VOLUME-8, ISSUE-8, AUGUST-2019 • PRINT ISSN No. 2277 - 8160

Original Research Paper

Medical Science

SEROLOGICAL STUDY OF BRUCELLOSIS IN SEPHERD AND BUTCHERS SUFFERING FROM PYREXIA OF UNKNOWN ORIGIN (PUO) ATTENDING A TERTIARY LEVEL HOSPITAL

Harender Simar — Assistant Professor, Department of Microbiology, Maharaja Agrasen Medical College, Agroha, Hisar

Suman Rana* — Associate Professor, Department of Pediatrics, Maharaja Agrasen Medical College, Agroha, Hisar *Corresponding Author

Reena Gandhi — Assistant Professor, Department of Pharmacology, Maharaja Agrasen Medical College, Agroha, Hisar

ABSTRACT Brucellosis is a disease of animals as well as human and distributed throughout the world including India. Brucellosis is a zoonotic disease. So person involved with rearing of animals are most commonly affected with brucellosis. Many times patients with brucellosis present with history of prolonged undiagnosed fever labelled as pyrexia of unknown origin (PUO). The objective of this study was to determine the seroprevalence of brucellosis among PUO cases in sephered & butchers admitted at Prathima Institute of Medical Sciences, Karimnagar. Serum samples from PUO cases sera were tested by Rose Bengal Plate Agglutination Test (RBPT), & Standard Tube Agglutination Test (STAT). Brucellosis might be missed if it is not considered in the differential diagnosis of PUO.

KEYWORDS : Brucellosis, Pyrexia of Unknown Origin (PUO), RBPT, STAT

INTRODUCTION

Brucellosis is one of the world's major zoonosis. It is an infectious disease of public health importance and economic concern in many parts of the world.[1] It is caused by a Gram negative bacillus belonging to the genus *Brucella*. Brucellosis in human beings is mainly caused by 4 species: *Brucella* melitensis, *Brucella* abortus, *Brucella*suis, and *Brucella* canis. Livestocks are the main reservoir of brucellosis.[2] Animals that are known to serve as sources to human infection are goats, sheep, cattle, buffaloes, and swine. It is an occupational hazard to dairy personnel, veterinary personnel, laboratory workers, butchers and abattoir workers, animal owners, milk vendors and shepherds.[3] Human beings acquire infection by consumption of raw milk or milk products, meat and contact with infected animals. Person to person transmission is rarely documented either in circumstances implicating sexual contact[4] or by transfer of tissue including blood and bone marrow.[5] World Health Organization reported that half million new human cases are reported annually worldwide and these numbers are greatly underestimated the true incidence of human disease as the actual number of cases is estimated to be at least 10 times the figures officially announced.[6] This is because of misdiagnosis and under reporting.

An outstanding characteristic of the brucellosis disease is its protean manifestation. Common symptoms are fever, muscular and joint pain, chill, sweat, weakness, loss of weight, abdominal pain, respiratory illness, central nervous system infections, heart disease, urogenital infection, scrotal swelling with testicular pain or as the chronic localized lesion.[3]

In this area of Telangana, where a large population is agriculture dependent, frequently come in contact with infected animals (goats, sheep, cattle, and other animals) and their products. And suffer from brucellosis like manifestation clinically. Brucellosis is treatable disease if the proper diagnosis is made early.[3] The objective of the present study was to determine the seroprevalence of brucellosis among PUO cases in sephered and butchers.

MATERIAL AND METHODS

The study was carried out in the Department of Microbiology, Prathima Institute of Medical Sciences, Nagunur, Karimnagar, for a period of one year. Institutional Ethical committee approval was obtained for the study after following the due procedure laid down by the ethical committee. In the present study included 77 cases (belongs to shepherd or butchers profession) of pyrexia of unknown origin (PUO). As per PUO definition by Petersdorf & Beeson (1961) and new definition, PUO is defined as febrile illness with temperature greater than 38.3°C (101.3°F) on several occasions accompanied by more than 3 weeks of illness and failure to reach a diagnosis, or after 3 outpatient visits or 3 days in the hospital without elucidation of a cause or 1 week of "intelligent and invasive" ambulatory investigations.[7] Patient who did not fit in PUO case definition were excluded from the study. 5ml of venous blood was collected aseptically in vacutainer and was allowed to clot at room temperature for half an hour, after which serum was separated by dislodging the clot and centrifuged at 1500 rpm for 5 minutes. Serum was stored frozen in a sterile Eppendorf tube in the freezer compartment of the refrigerator until they were tested. The sera received from patients with pyrexia of unknown origin (PUO) were tested by Rose Bengal Plate Agglutination Test (RBPT), & Standard Tube Agglutination Test (STAT. All relevant data were systematically recorded in predesigned data sheets for analysis.

RESULTS

The study material comprised 77 blood samples of patients suffering from pyrexia of unknown origin attending PIMS, Karimnagar. Out of 77, 42 (54.55%) were shephered & 35 (45.45%) were butchers. All samples were screened for brucellosis by RBPT, STAT test as per standard procedure.

Out of 77 serum samples tested, 9 (11.69%) & 8 (10.39%) found positive by RBPT & STAT test respectively (Table 1).

Table 1: Results of serological tests for Brucellosis in PUO patients

No. of sample screened	RBPT		STAT	
	Pos (%)	Neg (%)	Pos (%)	Neg (%)
77	9 (11.69%)	68 (88.31%)	8 (10.39%)	69 (89.61%)

The study included 51 (66.23 %) males and 26 (33.77 %) females. Out of 51 male patients, 6 (11.76%) were found positive by STAT, whereas only 2 (7.69%) out of 26 females patients revealed positive result.

5 (11.9%) out of the 42 samples from shepherds and 3 (8.57%) out of 35 samples from butchers were found positive for brucellosis.

Out of 77, 65 (84.42%) patients were belonging to rural areas and 12 (15.58%) were from the urban area. The prevalence of brucellosis was higher in person residing in a rural area (11.9%) than in urban areas (8.57%).

The study shows that the majority of cases had antibody titer in the range of 320 - 640 IU/ml. 4 samples were showed titre of 320 IU/ml followed by 640 IU/ml in 3 samples. only 1 sample registered a maximum 2560 IU/ml antibody titer.

Table 2: Rose Bengal Plate Test v/s Standard Tube Agglutination Test

RBPT		STAT		TOTAL
		POSITIVE	NEGATIVE	
	POSITIVE	8 (T.P)	1 (F.P)	9
	NEGATIVE	0 (F.N)	68 (T.N)	68
	TOTAL	8	69	77

T.P: True positive, F.P: False positive, F.N: False negative, T.N: True negative

Diagnostic efficacy of RBPT when compared with "widely used gold standard serological test" "STAT" (Table 2) is as given below

Sensitivity (true positive rate)	- 100%
Specificity (true negative rate)	- 98.55%
Positive predictive value	- 88.89%
Negative predictive value	-100%
Over all accuracy	- 98.55%

DISCUSSION

Brucellosis is an important public health problem it occurs in certain groups of people due to their occupational exposure. Such peoples acquire the disease, suffer usually from joint pains, myalgia, weakness, and prolonged un-diagnosed fever and remain morbid for long periods. These people go un-diagnosed and get symptomatically later on. The non-specific and protean manifestations of the disease add to this problem. Out of 77 PUO cases in our study, 8 (10.39%) cases were found seropositive for *Brucella* antibodies. Prevalence of brucellosis in our study is in concurrence with the Mrunalini et al; from Veterinary Biological and Research Institute Hyderabad, a reference center for brucellosis in South India reported a prevalence of 11.5%. [8]

In the present study, seropositivity is higher in male 11.76% (6 in 51) than in female 7.69% (2 in 26). Our results of male to female ratio (3: 1) almost correlates well with Kadri S M et al; (3:1).[9] Randhawa et al;[10] studies also shows that prevalence is more among males than in females. The increased incidence in males during the present study may be attributed to the fact that the majority of the males are exposed to animals compared to females.

Brucellosis is a disease more often seen in specific occupational groups like veterinarians, shepherds, dairy workers, butchers, farmers, etc. The present study reveals that the highest prevalence of brucellosis is among shepherds 5 (11.9%) followed by butchers 3 (8.57%). Spink et al; have similarly noted these occupational groups to have a higher prevalence. Direct contact (occupational contact with animals in the field, handling animals, engaged in parturition of animals) is found to be significantly most important predisposing risk factor than indirect contact (ingestion of raw milk, raw meat and laboratory personnel) which explains our results. [11] In the present study of PUO cases, 84.42% of cases were from rural areas and 15.58% were from urban areas. The result showed that the prevalence of brucellosis is higher in rural area 10.77% (7 out of 65) than in urban areas 8.33% (1 out of 12). This finding is in agreement with several other studies in which it is concluded that the higher prevalence in rural area people may be because they live in close proximity with animal livestock.

Antibody level among the seropositive patients in our study ranged from 320 IU/ml to 2560 IU/ml. The diagnostic titer suggested by IVRI is 80 IU/ml for human brucellosis. In the present study majority of positive cases showed titer of 320 IU/ml followed by 640 IU /ml. RBPT showed a sensitivity of 100% and specificity of 98.55% when compared with the serological gold standard for brucellosis. Our study well correlates with a study conducted by Amresh N et a;[12] who observed the sensitivity of 99.78% and specificity of 97% for RBPT when compared with STAT.

CONCLUSION

The over all prevalence of brucellosis in the study was found to be 11.69%. The prevalence of brucellosis in relation to sex revealed positivity of 13.11% in males and 6.25% in females. The prevalence of brucellosis in relation to occupation revealed that shepherds had the highest prevalence rate of 11.9% (5 out of 42), 8.57% (3 out of 35 patients). This suggested that brucellosis might be missed if it is not considered in the differential diagnosis of PUO specially if the patient belongs to shepherd or butchers profession. RBPT could be employed as an efficient screening test for brucellosis.

REFERENCES

1. Young EJ. Brucella Species. In: Mandell GL, Bennett JE, Dolin R, editors Mandell, Douglas and Bennet's Principles and Practice of Infectious Diseases, 6thed. Vol II. Philadelphia: Elsevier; 2005;2669-74.
2. Smits HL, Kadri SM. Brucellosis in India:a deceptive infectious disease. Ind J Med Res. 2005; 122: 375- 84.
3. Mantur B G, Biradar M S, Bidri R C, Mulimani M S, Veerappa K, Kariholu P L et al. Protean clinical manifestations and diagnostic challenges of human brucellosis in adults: 16 years' experience in an endemic area. J Med Microbiol. 2006; 55: 897- 903.
4. Mantur B G, Mangalgi S S, Mulimani M. Brucella militensis– a sexually transmissible agent ? Lancet1996Jun;347(9017):1763.
5. Corbel M J, Mac Millan A P. Brucellosis.In: Collier L, Sussman M, Balows A, editors. Topley and Wilson's Microbiology and Microbial infection, 9thed. New York, USA: Arnold publications, 1998;820-23.
6. Pappas G, Papadimitriou P, Akritidis N, Christou L, Tsianos EV. The new global map of human brucellosis. Lancet Infect Dis. 2006; 6: 91–99.
7. Jeffrey AG, Callahan MV. Fever of unknown origin. In : Longo DL, Fauci AS, Kasper DL, Hauser SL, Jameson JL, Loscalzo J. Editors. Harrison's Principles of Internal Medicine. Vol. I, 18th ed. New York, USA: McGraw-Hill Pub; 2012.p.158
8. Mrunalini N, Reddy M.S., Ramasastry P, M.R. Rao. Seroepidemiology of human brucellosis in Andhra Pradesh. Indian Veterinary Journal July 2004; 81(7): 744-47.
9. Kadri SM, Rukhsana A, Laharwal MA, Tanvir M. Seroprevalence of Brucellosis in Kashmir (India) among patients with Pyrexia of Unknown Origin. J Indian Med Assoc. 2000; 98: 170- 71.
10. Randhawa A S, Dhillon S S. Seroprevalence of brucellosis in humans and animals of Punjab. Ind J Pub Health. 1974;18:15-19.
11. Spink W W. The nature of brucellosis. The University of Minnesota Press Minneapolis. 1956;112-144.
12. Amaresh Nigudgi, Praveen Doddamani, Vinay Hazare, Sreekantha, Sunil Biradar, Kantha RC . Serology and antibiotic susceptibility in brucellosis. Int J Pharm BioSci. 2013 Apr; 4(2): (B)200-13.