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SEROPREVALENCE OF BRUCELLOSIS AMONG PEOPLE RESIDING IN THE RURAL AREAS OF WESTERN IRAQ

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ABSTRACT

This study was conducted to determine the seroprevalence of human brucellosis and identify the potential risk factors in a rural area of western part of Iraq. A simple random-sampling method was used for identifying 800 subjects for the study. Blood samples, collected from all the subjects, were studied following the methods of Rose Bengal slide agglutination and standard tube agglutination tests. More than 95% were seronegative, and 38 (4.75%) were seropositive. There was a statistically significant correlation between seropositivity and age. Sex, consuming fresh cheese and cream made from unboiled milk. Seropositivity was not related to educational level. It can be concluded from this study that pasteurization of milk and dairy products and education regarding eating habits must be pursued for eradication of human brucellosis from rural areas. The findings of the study suggest that human brucellosis is still an important public-health problem in the western parts of Iraq, especially in rural areas.

KEYWORDS: Brucella: Brucellosis: Seroepidemiologic studies; Iraq.

INTRODUCTION

Brucellosis is a public-health problem in many developing countries, including Iraq. The disease is zoonotic, and virtually all infections are derived directly or indirectly from animal exposure.[1,2]

Brucella strains cause diseases mainly in domestic animals, such as sheep, goats, cows, and pigs, resulting in important economic losses. Human brucellosis is notoriously a multisystem disease with varied manifestations, and the onset may be either acute or insidious. The diverse and sometimes deceptive manifestations of localized, sub-acute or chronic infection may lead to missing or delaying the diagnosis if the attending clinician has a low index of suspicion.[1,3]

Brucellosis is usually transmitted to humans by direct contact through abraded skin or mucosal surfaces, by consumption of contaminated milk and milk products, or by inhalation. Agricultural workers, veterinarians, and others involved in handling of animals or animal carcasses are at higher risk of direct inoculation. Individuals who ingest unpasteurized dairy products, especially from areas of endemic infection, are at significant risk of food-borne brucellosis.[1,2,4,5] Brucellosis can, therefore, be termed as an occupational disease. Person-to-person transmission of Brucella is extremely rare.[5]

Although the prevalence of Brucella is not exactly known in Iraq, the seropositivity has been reported to be about 2-6%.[6] The seroprevalence of brucellosis in sheep and cows in this country has been reported to be 1.97% and 1.43% respectively.[7]

The important characteristics of the studied region are the production and consumption of cream from unboiled milk and of fresh cheese from unpasteurized milk.

In this study, we aimed to determine the sero-prevalence of Brucella by Rose Bengal slide agglutination and standard tube agglutination methods in a region where domestic animal farming and agriculture is the main occupation.

MATERIALS AND METHODS

The study was carried out in the rural areas of seven districts of Anbar province/Iraq during April-May 2018, where nearly half of the population of the region live in rural areas. Of the1000 subjects invited to participate in the study, 200 did not agree to participate. Blood samples were collected from 800 subjects living in seven rural towns. The subjects were chosen by a simple random-sampling method from the area.

The subjects were informed about the nature of the study, and written consents were obtained. The participants were questioned as to whether they had ever eaten fresh cheese or raw cream until prior to three weeks before the date of questioning. Data were collected using a questionnaire comprising three groups of questions: (a) personal data (age, gender, education), (b) consumption of cream and fresh cheese from unboiled milk, and (c) direct contact with animals (milking, cleaning, shearing, handling, and feeding). The same physician completed the questionnaire by personal interview. The correlation between the habits of consumption of cream and fresh cheese from unboiled milk and brucellosis was examined.

Serum was separated from blood samples collected from the peripheral venous vessels. Whole blood was centrifuged, and serum was separated. The sera were stored at -40 °C. All the serum samples were studied by Rose Bengal slide agglutination test (RB) (Biotech, Barcelona, Spain) and standard tube agglutination test (SAT) (Wellcome Diagnostics, London, UK). All sera were routinely diluted from 1/20 to 1/1280. Each batch of the test included a positive control and a negative saline control. A definite agglutination of the suspension was read as a positive reaction. Agglutination was not seen in negative samples. For positive samples, the lowest positive titer was determined. Titration of 1/80 was accepted as exposure to Brucella, and 1/160 was accepted as brucellosis infection. Therefore, the titrations of 1/80 and over were accepted as positive.[6] The positive samples were screened with control serums four weeks later. We treated all the positive serum samples simultaneously with the 2-mercaptoethanol test for the detection of antibody classes either IgM or IgG. Although IgM titres are the first to rise after infection with brucellosis, IgM paradoxically remains elevated longer than IgG; thus, measures of IgG are traditionally used for monitoring the effectiveness of therapy.

The control samples that had increased or stabilized serum-specific IgG levels were referred to the local health institutions for follow-up and treatment. Other cases were informed about the results of laboratory investigations.

Statistical analysis

Data were processed using the SPSS software version 20 (SPSS Inc, Chicago, IL, USA). The mean age was given as M±SD. Statistical significance was tested with the chi-square test and Fisher's Exact test.

RESULTS

The 800 subjects (450 females and 350 males) enrolled in the study were aged 9-75 (median 34) years with a mean of 35.1±17.1 years. The seroprevalence of human brucellosis was 4.75%. Four percent was established as false negative according to SAT by the RB test. False positivity was not detected with the RB test. When compared with SAT, the sensitivity of RB was 94%, and its specificity was 95%. Serum levels of specific IgG increased in two of the positive cases after four weeks. The seroprevalence of human brucellosis was lower in cases aged less than 50 years compared to other age groups (p=0.0009). The seroprevalence of human brucellosis was higher among women (6.2%) than among men (2.9%) (Odds ratio [OR]: 0.44, 95% confidence interval [CI] 0.20-0.91, p=0.025). The educational status of the subjects was seronegative group (p=0.155).

Results showed that (26%) of the subjects had consumed cream from unboiled milk, and 9.3% of them were seropositive, while 74% had consumed cream from well-boiled milk, and 2.6% of them were seropositive (OR: 26, 95% CI 0.13-0.50, p=0.0005).

Human brucellosis was more prevalent among subjects consuming fresh cheese made from unpasteurized milk than among others (OR: 0.13, 95% CI 0.06-0.28, p=0.0005).

There was no statistically significant difference in sero-positivity in means of contact with animals (p=0.648). Contact with animals was defined as dealing with cattle, such as milking with automatic machines, handling and feeding. Not all of the persons who came in contact with animals were involved in the jobs of hand milking, shearing, cleaning and parturition.

Table (1): Distribution of seroprevalence of human brucellosis.

Characteristics of study subject	Seroprevalence (%)	Number(base)	Odds ratio	95% CI	P. Value
Age (years) groups					0.009
<50	20 (3.4)	590	1.0		
50-64	10 (8.3)	121	0.39	0.18-0.89	0.04*
≥65	8 (8.9)	89	0.36	0.15-0.88	0.04†
Sex					0.025
Male	10 (2.9)	350	1.0		
Female	28 (6.2)	450	0.44	0.20-0.91	
Education					0.155
illiterate	11 (7.6)	145	1.0		
Primary school	25 (4.1)	615	1.93	0.89-3.98	0.09†
Secondary school and Graduate –level college	2 (4.9)	40	3.18	0.51-71.22	0.270¶
Consumption of raw cream					
No	14 (2.6)	543	1.0		0.0005
Yes	24 (9.3)	257	0.26	0.13-0.50	
Consumption of fresh cheese					0.0005
No	10 (1.8)	565	1.0		
Yes	28 (11.9)	235	0.13	0.06-0.28	
Have contact with animals					0.648
No	8 (4.1)	195	1.0		
Yes	30 (4.9)	605	0.82	0.35-1.77	

* <50 with 50-64 years; + <50 with >65 years; † Illiterate with primary school; ¶iliterate with secondary school and graduate-level college; CI=Confidence interval

DISCUSSION

In the Asian countries where brucellosis is endemic and constitutes a serious public-health and economic problem in certain rural areas.[1,2,6] Many people are exposed to the disease each year and encounter physical invalidity, causing loss of work force.[8]

Results of studies conducted in Southern Italy, Jordan, and Southern Saudi Arabia showed that the overall prevalence of brucellosis was 3.1%, 16.7%, and 19.2% respectively.[4,9,10] The seropositivity of Brucella was assessed in various studies conducted in Iraq. The seropositivity of Brucella was reported to be 0.8-11.9% in various occupational groups of different countries.[6]

The seropositivity of brucellosis was found to be 3.2% in the elderly population in mid-Anatolia of Turkey.[6] Seropositivity among farmers was 6.2%.[11] Seropositivity in rural areas and dairy farms was reported to be 7.2% and 5.7% respectively.[12,13] The seroprevalence of human brucellosis found in the current study (4.75%) is similar to that found in other studies conducted in Iraq. We are unaware of any study about the prevalence of human brucellosis and animal brucellosis in our region.

Kaleli et al. reported the seropositivity of brucellosis as 8.9% in the 17-49-year age group and as 15.5% in the 50-69-year age group.[12] Al Sekait reported the seropositivity of brucellosis to be 9.7%, 16.0%, 18.7%, and 19.3% in the age groups of 0-14 years, 15-29 years, 30-44 years, and >45 years respectively.[14] In our study, seropositivity was higher in the older age group in agreement with other studies.

In the seroepidemiologic study carried out in a rural area of northwestern Greece, it was also reported that the incidence of brucellosis was higher in males (60.7%) than in females (39.3%).[8] Seropositivity in males and females has been reported to be similar in some studies.[4,6,14] In our study, the prevalence of brucellosis was higher in females (6.2%) than in males (2.9%).

In this study, the higher rate of seroprevalence of brucellosis in females compared to males was probably due to an increased involvement of women in farming domestic animals and handling their products. Males are generally employed in agricultural jobs. Contrary to our findings, seropositivity was higher (males 60.7%, females 39.3%) in males in a study in Greece.[8] The authors claimed that women who are occupied with household chores and raising of children spend less time with animals.

There was no correlation between the educational level and the seropositivity of brucellosis in our study. We think that the habit of consuming fresh cheese and raw cream is an acquired taste in all social-cultural-educational groups. This may be responsible for this finding.

In our study, the seroprevalence of brucellosis was higher in persons consuming fresh cheese and cream produced from unpasteurized milk (11.9% and 9.3% respectively). Consumption of fresh cheese and milk cream produced from unpasteurized milk has been reported to be a significant risk factor for brucellosis.[4-6,8,14] Contact with animals has been reported as one of the important risk factors for brucellosis.[4,6,8,14] However, contact with animals was defined as milking, cleaning, shearing and animal parturition. On the other hand, in our study, definition of contact with animals differed somewhat from the previous studies. Contact with animals was not so intense in our population. For example, milking was done by automatic machines, and shearing and animal parturition are performed by professionally, experienced persons. In addition, stable cleaning is done by machinery at definite periods. In this region, most stables are fenced with barbed wire, and the domestic animals live in a natural area which is called uncovered stable system.

Based on the findings of our study, we conclude that the main risk factor for brucellosis is consumption of fresh, unpasteurized dairy produce. This study also confirms that brucellosis continues to be a major public-health concern in the rural area of western Iraq. Increased risk has been attributed to certain dietary preferences, particularly fresh cheese, unpasteurized and unboiled milk, and cream. Production and consumption of cream made from boiled milk and consumption of cheese made from pasteurized milk or mature cheese (more than three months) may be the main protective factors for brucellosis.

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