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Brucellosis in Cattle and Occupationally Exposed Human Beings: A Serosurvey in Odisha, India

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The current serosurvey was carried to illuminate the status of brucellosis in cattle and high risk human beings of Odisha state, India. Serum samples (n= 346) comprising 258 from cattle and 88 from human beings were obtained and processed through RBPT, STAT, and I-ELISA. 87 milk samples were subjected to MRT. Cattle seropositivity by I-ELISA, RBPT and STAT were found to be 8.14%, 4.26% and 2.32% respectively. Milk sample analysis showed 3.44% positivity in MRT. Bull showed higher prevalence than female. In case of human beings the value obtained were 5.68% and 3.41% in RBPT and STAT respectively. The prevalence among veterinary officers, farmers, animal handlers, slaughter house workers and from patients with Pyrexia of Unknown Origin was 0.00, 9.09, 3.70, 12.5, 4.41 and 5% respectively. The current observation provides a baseline prevalence level of brucellosis in cattle and occupationally exposed human beings of Odisha. A broader investigation including small ruminants is required to elucidate the actual prevalence of the disease in the state.

Key words: Seroprevalence, Brucellosis, RBPT, STAT, I-ELISA, MRT.

Diagnosis and determination of prevalence of a disease in a population is important for effective control and eradication of the same. Brucellosis having zoonotic significance is an economically important reproductive disease of livestock that induces infertility, delayed heat, interrupted lactation and decreased production. The disease has been reported throughout the country, although there is wide variation in prevalence rate among states¹. In India, escalating prevalence of the disease in animals is may be due to their rapid movement and in human beings due to the prevailing unhygienic condition which is

congenial for the organism^{2,3}. Yet, no systematic study has been carried out in Odisha state regarding the seroprevalence of this disease.

Hence, the present seroepidemiologic study was conducted to illuminate the status of brucellosis in cattle and high risk human beings of Odisha, India.

MATERIALS AND METHODS

Three hundred forty six serum samples comprising 258 from cattle and 88 from human beings were obtained following the guidelines of Institutional Animal Ethics Committee existing at that time from different districts of Odisha state and subjected to Rose Bengal Plate Test (RBPT) and Standard Tube Agglutination Test (STAT) and Indirect ELISA test to diagnose presence of antibodies against *Brucella*. Further, milk samples (n=87) collected were screened through Milk Ring Test (MRT).

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RBPT and STAT were carried out using RBPT antigen and Plain Brucella abortus agglutinating antigen respectively procured from the Division of Biological Products, Indian Veterinary Research Institute (I.V.R.I.), Izatnagar, Uttar Pradesh following previously described literature⁴. The Indirect ELISA test was carried out using smooth lipopolysaccharide (S-LPS) based AB-ELISA kit supplied by the Project Directorate on Animal Disease Monitoring and Surveillance (PD-ADMAS), Hebbal, Bangalore, India for presence of Brucella antibodies⁵. MRT was conducted on pooled milk samples using Abortus Bang Ring antigen obtained from Indian Veterinary Research Institute, Izatnagar as per previously described protocol⁴.

RESULTS AND DISCUSSION

Brucellosis is a highly contagious, zoonotic disease that causes great economic losses in terms of infertility, abortion, still birth and decreased milk production. Serological tests like RBPT, STAT, indirect immune fluorescence, and ELISA specially Dot-ELISA & Indirect-ELISA are frequently used in detection of brucellosis. But, following single serological test is not enough as each test has its limitations in different epidemiological situations⁶. Therefore, serum samples should be screened initially through preliminary tests followed by confirmatory tests in suspected samples⁷.

In this present study, RBPT and AB-ELISA were used which have high sensitivity and have been prescribed as screening tests at national

level and for international trade. STAT, though lacks sensitivity, is still used in sero-surveillance study because of its high specificity⁷. Screening of cattle serum samples (n=258) for detection of brucellosis showed 11 (4.26%), 6 (2.32%) and 21 (8.14%) positive by RBPT, STAT and AB-ELISA respectively (Table 1). Similar results have been found earlier where 4.9%, 3.5% and 8.4% samples from cattle were found to be seropositive in RBPT, STAT and I-ELISA respectively⁸. Lower (2.65%, 2.34% and 1.56%) and higher rate of brucellosis seroprevalence (11.21%, 16.0% and 24.30%) has also been reported previously using RBPT, STAT and I-ELISA test respectively^{9, 10}. 4.02%, 11.2%, 20.1% and 27% seroprevalence were recorded on the basis of RBPT in different investigations at Kerala (India), Ethiopia and Khartoum (Sudan) respectively^{11, 12, 13, 14}.

The prevalence rate using I-ELISA was found to be 8.14%. A wide range of prevalence rate i.e 40.18%, 10.4%, 5%, 50% and 22.18%^{15, 16, 17, 18, 19} respectively have been reported in past. This broad variation in seropositivity is possibly due to variation in sampling as different authors collected samples from animals having history of reproductive disorders or from organized dairy farms where artificial insemination are carried out frequently²⁰.

The district-wise prevalence of the disease varied from 1.81% to 23.52% (Table 1). High prevalence was reported in Balasore district which may be because; the samples presented to the department by veterinary officers were from animals with history of reproductive disorders. The reason for very low seroprevalence in Cuttack district may

Table 1. Geographical distribution of Brucellosis in different districts of Odisha

Species	Districts	Number of samples tested	I-ELISA		RBPT		STAT	
			No. of samples +Ve	(%)	No. of samples +Ve	(%)	No. of samples +Ve	(%)
Cattle	Khurdha	69	2	2.89	1	1.45	0	0
	Cuttack	55	1	1.81	0	0	0	0
	Jagatsinghpur	39	3	7.69	2	5.12	1	2.56
	Balasore	17	4	23.52	2	11.76	1	5.88
	Bhadrak	25	3	12.00	1	4.00	1	4.00
	Sambalpur	28	5	17.85	3	10.71	2	7.14
	Kalahandi	25	3	12.00	2	8.00	1	4.00
	TOTAL	258	21	8.14	11	4.26	6	2.32

Table 2. Sex wise prevalence of brucellosis in cattle by using RBPT, STAT and I- ELISA

Species	Sex	Total number of serum samples tested	Number of sera found positive by		
			I-ELISA	RBPT	STAT
Cattle	F	248	19(7.66%)	10(4.03%)	5(2.01%)
	M	10	2(20.00%)	1(10.00%)	1(10.00%)
Total		258	21(8.14%)	11(4.26%)	6(2.32%)

Table 3. Prevalence of Brucellosis in occupational workers and patients of pyrexia of unknown origin (P.U.O.) group by RBPT and STAT

Category	Occupation	No. of sera Tested	RBPT		STAT	
			No. of samples +Ve	(%)	No. of samples +Ve	(%)
Occupationally Exposed Group	Veterinary officer	14	0	0.00	0	0.00
	Farmer	11	1	9.09	0	0.00
	Animal handler	27	1	3.70	1	3.70
	Slaughter House Worker	16	2	12.5	1	6.25
	Total	68	4	4.41	2	2.94
Pyrexia Of Unknown Origin		20	1	5	1	5
Total		88	5	5.68	3	3.41

be attributed to the fact that samples were mostly collected from frozen semen bank farm and exotic cattle breeding farm where the animals are regularly screened for brucellosis. Various other factors which lead to perpetuation of disease are multispecies rearing, blooming commercial farming system, unrestrained movement of animals from one place to other etc²¹.

Prevalence of anti Brucella antibodies in different sex of cattle using three serological tests has been depicted in Table 2 and the data showed a higher percentage of reactors i.e., 20% in male cattle as compared to cows which was 7.66% by I-ELISA test. The current observation is contradictory to most of the past and contemporary findings where significantly higher prevalence in females has been reported^{22,23}. But, this result is in accent with some researchers who reported that antibody titers against Brucella are independent of sex^{24, 25}. Observed high prevalence in bulls represents an alarming situation as those were field samples supplied by veterinary officers from rural India where natural bull service is predominant. This will eventually lead to continuous spreading of infection. Along with this free grazing and

movement will also add to the wide distribution of brucellosis in animals.

Milk samples as an alternate can be used efficiently in screening dairy herds following milk I-ELISA or Milk ring test. MRT is a low cost alternative tool against milk I-ELISA and has been used frequently all over world⁷. Screening of milk samples (n=87), by MRT gave a seroprevalence of 3.44% which was in agreement with earlier findings^{26,27}. However, higher seropositivity in MRT (12.82 %, 18.26% and 33.34%) has also been reported from northern, central and southern regions respectively^{28, 19, 29}.

Brucella organisms contaminate the surroundings through urine and vaginal discharge and infection occurs via consumption of unpasteurised milk or through contact with various secretion and excretion of infected animals^{30,20,31}. Worldwide more than 500,000 human cases are reported annually³² and many cases are missed due to misdiagnosis, underreporting, unavailability of diagnostic services in remote areas and communication gap between veterinary and public health services^{33, 34}.

In order to study the occurrence of *Brucella* antibodies in occupationally exposed human beings, 88 serum samples were tested by RBPT and STAT yielding 5 (5.68%) and 3 (3.41%) positive samples (Table 3). The finding is in accordance with the past result where, through RBPT 5.9% prevalence was observed in veterinarians and shepherds in Bangalore, Karnataka³⁵. A little higher incidence than the present findings i.e 13.3%, 17.8% and 24.5% by RBPT has been reported^{36, 37}. A much lower incidence of the disease i.e., 1.2% was also revealed earlier³⁸.

The prevalence of brucellosis among Veterinary officer, Farmer, Animal handler, Slaughter House Worker and from patients with Pyrexia of Unknown Origin was 0.00, 9.09, 3.70, 12.5, 4.41 and 5% respectively (Table 3). The high prevalence rate among farmers and slaughter house workers may be because they are constantly exposed to infection due to contamination of hands and also they spend most of their time with animals. In a similar study it was found that among veterinary officers and pharmacists, paraveterinarians, animal attendants and dairy farmers the prevalence rate was 17.8%, 1.3%, and 5.8% respectively³⁷. In a group of 414 and 3,532 patients with PUO, 28(6.8%) and 28(0.8%) were identified seropositive respectively^{39, 40}. This variation of results may be due to difference in the degree of exposure of human beings to disease, small number of sample size and variation in the environmental conditions.

CONCLUSIONS

The current observation provides a baseline prevalence level of brucellosis in cattle and occupationally exposed human beings of Odisha. A more comprehensive study including small ruminants should be undertaken to elucidate the actual prevalence of the disease in Odisha. Prevalence of antibodies in high risk human beings warrants adaptation of more hygienic measures and awareness to counteract the disease communication.

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