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Research Article

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Seroprevalence of Acute and Chronic Brucellosis, Iraq

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Abstract

The current study conducted to estimating the prevalence of acute and chronic cases of human brucellosis with detect the association of infection to some epidemiological risk factors. Totally, 276 individuals of different ages, sexes and residences were selected and subjected to sampling of blood that tested qualitatively by ELISA targeting anti-brucella IgM and IgG antibodies to detect of acutely and chronically infected cases, respectively. The results revealed an overall 17.39% acute and 43.12% chronic infected individuals. Concerning severity of infection, there were 47.92%, 33.33%, and 18.75% mild, moderate and severe acute infections, respectively; while chronically, there were 68.07%, 20.17%, and 11.76%, respectively. Relation to socio-demographic factors, the findings of acute infections showed a significant increase in rates of positivity and relative risk in populations aged 20-50 years old than those of ≤ 20 years and ≥ 51 years; females more than males, and in those inhabitants rural than urban areas. Regarding chronic infection, significant higher values were recorded in individuals of 20-50 and ≥ 51 years old than those of ≤ 20 years old; however, individuals of 20-50 years old appeared having greatest risk than ≥ 51 years and ≤ 20 years. Also, females were showed a significant increase in positivity and risk than males, and rural individuals more than urban areas. In conclusion, there was a remarkable increase in prevalence of acute and chronic brucellosis in study areas; therefore, population education, early diagnosis, population medication and medical precautions are especially important to prevent the harmful effects of the disease and its complications.

Keywords: Diseases of dairy products, Brucella melitensis, Zoonotic diseases, IgG antibodies, IgM antibodies, Al-Qadisiyah province

Introduction

Brucellosis is a common prevalent zoonotic infectious disease of bacterial origin, *Brucella* Genus in the *Hyphomicrobiales* Order of *Alphaproteobacteria* Class. Twelve *Brucella* species were detected, known to be transmitted from animals to humans; however, only four species have importance for human including *B. melitensis, B. suis, B. abortus,* and *B. canis* [1,2]. Brucella organisms are small aerobic facultative intracellular coccobacilli which localize in the reproductive organs of host animals such as sheep, goat, cattle, camels and pigs causing abortions and sterility [3,4]. The large shedding of organisms in urine, milk, and placental fluid demonstrated to act an observable part for transporting of organism to human [5]. Globally, many cases recorded yearly due to ingestion of unpasteurized dairy products, touching blood and body fluids of infected animals, direct contact with contaminated fomites [6-8]. People may also be infected by inhalation of contaminated dust or aerosols, and such as, it has labeled *Brucella* species as highly weaponizable [9].

The ability of brucellosis to affect any organ or any system results in variable clinical forms ranged from asymptomatic or nonspecific symptoms to severely debilitating illness with serious public health consequences [10,11]. Hence, definitive diagnosis based on culture, serologic and molecular techniques or both is necessary to warrant initiation of therapy and for epidemiologic surveillance [12]. Culture is the gold-standard method in diagnosis of most bacterial infections including *Brucella*; however, the rate of sensitivity depends on the phase of disease, previous use of antibiotic and the type of sample [13,14]. In addition, it is time consuming in particular in testing a large number if samples, required a standard biosafety level to avoid the risk of contamination by the specimens, and failure to detect the pathogen is frequent occurrence [15]. In absence of culture facilities, the diagnosis of brucellosis traditionally relies on serological testing with a variety of traditional assays such as Rose-Bengal, standard tube agglutination, and the Coombs tests [16,17]. However, these methods have important limitations like their poor sensitivity during the early stage of disease, and reduced their specificity in highly endemic areas and where frequent relapses of the disease occur [18,19]. ELISA has become increasingly popular as an advanced, well standardized diagnostic technique for brucellosis [20,21]. The sensitivity of ELISAs prepared in the laboratory may be high especially when the detection of specific IgM antibodies is complemented with the detection of specific IgG antibodies [22,23].

In Iraq, several studies have been conducted to estimate the prevalence of human brucellosis using different diagnostic assays [24-27]; however, no online available information concerned detection the prevalence of acute and chronic cases of human brucellosis using the ELISA. Therefore, this study conducts to estimate the seroprevalence of anti-brucella IgM and IgG antibodies in Al-Qadisiyah province (Iraq), with evaluate the association the rate of positivity to some epidemiological risk factors (age, gender and residence).

Materials and Methods

Ethical approval

The Scientific Committee in the Department of Microbiology (College of Medicine, University of Al-Qadisiyah) licensed the current study.

Samples

Totally, 276 individuals of varied ages and genders were selected from different rural and urban areas localized in Al-Qadisiyah province (Iraq) during April-August (2023). Under aseptic conditions, 2.5 ml of venous blood was samples from each one into plastic anticoagulant EDTA tubes using of disposable syringes. In laboratory, the samples of blood were centrifuged (5000 rpm/5 min), and the obtained sera were kept frozen into labeled Eppendorf tubes at 4°C. Epidemiological risk data of study population were recorded to estimate their association to rate of positivity.

Serology

Two qualitative ELISAs' kits were invoiced from the SunLong Biotech Company (China); Human Brucella IgM ELISA Kit (Cat. No. SL2196Hu) and Human Brucella IgG ELISA Kit (Cat. No. SL0378Hu) and served in this study to estimate the prevalence of acute and chronic brucellosis. Following the manufacturer instructions of each kit; the sera and kit contents were prepared and processed and the optical density (OD) were measured at 450 nm with finally calculation the effectiveness and critical value (CUT OFF). Study individuals were considered positive if the ODs of each ELISA's kit were ≥ CUT OFF value and negatives if they < CUT OFF value. According to their severity, the positive ODs were divided into three levels; Mild, moderate and severe infections.

Statistical analysis

GraphPad Prism Software (*version* 6.0.1) was served for evaluation significant variation among positive findings at $P < 0.05$ using the *t*-test; whereas, association of positive rates to epidemiological risk factors were determined throughout the Odds Ratio. Values were recorded as either mean ± standard deviation or as number (percentage), [28].

Results

Among 276 individuals tested by specific ELISA kits, the findings were revealed 48 (17.39%) acute infections and 119 (43.12%) chronic infected individuals at a significant difference of P0.0264 (Figure 1).

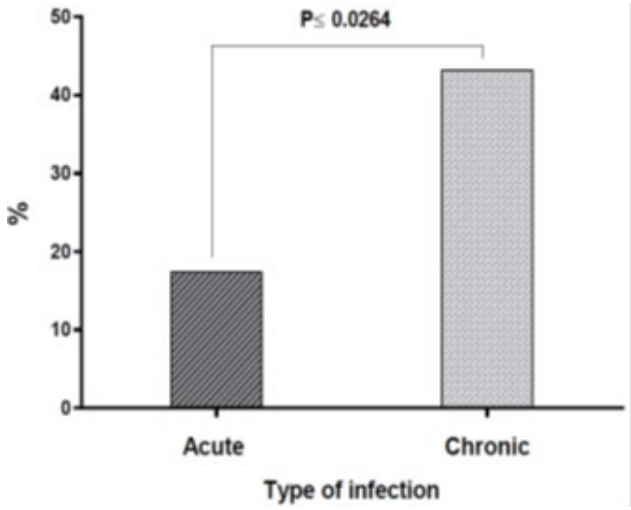

Figure 1: Total results of acute and chronic infections detected by ELISA (Total No: 276).

Concerning severity of infection, the results of acute infections showed that 47.92% (23/48), 33.33% (16/48), and 18.75% (9/48) have mild, moderate and severe infections, respectively, at a significance of $P < 0.0416$ (Figure 2). According the level of ODs, the mild, moderate and severe infections were reported 0.290 ± 0.031, 0.377 ± 0.016, and 0.416 ± 0.014, respectively at a significance of $P < 0.0349$ (Figure 3).

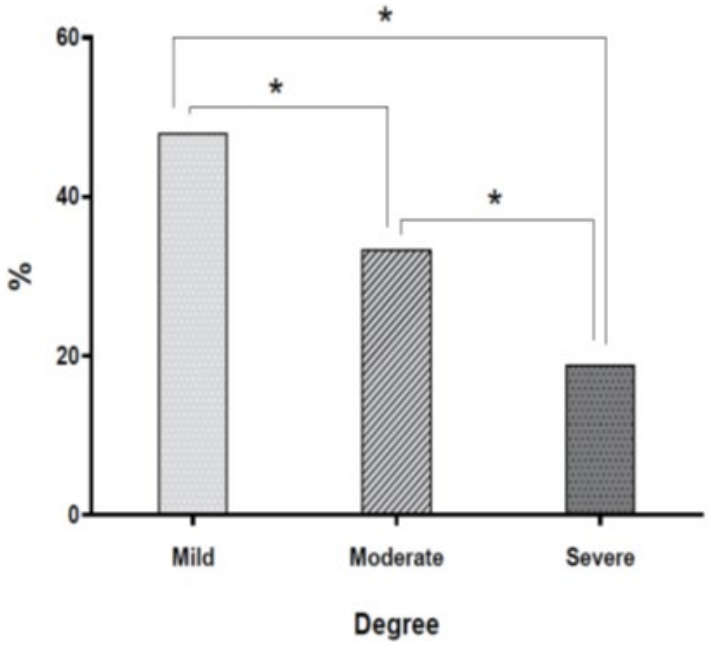

Figure 2: Degree of severity according to number of acute cases (Total No: 48).

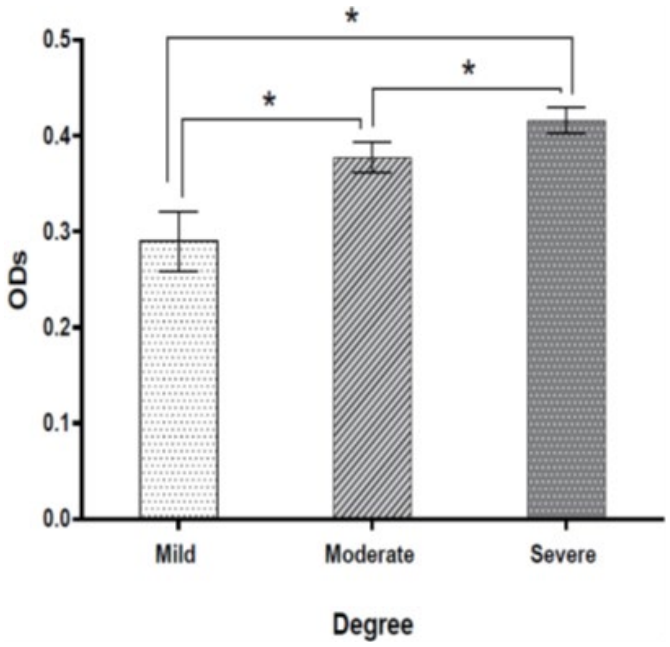

Figure 3: Degree of severity according to ODs of acute cases (Total No: 48).

While, the results of chronic infections were showed 68.07% (81/119) mild, 20.17% (24/119) moderate, and 11.76% (14/119) severe infections at a significance of $P < 0.0207$ (Figure 4). For the level of ODs, values of mild, moderate and chronic infections were 0.281 ± 0.031, 0.379 ± 0.014, and 0.431 ± 0.021, respectively at a significance of $P < 0.0271$ (Figure 5).

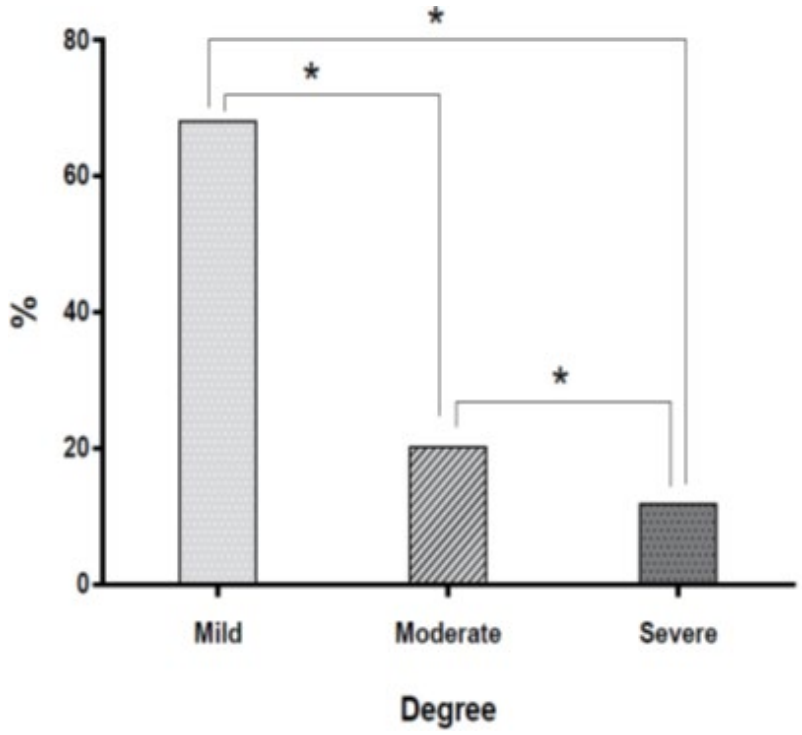

Figure 4: Degree of severity according to number of chronic cases (Total No: 119).

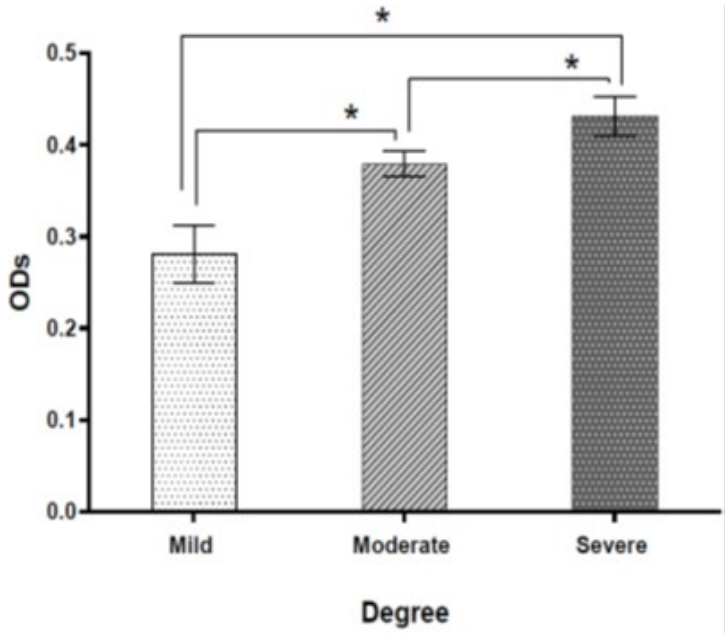

Figure 5: Degree of severity according to ODs of chronic cases (Total No: 119).

Relation to socio-demographic factors, the findings of acute infections were showed an observable difference ($P < 0.05$) in their values (Table 1); however, individuals aged 20-50 years old were reported a higher rate of positivity (21.43%) and risk (1.529) than values of ≤ 20 years (12.68% and 0.668, respectively) and ≥ 51 years (15.19% and 0.831, respectively). Females were showed a significant elevation ($P < 0.05$) in positive rate (23.44%) and risk (1.5) when compared to males (15.57% and 0.667, respectively). Significantly, individuals of rural areas were recorded an increasing in rates of positivity (29.71%) and risk (5.824) in comparison with those of urban areas (5.07% and 0.172, respectively).

Factor (Group)	Total No.	Positive		Odds Ratio	Risk
		No.	%		
Age (Year)					
≤ 20 years	71	9	12.68	0.617	0.668
20-50	126	27	21.43	1.675	1.529
≥51 years	79	12	15.19	0.799	0.831
p-value			0.0473	0.0135	0.0176
Sex					
Female	64	15	23.44	1.663	1.5
Male	212	33	15.57	0.601	0.667
p-value			0.0476	0.0001	0.0001
Residence					
Rural	138	41	29.71	7.981	5.824
Urban	138	7	5.07	0.125	0.172
p-value			0.0169	0.0001	0.0001

Table 1: Association of acute infection to some socio-demographic risk factors.

Regarding the results of chronic infection, significant increases ($P < 0.05$) in rate of positivity were recorded in individuals of 20-50 years old (56.35%) and ≥ 51 years old (54.43%) when compared to those of ≤ 20 years old (7.04%). However, individuals of 20-50 years old appeared at higher risk of human brucellosis (1.763) than those of ≥ 51 years (1.409) and ≤ 20 years (0.126). Females were showed a significant increase ($P < 0.05$) in rate of positivity (51.56%) and risk (1.271) than males (40.57% and 0.787, respectively). Also, rural individuals were recorded more rates of positivity (51.45%) and risk (1.552) than those of urban areas (35.51% and 0.689, respectively), (Table 2).

Factor (Group)	Total No.	Positive		Odds Ratio	Risk
		No.	%		
Age (Year)					
≤ 20 years	71	5	7.04	0.095	0.126
20-50	126	71	56.35	2.741	1.763
≥51 years	79	43	54.43	1.901	1.409
p-value			0.0361	0.0001	0.0001
Sex					
Female	64	33	51.56	1.567	1.271
Male	212	86	40.57	0.638	0.787
p-value			0.0438	0.0001	0.0001
Residence					
Rural	138	71	51.45	1.924	1.552
Urban	138	49	35.51	0.52	0.689
p-value			0.0419	0.0001	0.0001

Table 2: Association of chronic infection to some socio-demographic risk factors.

Discussion

During the last two decades, the breakdown of public health systems in resources-poor and politically troubled countries has resulted in new foci of disease in Asia and a worsening situation in many countries including Iraq [27]. In this study, the findings revealed that the seroprevalence of chronic cases (43.12%) was more prevalent than acute cases (17.39%). Subsequently, the mild infection of both acute and chronic cases was appeared more significantly than moderate and severe infections. Several local and global researchers have been studied the prevalence of acute and/or chronic brucellosis in humans using of various diagnostic assays with recording different results [24,26,29, 30]. Comparatively, there were 13.13% in Pakistan [31], 14.96% in Ethiopia [32], 17% in Uganda [33], 18% in Turkey [34], 23.3% in

Sudan [35], 29.5% in Iran [36], and 30.07% in Iraq [27]. However, the high prevalence of mild infections in both acute and chronic cases of our study might refer to endemic status of disease in study areas, frequent exposure to causative pathogen through ingestion of unpasteurized dairy products, directly and indirectly contacting with diseased animals or their contaminated reproductive fluids.

Significant variable distribution of acute and chronic human brucellosis among different socio-demographic risk factors was seen in this study. Several studies demonstrated that human brucellosis can occur at any age, but the peak occurs in young adults as observed in acutely and chronically infected cases of our study in young adults of 20-50 years followed in ≥ 51 years [26,37,38]. Also, this study noted that brucellosis increased significantly in females than males, and in individuals inhabitant rural than urban areas. In a previous study, Gür et al. [39] reported the incidence of brucellosis in 63% of patients aged 15-45 years, and 19% in patients aged 7-14%; with significant recognizing of disease in adult men. Dean et al. [40] mentioned that the proportion of male patients was greater than females in both children and adults attributing this variation for accessing to health care and household responsibilities. Al-Bayaa [41] found that population aged 18-39 years having a higher rate of brucellosis (22%) than other age groups; 40-59 (15%) and ≥ 60 (7%) years with lack of significance between females (13.7%) and males (11.1%). He thinks that the link between brucellosis and sex factor depends on the lifestyle of the community. In Saudi Arabia, Aloufi et al. [42] found that the greater number of brucellosis in the 15-44 years age than in any other age group; and males has a significantly higher risk than females. In Kenya, Akoko et al. [43] reported that 38.5% of human samples were positives for *Brucella*, with significant increases in population aged 21-40 years (52.5%) than ≤20 (27.6%) and < 40 (31.5%) years, with lack of significance between males (36.7%) and females (39.9%). High prevalence of brucellosis in rural areas might be interpreted primarily by that the rural population engaged in agriculture for which field animals are used; hence, occupational exposure to these animals and their secretion/excretions results in increasing the risk of disease [44]. Munyua et al. [45] reported the high incidence of human brucellosis in a rural pastoralist community in particular in population aged 20-41.5 years (75%) and females (75%) suggesting the occurrence of higher rates of brucellosis is correlated with *Brucella*-seroprevalence in livestock and contact with unpasteurized dairy products.

Conclusion

The prevalence of acute and chronic cases indicates that the study areas are endemic to brucellosis and the wide diversity of disease in animals that serve as a source for transmission of infection. Hence, disease control in endemic areas relies heavily on identifying the related risk factors, implementing measures to reduce the spread of infection, and using the precise diagnostic tests at the right time, since misdiagnosis and incorrect treatment ensure when tests fail to accurately detect the disease.

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Conflict of Interest

No.

Funding

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