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To Study Prevalence of Brucellosis, Leptospirosis & Scrub Typhus in Cases of Acute Febrile Illness in A Tertiary Care Hospital

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Abstract

Introduction: Acute febrile illness is the most prevalent symptom among individuals seeking medical attention in tropical countries like India. Healthcare professionals have a diagnostic and therapeutic challenge, particularly in places with limited resources where the incidence of AFIs is high.

Aim: To estimate the prevalence of Brucellosis, Leptospirosis & Scrub typhus in cases of acute febrile illness and to assess the sociodemographic and clinical profile of these cases.

Materials and methods: The study population constitutes patients with an acute undiagnosed febrile illness presenting to the Outpatient and Inpatient Department of Medicine of Government Medical College, Amritsar during the study period from July 1,2023 to April 30, 2025. Serum sample from suspected cases was tested for brucellosis, leptospirosis and scrub typhus by IgM ELISA in the Department of Virology.

Results: Overall seroprevalence of leptospirosis, scrub typhus and brucellosis cases among acute febrile illness cases was found to be 19.28% (54/280), 6.07% (17/280) and 6.42%(18/280) respectively. Co infection was also

observed among 5 patients for both leptospirosis and scrub typhus.

Conclusion: The current investigation indicates a significant seroprevalence of leptospirosis, scrub typhus, and brucellosis among undifferentiated AFI cases in patients presenting in GNDH, Amritsar, despite the fact that these diseases were underreported in this area. It is therefore strongly advised that a programmatic approach for the management, prevention, and control of these diseases should be initiated.

Keywords: Acute Febrile Illness, Fever, Zoonotic

Introduction

In tropical nations like India, acute febrile illness is a common complaint among people seeking medical care. Healthcare providers face diagnostic and treatment challenges, especially in resource-poor areas where AFIs are prevalent.

It encompasses a wide range of infectious and non-infectious causes, including autoimmune, inflammatory, parasitic, viral, and bacterial illnesses. Based solely on clinical appearance, it can be challenging to differentiate between these various disorders.[1]

In tropical regions like India, the majority of AFI cases occur during the monsoon season and in between seasonal variations. The environment is favourable for the transmission of disease because of factors including humidity, rising temperatures, prolonged rains, standing water, floods, an abundance of vegetation growth, vector density, and biting patterns. Potentially dangerous illnesses like dengue, enteric fever, chikungunya, malaria, leptospirosis, and rickettsial diseases are common in developing countries. In developed countries, however, viral etiologies are the main cause. [2]

Leptospirosis is a potentially fatal zoonotic illness that can cause widespread epidemics after times of flooding and heavy precipitation. Leptospirosis occur after contact with infected reservoir host animals that contain the pathogen in their renal tubules and excrete pathogenic leptospires in their urine, directly or indirectly. Infections in humans are most commonly caused by brown rats (Rattus norvegicus). [3] There are many different natural rodent and non-rodent reservoir hosts for leptospirosis, such as rabbits, cattle, buffalo, goats, sheep, and pigs

The condition may present with a wide range of clinical symptoms that can range from a mild "flu"-like illness to Weil's syndrome, a severe and sometimes fatal icteric illness. Icterus, or jaundice, is a common symptom of leptospirosis, but it can also be seen in a number of other liver-related conditions, such as various forms of hepatitis. [4]

Leptospirosis is responsible for an estimated 1.03 million infections and 58,900 deaths each year. The largest estimates of disease morbidity and death are found in the Global Burden Disease regions of East Sub-Saharan Africa, South and Southeast Asia, Oceania, the Caribbean, Andean, Central, and Tropical Latin America.[5]

In India, leptospirosis is more frequently found in coastal districts of Andaman and Nicobar (also known as Andaman hemorrhagic fever), Kerala, Gujarat, Maharashtra, and Tamil Nadu. Andhra Pradesh, Karnataka, Dadar and Nagar Haveli, Daman and Diu, Puducherry, Goa, and Odisha are next in line. The most common serovars in India are L. Andamana, L. Pomona, L. Grippotyphosa, L. Hebdomadis, L. Semoranga, L. Javanica, L. Autumnalis, and L. Canicola.[6]

The rickettsial bacterium, Orientia tsutsugamushi is the cause of scrub typhus, a zoonotic disease also referred to as Tsutsugamushi disease. This bacterium is obligatory intracellular and gram negative organism. This disease is contracted by humans through the bite of a chigger, a trombiculid mite larva belonging to the Leptotrombidium

group. Because infected chiggers are most common in areas with dense scrub vegetation during the wet season, when mites lay their eggs, this disease has also been called river/flood fever. It usually takes place between the months of June and November. [7,8]

Worldwide, it poses a hazard to one billion individuals and infects one million people annually .[9]In the Asia-Pacific area, this illness is a major public health concern. It affects more than one billion people and covers an area of more than eight million square kilometers. The "tsutsugamushi triangle" is the endemic area for scrub typhus. In addition to Japan, Taiwan, China, South Korea, Nepal, Northern Pakistan, and Papua New Guinea, this also covers the Australian states of Queensland and Northern New South Wales. For many years, scrub typhus has been known to exist in India. The following states have reported cases of the disease: Haryana, Jammu and Kashmir, Himachal Pradesh, Uttaranchal, West Bengal, Assam, Maharashtra, Kerala, and Tamil Nadu.[10,11]

Brucellosis is a primarily a zoonotic disease affecting various domestic animals such as sheep, goat or cattle. Three species of Brucella are thought to be significant human disease agents: B. melitensis, B. abortus and B.suis .Human infection is usually associated with occupational exposure to infected animals or their products. Brucella was named after British army physician Sir David Bruce (1886), who isolated the first recognized species, Brucella melitensis. [12]Human brucellosis is endemic in areas where animals are raised in large numbers, such as countries of Mediterranean zone, Eastern Europe, Central Asia, Mexico and South America.[13] Weight loss, undulating fever, nocturnal sweats are primary signs of brucellosis. It primarily infects organs of the reticuloendothelial system, such as lymph nodes, spleen, liver and bone marrow. Other organs such as placenta, musculoskeletal tissues and genitourinary systems are also involved. [14]

Materials and Methods

Our study was hospital based cross-sectional study conducted from July 1, 2023 to April 30, 2025(22 months) at Government Medical College, Amritsar. The study population included patients presenting with an acute undiagnosed febrile illness attending outpatient and inpatient units of Medicine department.

Inclusion Criteria

- Patients of any age group with an acute undiagnosed febrile illness (body temperature >38.2 ºC)
- Suspected case of leptospirosis, scrub typhus and brucellosis (according to clinical case definition).
- Patients giving consent for participation in the study.

Exclusion Criteria:

- Cases of acute febrile illness who do not fit in to the clinical case definition of leptospirosis, scrub typhus & brucellosis.
- Patients who refuse to consent in research participation.

Case Definition

- **Clinical Case Definition : Leptospirosis**

Acute febrile illness with headache, myalgia and prostration associated with any of the following symptoms: conjunctival suffusion, meningeal irritation, anuria or oliguria and/or proteinuria, jaundice, hemorrhages (from the intestines; lung bleeding is notorious in some areas), cardiac arrhythmia or failure, skin rash.[15]

- **Clinical Case Definition :Scrub typhus**

A clinical case of scrub typhus is one with acute undifferentiated febrile illness of 5 days or more with or without eschar should be suspected as a case of Rickettsial infection. If eschar is present, fever of less than 5 days should be considered as Scrub typhus. Other

signs and symptoms may be headache, rash, lymphadenopathy, multi-organ involvement like liver, lung and kidney involvement. [16]

- **Clinical Case Definition :Brucellosis**

An illness characterized by acute or insidious onset of fever and one or more of the following: night sweats, arthalgia, fatigue, anorexia, myalgia, weight loss, meningitis or focal organ involvement (endocarditis, orchitis, hepatospenomegaly, splenomegaly).[17]

Sample Collection and Processing

5 ml of whole blood sample was drawn from the suspected cases using a plain vacutainer while adhering to very rigorous aseptic guidelines. Serum was separated and stored in the refrigerator till processing was done. IgM ELISA was performed on the samples to detect IgM antibodies specific for brucellosis, leptospirosis, and scrub typhus respectively in the Department of Virology. ELISA testing was performed as per following kit manufacturer's instructions.

- RecombiLISA (Leptospira IgM)
- MICROLISA (Scrub Typhus IgM ELISA)
- Calbiotech (Brucella IgM ELISA)

Software called SPSS 20 (IBM Corporation, Armonk, NY, USA) was used to analyze the data. Percentages were used to represent categorical data. The Chi-Square test for non-parametric data, the Posthoc Anova test, the Student's "t" test, and Pearson's correlation test were used to compare the numerical variables, which were normally distributed. The degree of significance was then assessed using the p-value. To make pertinent inferences, the findings were examined and contrasted with those of earlier research.

Results

Out of total 280 cases, 54 (19.28%) cases were positive for leptospirosis. Positive cases for scrub typhus and brucellosis were 17(6.07%) and 18 (6.42%) respectively (Fig.1). Coinfection was observed in 5 cases for leptospirosis and scrub typhus. The demographic distribution of the positive cases is as follows (Table 1). Seroprevalence of leptospirosis among male and female AFI cases was 16.36% and 23.47% respectively. Maximum seroprevalence was seen in 0-20 years (29.03%) followed by >60 years age group (26.53%), 21- 40 years group (21.23%) and 41-60 years (9. 19%).Seroprevalence was observed more in the rural areas (20.8%)than in urban in leptospirosis. The rate was highest during the monsoon (25.96%) followed by summer (21.95%), autumn (17.54%) and winter (10. 25%).The predominant occupation affected was the agricultural group (28.26%).

In Scrub typhus, seroprevalence of scrub typhus among male and female AFI cases was 6.66% and 5.21% respectively. Maximum seroprevalence was seen in 0-20 years age group (9.67%), followed by 41-60 years group (8. 04%).More seroprevalence was found in the rural areas (6.71%). seroprevalence rate was highest during the summer (17.07%), followed by autumn (5.26%), monsoon (4.80%) and winter (2. 56%).The predominant population affected belonged to agricultural group (8.7%).

In Brucellosis, seroprevalence of brucellosis among male and female AFI cases was 8.48% and 3.47% respectively. Maximum seroprevalence was seen in >60 years age group (10.20%), followed by 0-20 years group (9.67%), 21-40 years (7.07%) and 41-60 years (2. 29%).Seroprevalence was found more in the urban areas (9.16%). The rate was highest during the winter (12.82%) followed by summer (4.87%), monsoon (3.84%) and autumn (3.50%). (Fig 2) The predominant population affected belonged to service / student group (11.11%) (Table 1).

Figure 1: Seroprevalence of leptospirosis, scrub typhus and brucellosis (total cases=280)

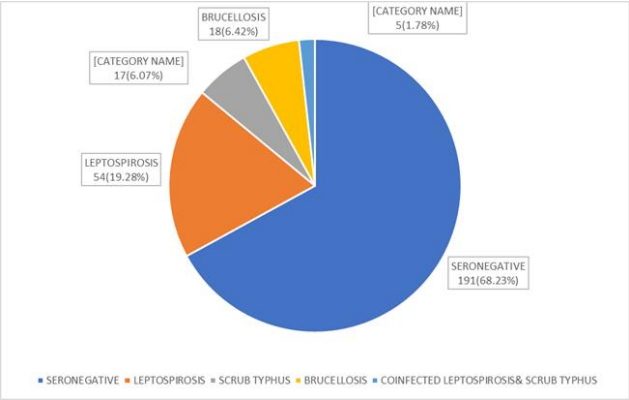

Table 1: Seroprevalence & Demographic Distribution of The Positive Cases

Parameter	Leptospirosis(n=54)			Scrub Typhus(n=17)			Brucellosis (n=18)		
	Seroprevalence	Chi Square	P Value	Seroprevalence	Chi Square	P Value	Seroprevalence	Chi Square	P Value
Gender									
Male	16.36%	1.476	0.224	6.67%	0.249	0.617	8.48%	2.823	0.243
Female	23.47%			5.21%			3.47%		
Age Group(Years)									
0-20	29.03%	6.627	0.084	9.67%	2.509	0.473	9.67%	3.775	0.286
21-40	21.23%			5.30%			7.07%		
41-60	9.19%			8.04%			2.29%		
>60	26.53%			2.04%			10.20%		
Residence Area									
Rural	20.8%	0.472	0.491	6.71%	0.228	0.632	4.02%	3.053	0.217
Urban	17.55%			5.34%			9.16%		
Seasonal Variation									
Summer(April-June)	21.95%	5.098	0.164	17.07%	8.841	0.031	4.87%	6.309	0.097
Monsoon(July - September)	25.96%			4.80%			3.84%		
Autumn(October – November)	17.54%			5.26%			3.50%		
Winter(December-March)	10.25%			2.56%			12.82%		
Occupation									
Agriculture	28.26	0.28	0.9908	8.7	0.49	0.9748	10.87	2.74	0.6019
Labourer/Construction Worker	23.53			5.88			5.88		
Service/Student /Self Employed	24.69			8.64			11.11		
Retired/ Housewife	25			8.33			4.17		
Others	23.81			4.76			4.76		

Figure 2: Seasonal Distribution of the Positive Cases

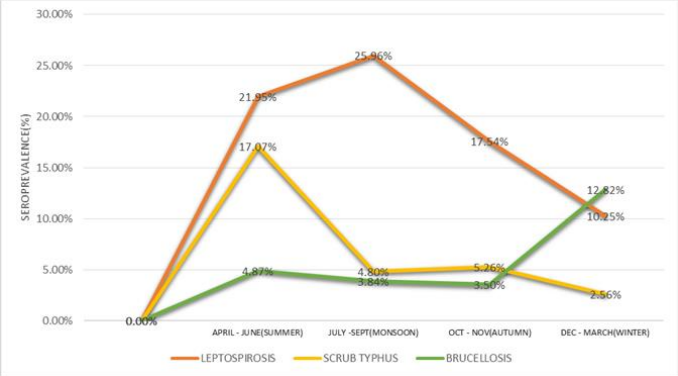

Figure 3: Comparison of Clinical Features of Leptospirosis, Scrub Typhus And Brucellosis Cases

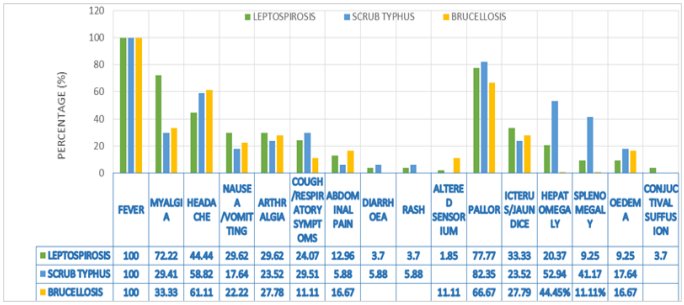

Table 2: Comparison of Lab Parameters of The Positive Cases

Laboratory Parameters	Leptospirosis (%) N=54	Scrub Typhus (%) N=17	Brucellosis (%) N=18
• Hematological Investigations			
Hemoglobin (<10g/dL)	77.78	82.35	66.67
TLC(>11000/µl)	53.70	94.11	22.22
Platelets(<100000/µl)	20.37	58.82	83.33
• Biochemical Investigations			
S. Creatinine (>1.5 mg/dl)	37.03	11.76	16.67
Blood urea (>40 mg/dl)	38.89	11.76	33.33
S. Bilirubin (Total) (> 2 mg/dL)	51.85	23.52	27.79
SGOT (>80 IU/ml)	46.29	17.64	22.22
SGPT (>80 IU/ml)	44.44%	17.64	27.79

Discussion

Brucellosis, leptospirosis and scrub typhus have returned to prominence as the primary causes of AFI in many regions of India in recent years, particularly during the monsoon and post-monsoon seasons. The public health burden and worldwide spread of these diseases are still poorly understood, despite growing awareness in endemic areas. Studies on the seroprevalence of these illnesses in Punjab, particularly in the Amritsar district, are scarce.

Clinically suspected AFI cases(N=280) of leptospirosis, scrub typhus and brucellosis presenting in outpatient and inpatient Department of Medicine, GNDH complex Amritsar were tested using IgM ELISA. Out of the total cases of AFI (N=280), overall seroprevalence of leptospirosis, scrub typhus and brucellosis cases was found to be 19.28% (54/280), 6.07% (17/280) and 6.42% (18/280) respectively. Co infection was observed among 5 patients in leptospirosis and scrub typhus. Seroprevalence of leptospirosis (23.47%) was higher in females than males (16.36%) but seroprevalence of scrub typhus among males (6.67%) was higher than females (5.21%). In a study by Ramlingam et al., males were found to have higher seroprevalence (57.1%) than females (42.9%) in both leptospirosis and scrub typhus [18]. Gender distribution however in leptospirosis and scrub typhus was not found to be statistically significant (p =0 .137) for leptospirosis and (p= 0.617) for scrub typhus respectively. Seroprevalence of brucellosis among males was (8.48%) and females (3. 47%).Gender distribution in

brucellosis was not found to be statistically significant (p=0.241). Maximum seroprevalence of cases of leptospirosis and scrub typhus were seen in 0-20 years age group. Ramlingam et al, reported more seroprevalence among 21-40 years age group followed by 0-20 years age group for both leptospirosis and scrub typhus cases[18].Whereas in brucellosis more seroprevalence was seen among > 60 years of age group. In a study by Modak et al at West Bengal, majority of the patients affected with brucellosis belonged to the age group 51–60 years (23.5%)[19].

Leptospirosis was most common during monsoon season (25.96%). The monsoon season creates a perfect storm for leptospirosis due to more contaminated water and soil due to animal urine and flooding, combined with increased human contact with these contaminated environments. In a study conducted at Wardha district, Deshmukh et al observed the highest incidence of leptospirosis (18%) in the monsoon season. [20]Scrub typhus was most common in the summer season (17.07%). Increased mite populations brought on by higher temperatures and humidity can enhance the danger of spreading scrub typhus. Seroprevalence of brucellosis was maximum during the winter season (12. 82%).It could be due to longer environmental survival of the bacteria in cold and moist environments. Direct exposure to sunlight and warm weather significantly reduces their survival. (Fig.1).

Majority of patients were from rural areas both in leptospirosis and scrub typhus accounting for 20.8% and 6.71% respectively. Whereas in brucellosis more cases (9.16 %) belonged to urban area. Sethi et al reported higher seropositivity in rural area (76.7%) than urban areas (23.3%) in his study among leptospirosis cases .[21] A study conducted by Devamani et al in hilly areas in South India reported rural preponderance (28.1%) in scrub typhus. [22]. Among brucellosis cases, Parai et al reported higher seroprevalence from rural areas than urban dwellings which is contradictory to the current investigation[23].The predominant occupation affected were agriculture in leptospirosis and scrub typhus. (Table 1). Brucellosis is traditionally considered an occupational hazard for those directly involved with livestock. In the current study, the predominant occupation affected were servicemen / students in brucellosis. This suggests several possibilities such as frequent consumption of raw milk, unpasteurized dairy products, undercooked meat, people in service roles in healthcare (nurses, doctors, lab technicians) could be at risk. Students living in rural or semi urban areas might be exposed to airborne bacteria from nearby animal enclosures, even if they don't directly handle animals.

The most common presentation observed in leptospirosis was fever followed by myalgia, headache, arthralgia, cough, abdominal pain and diarrhroea. In scrub typhus cases, the most common presentation after fever was myalgia followed by cough, arthralgia, breathlessness and abdominal pain. Brucellosis cases mostly presented with fever followed by myalgia, breathlessness, chills, arthralgia, hemoptysis and altered sensorium. (Fig .2) The most common altered lab parameter was anaemia and leucocytosis in both leptospirosis and scrub typhus followed by hyperbilirubinemia. Thrombocytopenia and raised blood urea were the predominant altered parameters in brucellosis. (Table 2).

5 (1.78%) samples were found to be positive for both leptospirosis and scrub typhus, 49 (17.5%) for leptospirosis only and 12 (4.28%) for scrub typhus only (Fig 1). In a study conducted by Borkakoty, et al. in Longding district of Arunachal Pradesh in 2013, scrub typhus IgM was reactive in 97% (30/31), and 25% (8/31) cases were co-infected with leptospira.[24] Similarly a

study conducted at Chennai by kanagasabai et, al., coinfection was seen with scrub typhus and leptospira in 23 patients out of 354 serum samples. Coinfection can be attributed to several overlapping factors such as common epidemiology, shared risk factors, exposure and environmental conditions.[25]

Conclusion

In developing tropical countries, acute febrile diseases (AFIs) like dengue, chikungunya, leptospirosis, scrub typhus, and malaria result in significant morbidity, mortality, and economic impact. Since the clinical characteristics of many illnesses are so similar and non-specific, it is impossible to identify the underlying cause. Brucellosis, leptospirosis, scrub typhus and have taken over as the main causes of AFI in many regions of India. As per literature, very few studies have been documented on seroprevalence of brucellosis, leptospirosis and scrub typhus, particularly in the North-West (Majha) region of Punjab. The current investigation indicates a significant seroprevalence of leptospirosis, scrub typhus, and brucellosis among undifferentiated AFI cases in patients presenting in GNDH, Amritsar. It is therefore strongly advised that a programmatic approach for the management, prevention, and control of these diseases should be initiated.

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