac was reported in 2008<sup>[17]</sup>, whereas in 2009, in Netherland, 24 cases were reported<sup>[18]</sup>. In England and Wales there were 52 laboratory-confirmed infections in 2009<sup>[19]</sup>. Leptospirosis has been implicated in 13.7% cases in Mayotte of Comoros archipelago during 2007–2008<sup>[20]</sup>. In the current study, we detected 214 confirmed leptospirosis cases from 404 serum samples from patients attending the CSTM, Kolkata for treatment during 2002–2008.

According to NICD report, prevalence rate of leptospirosis was high in several states and union territories including West Bengal<sup>[10]</sup>. In this communication, the highest incidence was recorded from Kolkata, in 2007 on the eve of post monsoon period (September–October); the children (age group 5–10 years) were mostly (27.57%) affected with higher number of males in the same age group. In Orissa, the incidence was higher (5.95%) among males in the age group of 6–15 years while the case fatality was higher among females in the same age group<sup>[11]</sup>. In Northern India, the mean age of patients was 32.6 years, and males (57%) outnumbered females (43%) with a peak during the rainy season<sup>[4]</sup>. In Trivandrum, 80% of patients were below 50 years but a higher mortality (27.7%) was seen in patients above 50 years, and 12.2% in patients below 50 years<sup>[2]</sup>. In Coimbatore, the male–female ratio was 53:37 with 63.75% in 21–45 years age group<sup>[9]</sup>. In Thailand, despite a decreasing male: female ratio (from 9:1 in 1995 to 3:1 in 2003), males remain to be more affected than females<sup>[21]</sup>, but different pattern has been reported from Vietnam<sup>[22]</sup>. The factors like regional climate, population density, contact between humans (accidental hosts) and animals (maintenance hosts) contribute to maintaining a high transmission of leptospirosis<sup>[23]</sup>. We found that the peak incidence of the disease was during September–October followed by July–August, the monsoon season, which may explain the high incidence of seropositivity during this period. In Gujarat the majority of cases and deaths were prevalent from June to September 2009; the Surat, Navsari, Valsad and Tapi districts were mostly affected<sup>[12]</sup>. Humans frequently come in contact with contaminated water during and after heavy rainfalls. Most of the patients in our study were labourers, students, housewives, indoor non-manual workers. Transmission is not only occupational as is observed in temperate climates, but is the consequence of a wider environmental contamination<sup>[7]</sup>. In the present study, a maximum number of patients were from Kolkata, a metropolitan city; this epidemiological form includes urban leptospirosis, often seen in overcrowded cities and towns where environmental sanitation and personal hygiene are poor. The patient input from the nearby adjoining districts of Kolkata, such as Howrah, 24 Parganas (south and north), Nadia and Hooghly was more than the other distant districts, and this is perhaps due to inaccessibility of the testing centre (Kolkata) far away from the residing area. An increasing trend of the received suspected cases at CSTM, Kolkata, was observed from 2003 to 2008, it may be either due to unawareness about the newly opened testing centre or due to leptospiral jaundice cases being treated as viral hepatitis. Leptospirosis is common in occupations having frequent exposure to contaminated environment and contacts

with carrier animals such as rats, pigs, cattle, bandicoots, dogs, and cats<sup>[24]</sup>. Debnath *et al*<sup>[3]</sup> found that in Kolkata, *Leptospira interrogans* serovar *Leptospira pomona* was the most prevalent serovar in the porcine population and human patients, indicating pigs as the natural reservoir for this species of leptospira. Besides, the huge rodent population, and the intermingling of rodents with the pigs and close association with humans might be a source of infection for these patients with this serovar<sup>[3]</sup>. The epidemiological risk factors might be the infestation of dwelling with rats, contact with animals, especially cattle, bathing in public places, history of unprotected contact with dirty stagnant water, wet environmental living conditions, lack of protective footwear, working in farm lands. Prevalence rates were the highest in the 5–10 age groups probably due to greater number of patient being referred from B. C. Roy children hospital. Besides rural and urban form of leptospirosis, there are two other forms, namely the recreational leptospirosis exemplified by the outbreak among participants of the multi-sport racing expedition in Malaysia; and post-disaster leptospirosis outbreaks exemplified by Orissa cyclone and the Mumbai flood<sup>[11, 13, 22, 25, 26]</sup>.

It is mainly an epidemiologic and clinical diagnosis during early stage of the disease as the serological and bacteriological finding is usually not available before 5th day of illness. The modified Faine's criterion was applied in our study to diagnose leptospirosis in 404 patients. Sivakumar *et al*<sup>[6]</sup> modified the original Faine's criteria to include factors like rainfall, IgM ELISA, SAT (Slide Agglutination test) without changing any clinical criteria. Sethi *et al*<sup>[4]</sup> used the modified Faine's criterion without MAT.

The common presenting features of leptospirosis in our study included flulike symptoms (fever and bodyache), jaundice, pedal oedema, extreme muscle tenderness, suffusion of conjunctiva and sub-conjunctival haemorrhage, hepatosplenomegaly. The prevalent signs and symptoms in a study by Prabhu *et al*<sup>[9]</sup> were fever and headache (100%), arthralgia (56%), conjunctival suffusion (41%), jaundice (35%), renal failure, splenomegaly, nausea and vomiting (15%) each and hepatomegaly (10%). Leptospirosis related acute renal failure (ARF) is commonly characterized by polyuria and hypokalaemia, and also the severe oliguric form. This may be related to tubular dysfunction, decreased proximal sodium absorption leading to increased distal potassium secretion and vasopressin resistance causing polyuria<sup>[8]</sup>. In a study by Unnikrishnan *et al*<sup>[2]</sup>, ARF was present in 73% patients, and 67% of renal failure was oliguric. In the present study, 3 patients had severe renal failure with serum creatinine > 5 mg/dL and the mean value of blood urea level was 114.2 mg/dL in overall patients with abnormal renal function; about 94% of leptospirosis was icteric compared with 6% of anicteric forms. The mean value of serum bilirubin levels among those with abnormal liver function was 14.61 mg/dL. In a study by Prabhu *et al*<sup>[8]</sup>, jaundice was observed in 44% patients most of whom also had renal failure as well as severe bleeding episodes in the form of adult respiratory distress syndrome. Our study highlighted that leptospirosis induced impaired renal function is generally accompanied by hyperbilirubinaemia and jaundice with elevated transaminases (mean SGOT and SGPT being 158.09 IU/L and 198.23 IU/L respectively). Sethi *et al*<sup>[4]</sup> described cases with high ALT and AST levels (>200 IU/m L, 2 cases) and very high bilirubin

levels ( $>8$  mg/d L, 2 cases). There have also been reports on pulmonary involvement in leptospirosis<sup>[27, 28]</sup>. The incidence of pulmonary manifestations of leptospirosis ranged from 20% to 70%, along with cough, dyspnoea, chest pain, haemoptysis or respiratory failure<sup>[29]</sup>. Haemorrhagic manifestations of leptospirosis in our study were evident in 21.49% patients through conditions such as hematuria, hemoptysis, hematemesis, epistaxis and petechiae. Thrombocytopenia is especially common, while minority of patients also present with prolonged prothrombin time due to hypoprothrombinemia<sup>[30]</sup>. The haemorrhagic manifestations in leptospirosis constituted the major clinical features in thrombocytopenic cases as highlighted by some investigations that thrombocytopenia as the significant predictor of the development of acute renal failure<sup>[31]</sup>. Neurological manifestations were seen in 10%–15% of leptospirosis patients with signs of altered sensorium and neck stiffness<sup>[32]</sup>.

In the present study we used IgM ELISA of Automation Diagnostic, USA (sensitivity 100% and specificity 90%) to screen for leptospirosis in well timed acute and convalescent blood samples against *Leptospira biflexa* Patoc I (serovar patoc I) strain. Samples interpreted as non-reactive (absorbance ratio  $\leq 0.90$ ) indicated absence of antibody in the sample during the early phase of the disease, i.e., 5–8 days of incubation. In such cases the test was repeated 2–3 weeks later, during which the patients showing equivocal reactions (absorbance ratio 0.91–1.20) were further tested 10–14 days later. A convalescent serum with a significant reaction (absorbance ratio  $\geq 1.21$ ) indicated the formation of specific antibody against leptospira. An initially negative result followed by a positive result implied seroconversion; in our study no such phenomenon was documented. In samples showing initially equivocal results, confirmation was done using a sample collected 2–3 weeks later with paired acute and convalescent sera and absorbance ratio  $\geq 1.21$  in the second sample confirmed the presence of recent specific antibody according to the manufacturer protocol. In our study, out of 53 equivocal samples, the repeat test showed positive reaction for 10 cases and 43 nonreactive cases. A cross-reactive antibody was interpreted if the convalescent serum sample did not show a higher antibody level than the acute sample; no such cross-reaction was reported in our study. The present antigen Biflexa Patoc I is known to cross react with most serovars, excepting animal strains. A sample with an absorbance ratio  $\geq 1.21$  was interpreted as reactive indicative of the presence of specific antibody. Sensitivity of PanBio IgM ELISA has been reported to be 76–90%<sup>[33,34]</sup>. Chaudhry *et al*<sup>[35]</sup> used Serion Immunodiagnostica ELISA with sensitivity and specificity of 96% and 97% respectively. IgM ELISA showed a sensitivity of 97.3% and specificity of 100% compared to MAT<sup>[9]</sup>.

It is essential to include leptospirosis in the differential diagnosis and then institute early empirical therapy to reduce inadvertent deaths. In our study, treatment and chemoprophylaxis of leptospirosis were started as early as possible. All clinically suspected cases of leptospirosis after demonstration of serum reactivity through IgM ELISA were subjected to chemotherapy. The patients responded to treatment with parenteral antibiotics, penicillin, ceftriaxone and cefotaxime. In non-reactive cases, doxycycline was given in a dose of 100 mg twice a day for 7 days. In reactive cases, patients were treated by crystalline

penicillin injection of 20 lac units intravenously every 6 hour for 7 days. Patients receiving penicillin were not given doxycycline capsule. Follow up did not reveal any case fatality. Levett and Haake<sup>[36]</sup> recommended the following dosage for chemoprophylaxis and treatment; doxycycline 200 mg PO once weekly for chemoprophylaxis. For treatment of mild leptospirosis, doxycycline 100 mg bid PO, ampicillin 500–750 mg q6h PO and amoxicillin 500 mg q 6h PO dosages were suggested. For the treatment of moderate to severe leptospirosis, penicillin G 1.5 MU IV q6h, ceftriaxone 1 g IV q 24h, ampicillin 0.5–1 gm IV q6h was indicated. Prabhu *et al*<sup>[8]</sup> treated the milder form of leptospirosis with doxycycline of 50 mg/kg and oral amoxicillin of 40 mg/kg; the severe leptospiral cases were treated with crystalline penicillin. In Gujrat treatment was provided with doxycycline 100 mg twice a day for 7 days in all cases of fever due to the possibility of simultaneous leptospiral infection in the endemic areas during monsoon<sup>[12]</sup>. At Trivandrum medical college, Kerala, treatment included adequate hydration, venous pressure monitoring, and appropriate antibiotics<sup>[2]</sup>. Penicillin was the antibiotic started in most patients, 4 patients allergic to penicillin received doxycycline<sup>[2]</sup>. Twelve patients received additional antibiotics (aminoglycoside, quinolone, or macrolide) for coexistent pulmonary or urinary tract infections. Sethi *et al*<sup>[4]</sup> documented once a day ceftriaxone therapy to have equal efficacy with penicillin therapy, and preferred ceftriaxone therapeutic regimen. Seven cases of primary neuroleptospirosis were put on high dose systemic corticosteroids<sup>[4]</sup>. Further study on the role of systemic steroids in this disease is warranted. Out of 80 thrombocytopenia associated leptospirosis patients, 20 received platelet transfusion<sup>[8]</sup>. Levett and Haake<sup>[36]</sup> recommended supportive therapy for hospitalized leptospirosis cases. Patients with early renal disease with high-output renal dysfunction and hypokalemia should receive aggressive volume repletion and potassium supplementation to avoid severe dehydration and acute tubular necrosis. In patients who progress to oliguric renal failure, rapid initiation of hemodialysis reduces mortality and is typically required only on a short-term basis. Patients requiring intubation for SPHS (severe pulmonary hemorrhage syndrome) have decreased pulmonary compliance and should be managed as cases of acute respiratory distress syndrome (ARDS). Protective ventilation strategies involving low tidal volumes ( $< 6$  mL/kg) to avoid alveolar injury caused by high ventilation pressures have been shown to improve survival rates in ARDS dramatically<sup>[36]</sup>.

In our part of the globe, occurrence of leptospirosis is not uncommon because the risk factors already prevailing here predispose this area for the persistence of leptospires outside and inside the host, and thus, humans are being affected every year producing a serious health hazard<sup>[3]</sup>. However, considering the number of samples studied, the current scenario did not represent the true reflection of its prevalence in the community because not all suspects undergo diagnosis and thus treatment for leptospirosis. Nevertheless, the study underlines the importance of serological testing of suspected cases from in and around Kolkata, India, for the early diagnosis and identification of the cases with this potentially dangerous disease, and to institute treatment before complications set in. The organism responds readily to antibiotics like penicillin, doxycycline and third generation cephalosporins (ceftriaxone and



cefotaxime). However, in complicated cases life supporting measures like dialysis, hydrocortisone and inotropic drugs may be included in the treatment schedule.

### Conflict of interest statement

We declare that we have no conflict of interest.

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