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Original Article

Seroprevalence of Brucellosis among General Population in Famenin City, Western Iran in 2016: The Famenin Brucellosis Cohort Study

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Abstract
Background: Brucellosis is endemic in Iran with a higher level of endemicity in western areas, including the Hamadan province. This study aims to define the seroprevalence of brucellosis and it,s risk factors in general the population of Famenin, Hamadan province, in western Iran.
Methods: This survey was conducted on 2367 participants in Famenin and its villages from September to November 2016. After receiving written consent from subjects, demographic information was obtained through questionnaires and 10cc blood samples were taken from the participants. Blood samples were sent to the Core facility of Hamadan University of Medical Sciences and were tested using Wright and 2ME kits (Pasteur Institute, Iran) for serological detection of brucellosis. The seroprevalence of brucellosis was reported as percentage with 95% confidence interval (CI).
Results: Totally, 2367 individuals with the mean age (SD) of 34.6 (20.9) (range: 2 to 95) years were enrolled. Of these, 1060 (44.8%) were men and 1610 (68.0%) lived in rural areas. The seroprevalence of brucellosis according to the Wright titer (equal to or greater than 1:80) was 6.6% (95% CI: 5.62%, 7.66%). The corresponding prevalence based on 2ME titers (equal to or greater than 1:40) in subjects with positive Wright test was 37.2% (95% CI: 29.5%, 44.84%). We saw a significant association between the incidence of brucellosis and occupation ($P < 0.001$) and type of contact with livestock ($P = 0.009$) as two important risk factors.
Conclusion: The seroprevalence of brucellosis in Famenin population was considerable. Contact with livestock, animal husbandry, farming and history of brucellosis were risk factors for brucellosis infection.
Keywords: Brucellosis, Famenin, Iran, Seroprevalence, Zoonotic infectious disease
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Introduction

Brucellosis is a common bacterial disease in both humans and animals, which is transmitted through unsafe contact with infected animals or consuming contaminated dairy products. In brucellosis, we encounter a range of clinical manifestations from asymptomatic infection to severe forms.[1] *Brucella* is a gram-negative coccobacillus without spores and capsules with at least four species that have been pathogenic for humans; *Brucella abortus, Brucella suis, Brucella melitensis* and *Brucella canis*.[2] The role of phagocytosis and cellular immunity in brucellosis immune response have been demonstrated.[2] *Brucella* involves various organs and can exhibit non-specific symptoms such as weakness, bone pain, fever, muscle aches, weight loss, sweating and headache.[2,3]

In Iran, brucellosis is an endemic disease with *B. melitensis* as the most common pathogen. The incidence of brucellosis is high in some provinces of Iran such as Lorestan, Kermanshah and Hamadan.[4,5] Hamadan, with incidence rates of 31.7 per 100 000 population in 2008, 36.7 per 100 000 population in 2010, as well as 62.9, 81.4 and 91.0 per 100 000 population in the years 2012, 2013, and 2014, respectively, was among the provinces with the highest prevalence of brucellosis.[4,6] Administrative reports from national and provincial notifiable diseases surveillance systems indicated that the incidence rate of brucellosis in Iran was 21 per 100 000 population, while in Hamadan, it was approximately three times the rate of national occurrence in 2012. The corresponding incidence rate per 100 000 population in Famenin was 105 in 2014.

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In areas where brucellosis is endemic, there are many undiagnosed patients with asymptomatic exist who are not diagnosed and fewer than 10% are detected after an infection in primary phases.[7,8]

The diagnosis of brucellosis is usually confirmed by *Brucella* isolation from blood or bone marrow or by demonstrating rising antibody titers to *Brucella* antigens in blood samples. Numerous serological tests are used for detection of brucellosis.[3] In our country, Wright and 2ME tests are routinely used for establishing the diagnosis.[9-11]

Our aim in this study was to assess the brucellosis seroprevalence and the most important risk factors for brucellosis infection in order to define effective strategies for control and reduction of brucellosis in the general population of Famenin (a city in the province of Hamadan).

Material and Methods

Study Design and Sampling Method

This study was carried out on the general population of Famenin and its surrounding villages during a three-month period (September to November 2016) .
The number of samples was calculated using the following formula.

$$n = \frac{z^2_{1-\frac{\alpha}{2}}(p)(1-p)}{(d)^2}$$

Considering the prevalence rate of brucellosis, with 20% of people having Wright's test titers between 1:20 and 1:5120 in the region ($P = 0.20$), we considered z = 1.96 (for $\alpha = 0.05$) and d (absolute error) = 0.018. Finally, the sample size was calculated at 1898; by applying a design effect equal to 1.25, the required blood sample size was calculated at 2371.

The cluster sampling method was used after classification of the Famenin population based on urban and rural areas of residence. The urban population of Famenin in 2016 was 17979 (~42%) while the rural population was 25065 (~58%). The total clusters were selected proportional to the urban and rural population. Each cluster included 10 households or 40 people. So based on this, 1440 people (36 clusters) in villages and 960 people (24 clusters) in cities were calculated.

To select the head of the clusters, the first and last postal code of each division was first obtained from the post office. Based on the aggregation of the postal codes, the mentioned clusters were proportional to the density of the postal codes of each division. Large divisions with more households accounted for a greater fraction of the sample size. Then, using systematic sampling considering each category sample size, the postal codes of head clusters from rural and urban areas were selected and sampling in each cluster was performed consecutively from the first postal code and continuing to include 40 people or 10

households. The questionnaire was administered to the any household head of cluster, and continued to reach 10 households or 40 individuals (preferably at least 40 in 10 households). Using this systematic approach, we achieved a geographically representative distribution of eligible clusters. The address of the head of clusters was obtained from the post office to dispatch the questionnaire and collect data. To select a random number to start systematic sampling, Excel software was used. If the postal code did not belong to a home, the nearest postal code would be replaced. In case of absence of a household, the questionnaire was completed twice within 24 hours with on-site presence and on the third referral (second follow-up), by giving a special form of referral to neighbors; to invite the family members for questioning and sampling. If a member of the household wished to participate in the study, the interviewer completed the questionnaire with informed consent, regardless of the non-cooperation of the other household members. If the household did not cooperate, the interviewer continued by completing the questionnaire for the next household. All subjects of the urban and rural population of Famenin entered the study between September to November 2016. Individuals younger than two years were not included in the study. According to the cluster sampling, households in Famenin were determined by coding the areas and households.

Data Gathering

After receiving written consent, health workers in the area asked each participant about the maximal data relating to brucellosis including sex (male, female), pregnancy (yes, no), marital status (married, single), education (illiterate, elementary, middle school, high school, diploma, super-diploma and higher), residence (city, village), occupation (animal husbandry farmer, housewife, other), history of brucellosis (yes, no), history of infection (yes, no), recurrence history among people with a history of infection (yes, no), number of relapses (among people with recurrence history), current treatment for brucellosis among people with a history of infection (yes, no), contact with livestock (yes, no), livestock species (cow, small ruminants), type of contact with the livestock (contact with aborted fetus, contact with live animals, contact with discharge after delivery, keeping livestock, slaughtering the livestock, contact with meat and organs, contact with carcasses and livestock secretions after slaughter, milking livestock), use of masks in people who have contact with the livestock (yes, no), using local dairy (yes, no), local dairy type (milk, skim milk, cheese, butter, cream curd, colostrum, traditional ice cream) and brucellosis infection in family members during the past two years (yes, no) which were entered into questionnaires by questioners.

Diagnostic Test

Brucellosis was considered in any subject with Wright test

equal to or greater than 1:80 and a 2ME test equal to or greater than 1:40 with clinical symptoms consistent with brucellosis (fever, sweating, weight loss, loss of appetite and muscle or bone pain) or positive blood cultures.[6] Brucellosis infection was defined by positive serology with or without clinical symptoms compatible with brucellosis.[3] The sensitivity and specificity of Wright test based on previous studies were reported at 91.8%–100% and 63.6%–100%, respectively.[9,12-14] The sensitivity and specificity of 2ME was reported at 52%–93.7% and 75%–100%, respectively.[9,15] Wright and 2ME kits by Pasteur Institute of Iran were used to assess the seroprevalence of brucellosis. Ten cc blood samples were taken from each subject and sent to the core facility of Hamadan University of Medical Sciences (microbiology laboratory) for Wright and 2ME tests. 2ME tests were performed for those who had Wright titers equal to or greater than 1:40.

Statistical Analysis

After reviewing the questionnaires, we analyzed all data using SPSS software version 16. Numerical variables were presented as mean and standard deviation and qualitative variables as frequency and percentages. The seroprevalence of brucellosis was reported as percentage with 95% confidence interval. We used the chi-square test to investigate the relationships between qualitative variables. The comparison between quantitative variables in different groups was performed using t test. Because of cluster sampling, to define the effective factors and risk factors for the occurrence of brucellosis and its prevalence, a mixed effect logistic regression model was used. In the mixed effect model, the estimation of standard error (confidence interval) of parameters adjusts because of cluster sampling. To fit the mixed effects logistic regression model and to determine the most important factors influencing Wright test positivity, the method of backward selection of variables was used. In this method, we considered a P value of less than 0.05 to enter variable into the model and a P value of less than 0.1 to exit the model. The significance level in all statistical tests was less than 0.05. It is notable that the missing data for variables were few (less than 2%) and inconsiderable and they were deleted when analyzing.

Results

Out of a total of 2400 eligible individuals who enrolled in the study and completed the questionnaire, due to repeated codes or lack of questionnaires, finally, 2367 blood samples entered the final analysis. Table 1 shows the sample's main characteristics. The mean age of 2367 participants was 34.6 years with a standard deviation of 21.0; median age was 34 (range 2 to 95) years. Out of these, 1060 (44.8%) were men, and 1472 (62.1%) were married. A total of 1610 subjects (68%) lived in villages. Pregnancy was registered in 31 out of 604 (13.5%) married women from 10 to 49 years of age (Table 2).

Table 1. Participants' Main Characteristics

Comparison of Literacy Ratio of Study Population and Population of Famenin County (in Those Aged Over 6)

	Population of Famenin County									Study Population								
	Village			City			Total			Village			City			Total		
Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)
	71.59	83.11	77.66	82.05	90.02	86.15	75.44	85.57	80.73	70.20	82.00	75.55	81.89	89.18	85.61	73.67	84.58	78.48

Comparison of Sex Ratio of Study Population and Population of Famenin City

	Population of Famenin County									Study Population								
	Village			City			Total			Village			City			Total		
Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)	Women (%)	Men (%)	Total (%)
	47.37	52.63	100	48.38	51.62	100	47.71	52.29	100	54.33	42.67	100	50.73	49.27	100	55.22	44.78	100

Comparison of Age Groups of Study Population and Population of Famenin City

	Population of Famenin County				Study Population			
Total (%)	Under 15 years old (%)	Between 15–65 years old (%)	>65 years old (%)	Total (%)	Under 15 years old (%)	Between 15–65 years old (%)	>65 years old (%)	Total (%)
	9993 (29/39)	26467 (67/25)	2899 (7/37)	39359 (100)	537 (22/69)	1605 (67/81)	225 (9/50)	2367 (100)

Table 2. Demographic Characteristics of Participants in Famenin Brucellosis Cohort Study

Variable	Variable Levels	No. (%)
Sex	Male	1060 (44.8)
	Female	1307 (55.2)
	Total	2367 (100)
Pregnancy (In married women, ages 10–49)	Yes	31 (5.13)
	No	573 (94.87)
	Total	604 (100)
Marital status	Married	1471 (62.147)
	Single	894 (37.769)
	Missing data	2 (0.084)
	Total	2367 (100)
Education	Illiterate	635 (26.830)
	Elementary	916 (38.698)
	Guidance	399 (16.856)
	High school	145 (6.125)
	Diploma	194 (8.196)
	Super-diploma and higher	78 (3.295)
	Total	2367 (100)
Habitat	City	757 (31.98)
	Village	1610 (68.02)
	Total	2367 (100)
Occupation	Animal husbandry	171 (7.224)
	Farmer	258 (10.900)
	Veterinarian, vaccinator, slaughterhouse staff, butcher, lab staff, dairy sales, or cook	8 (0.336)
	Housewife	1006 (45.501)
	Other (Students, children, non-respondents, etc.)	1053 (44.487)
	Total *A	22496
History of brucellosis	Yes	174 (7.351)
	No	2182 (92.184)
	A history of more than one times	1 (0.042)
	Missing data	10 (0.423)
	Total	2367 (100)
History of infection	Forgotten	4 (2.281)
	From 1976 to 1986	19 (10.858)
	From 1987 to 1996	62 (35.428)
	From 1997 to 2006	52 (29.714)
	From 2007 to 2016	35 (20)
	Missing data	2 (1.714)
	Total	174 (100)
Recurrence history (among people with a history of infection)	Yes	12 (6.89)
	No	162 (93.11)
	Total	174 (100)
Number of relapses (among people with recurrence history)	One or two times	8 (66.666)
	Missing data	4 (33.334)
	Total	12 (100)
Current treatment for brucellosis (among people with a history of infection)	Yes	2 (1.15)
	No	172 (98.58)
	Total	174 (100)
Contact with livestock	Yes	785 (33.164)
	No	1579 (66.708)
	Missing data	3 (0.128)
	Total	2367 (100)
Livestock species	Cow	206 (26.424)
	Sheep	684 (87.134)
	Goat	188 (23.949)
	Missing data	5 (0.637)
	Total *B	1083

Table 2. Continued

Variable	Variable Levels	No. (%)
Type of contact with livestock	Contact with aborted Fetus	176 (22.420)
	Contact with live animals	625 (79.618)
	Contact with discharge after delivery	160 (20.382)
	Keeping livestock	560 (71.338)
	Slaughtering the livestock	73 (9.229)
	Contact with meat and organs	144 (18.334)
	Contact with carcasses and livestock secretions after slaughter	79 (10.064)
	Milking livestock	130 (16.561)
	Missing data	3 (0.382)
	Total *C	1950
Use of masks (people who have contact with the livestock)	Yes	137 (17.452)
	No	641 (81.656)
	Missing data	7 (0.892)
	Total	785 (100)
Using local dairy	Yes	2082 (88.4)
	No	274 (11.6)
	Total	2356 (100)
Local dairy type	Milk	1977 (95.5)
	Skim milk	326 (15.7)
	Cheese	644 (31)
	Butter	237 (91.6)
	Cream	200 (17.6)
	Curd	366 (17.96)
	Colostrum	47 (2.3)
	Traditional ice cream	170 (8.2)
	Missing data	2 (0.096)
	Total *D	2082
Brucellosis infection in family members during the past two years	Yes	51 (2.2)
	No	2316 (97.8)
	Total	2367 (100)

*A: Due to more than one job for some people, the sum is more than 100 %.
*B: Due to contact with more than one type of livestock, total percentages are more than 100%.
*C: Due to several methods of contact with the livestock for an individual, the percentage is more than 100%.
*D: Due to consumption of more than one type of dairy products, the sum of percentages is more than 100%.

In the population studied, 1859 subjects (78.5%) had a negative Wright test and in the remaining 508 (21.5%) the Wright titers were between 1:20 and 1:5120. The prevalence with direct standardization based on age of the population is reported in Table 3. The ICC (Intra class correlation) obtained was less than 0.01 and we did not consider the design effect in prevalence calculation. However, in 156 participants, Wright titers were equal to or greater than 1:80 (Table 3). In total, the seroprevalence of brucellosis in the examined population of Famenin based on the serology titer of Wright test equal to or greater than 1:80 was 6.6% with a standard deviation of 0.005 and 95% confidence interval 5.62%–7.66%. Table 4 shows that in the total population studied in Famenin (city and villages), 156 had a Wright test ≥1:80, 58 of them had 2ME titer ≥1:40 and in the rest of 98

Table 3. Distribution of Wright Test Results Based on the Location of the Participants

Wright titer*	City		Villages		Total		
	F	FP	F	FP	F	FP	DSP
Negative	565	74.6%	1294	80.4%	1859	78.5%	—
1:20	64	8.5%	111	6.9%	175	0.39%	77.1
1:40	76	10%	101	6.3%	177	7.47%	39.0
1:80	43	5.7%	57	3.5%	100	4.22%	20.2
1:160	9	1.2%	28	1.7%	37	1.60%	14.8
1:320	0	0%	12	0.7%	12	0.50%	3.3
1:640	0	0%	5	0.3%	5	0.21%	2.0
1:1280	0	0%	0	0%	0	0%	0.0
1:2560	0	0%	1	0.1%	1	0.04%	0.4
1:5120	0	0%	1	0.1%	1	0.04%	0.4
Total	757	100%	1610	100%	2367	100%	157.2

F, frequency; DSP, direct standardized prevalence; FP, frequency percent.
* The prevalence rate based on Wright titer ≥1:20 in all samples (2367) was 21.46% (confidence interval: 19.81% – 23.10%). The prevalence rate based on Wright titer ≥1:80 in all samples (2367) was 6.59% (confidence interval: 5.62% – 7.66%).

subjects, the 2ME titer was ≤1:20. The seroprevalence of brucellosis based on the 2ME titer of ≥1:40 was 37.2% when Wright titers were ≥1:80 and it was 2.4% among all 2367 subjects (Table 5). Besides, there was a significant difference between mean age ± SD of subjects with and without positive serology of Wright test (42.2 ± 19.8 and 34.0 ± 20.9 years, respectively; $P < 0.0001$).

Table 6 shows that there was a correlation between occupation and positive serology of Wright test in the population studied in Famenin. Also, there was a significant relationship between history of contact with livestock as well as type of contact with livestock and positive serology in the population studied in Famenin.

Moreover, Table 7 shows the final model that was obtained with backward method in using multiple mixed effects logistic regression. The odds ratio of the disease was 3.74 (2.818–4.963), ($P < 0.001$); in other words, a history of brucellosis had a positive effect on Wright test results.

Discussion

In Iran, brucellosis is regarded as an endemic disease, and its most common cause is *B. melitensis*.[4] As a result of the highly endemic epidemiological state, the accurate incidence of clinical brucellosis is not clear.[16] The seroprevalence of brucellosis in our study according to the serologic titers of Wright test equal to or greater than 1:80 in the total sample was 6.6% and the seroprevalence of brucellosis based on 2ME titers ≥1:40 in subjects with positive Wright titers was 37.2% while in total, it was 2.4%; it appears that the number of brucellosis cases was higher than those reported during the same year. In a study by Bokaie et al in Birjand among 422,106 individuals, the prevalence of brucellosis was 37 per 100 000 people, which is equal to 0.03% and less than the prevalence we found.[17]

In our study, men constituted less than half of the sample, and the mean age was almost 35 years; in addition, 67.8% lived in rural areas. Brucellosis has the ability to infect individuals of all ages, but is more likely to be seen

between the ages of 20 and 30 years. Also, the disease is seen in both genders with various predominance.[4] In the study by Bokaie et al, brucellosis was more prevalent in men than women, and in rural parts than urban areas, with the age group of 20–40 years most frequently affected, which is similar to our findings.[17] In a study by Poorhajibagher et al, out of 318 patients, 42 (13.2%) had a Wright test equal to or greater than 1:80 and brucellosis was more common

Table 4. Distribution of Subjects According to Wright (≥1.80) and 2ME Titers

2ME Test*	Wright Test						Total
	1/80	1/160	1/320	1/640	1/2560	1/5120	
Negative	61	12	1	0	0	0	74
1:20	17	7	0	0	0	0	24
1:40	12	6	1	0	0	0	19
1:80	10	7	6	1	0	0	24
1:160	0	5	4	0	0	0	9
1:320	0	0	0	1	1	0	2
1:640	0	0	0	3	0	1	4
Total	100	37	12	5	1	1	156

*The prevalence of brucellosis in subjects with a 2ME titer ≥1:20 in all samples (156) was 52.66% (confidence interval: 44.64%–60.48%) and the prevalence of brucellosis in subjects with a 2ME titer ≥1:40 in all samples (156) was 37.17% (confidence interval: 29.50%–44.84%)

Table 5. Seroprevalence of Brucellosis Based on 2ME Titers in All Subjects (2367)

2ME Titers	Number of Subjects	Brucellosis Seroprevalence in A
≥1:20	82	3.46 (Confidence interval: 2.72%–4.20%)
≥1:40	58	2.45 (Confidence interval: 1.82%–3.07%)

Table 6. Multiple Mixed Effect Logistic Regression Analysis in Subjects with a History of Brucellosis Associated with the Positive Wright Test

Variables	Odds Ratio (95% CI)	P Value
Constant factor	0.289 (0.183–0.456)	<0.001
History of brucellosis	3.74 (2.818–4.963)	<0.001

Table 7. Distribution of Risk Factors Associated with Wright Test Positivity

Variables	Subgroups	Positive Wright Test		Total (%)	P Value
		Yes (%)	No (%)		
Contact with livestock	Yes (%)	69 (8.8)	716 (91.2)	785 (100)	0.002
	No (%)	87 (5.5)	1492 (94.5)	1579 (100)	
Type of contact with livestock	Contact with aborted fetus	24 (13.6)	152 (86.42)	176 (100)	$\chi^2 = 18.8288$ P = 0.009
	Contact with live animals	53 (8.5)	572 (91.5)	625 (100)	
	Contact with discharge after delivery	24 (15)	136 (85)	160 (100)	
	Keeping livestock	49 (8.8)	511 (91.3)	560 (100)	
	Slaughter of livestock	9 (12.3)	64 (88.7)	73 (100)	
	Contact with meat and organs	20 (13.9)	124 (86.1)	144 (100)	
	Contact with carcasses and livestock secretions after slaughter	9 (12)	66 (88)	75 (100)	
	Contact with aborted fetus	14 (10.8)	116 (89.2)	130 (100)	
	Contact with live animals	104 (10.4)	1741 (89.6)	1943 (100)	
Livestock species	Sheep and goat	88 (10.1)	784 (89.9)	206 (100)	P = 0.094
	Cow	13 (6.3)	193 (93.7)	188 (100)	
	Total	101 (9.4)	977 (90.6)	1078 (100)	
Use of masks and gloves in contact with the livestock	Yes (%)	45 (32.8)	92 (67.2)	137 (100)	P = 0.202
	No (%)	147 (22.9)	494 (77.1)	641 (100)	
Consumption of unpasteurized dairy products	Yes (%)	439 (21.1)	1643(78.9)	2082 (100)	P = 0.790
	No (%)	64 (23.7)	209(76.3)	274 (100)	
Consumption of different types of dairy products	Milk	134 (6.8)	1843 (93.25)	1977 (100)	$\chi^2 = 12.2977$ P = 0.091
	Skim milk	22 (6.7)	304 (93.3)	326 (100)	
	Cheese	42 (6.5)	602 (93.5)	644 (100)	
	Butter	14 (5.9)	223 (94.1)	237 (100)	
	Cream	13 (6.5)	187 (93.5)	200 (100)	
	Curd	38 (10.4)	328 (89.6)	366 (100)	
	Colostrum	1 (2.1)	46 (97.9)	47 (100)	
	Traditional ice cream	6 (3.5)	164 (96.5)	170 (100)	
	Total	270 (6.8)	3699 (93.2)	3969 (100)	
Occupation	Rancher	21 (12.3)	150 (87.7)	171 (100)	$\chi^2 = 21.6169$ P < 0.001
	Farmer	30 (11.6)	228 (88.4)	258 (100)	
	Housewife	78 (7.8)	928 (92.2)	1006 (100)	
	Other	44 (.05)	841 (95.0)	885 (100)	
	Total	173 (7.5)	2149 (92.5)	2322 (100)	

in men compared to women. The sample size in their study was small and, in addition, people with symptoms suspected of brucellosis had been referred, whereas our study was based on the general population of Famenin.[15] In a study by Hashemi et al, 77% of patients were living in villages.[18] Also, in a study by Hajia et al, the participants were aged 15 to 110 years and 61.2% were men and 38.8% were women.[19] In a study by Keramat et al, 59.9% were men, the mean age was 40.69 years, and 64% of the patients lived in villages.[20] In another study by Hajia and colleagues at Sina Hospital in Hamadan, 67.9% of brucellosis patients lived in rural and 32.1% lived in urban areas.[21] Also, in a study by Eini and colleagues between 2005 and 2010 in Hamadan, 43.5% of the subjects were men with a mean age of 40.84 years and 72.2% of the patients lived in villages.[22] In various studies,[18-22] the mean age and mean rate of disease were higher in men than women and most patients lived in villages, which is similar

to the results of this study.

However, in a study by Ranjbar and colleagues on 228 brucellosis patients in Hamadan between 1999 and 2001, 107 (48.6%) of patients were men, the average age of patients was 36.35 years and 50% were rural residents.[23] Another study by Hashemi and colleagues in Hamadan on 219 brucellosis patients between 2008 and 2010 showed that 48.4% (106) of the patients were men and 113 (51.6%) were women with an average age of 39.67 years, of whom 73 (33.3%) were rural residents.[24] In recent studies, brucellosis cases were reported more frequently in women than men, and cases who lived in urban areas were more than or equal to cases in rural areas.[23,24] This is in contrast to our study, and this difference may be due to the fact that analysis was applied only on patients who were admitted to the hospitals, while our study was in the general population; also another important reason may be the transformation of rural areas to urban areas based on

changing country divisions over time.

One of the reasons for the spread of brucellosis in our country is the poorer hygiene among some villagers and farmers; also, agriculture and animal husbandry are very mixed with the rural and nomadic lives of our people. Still, the tent of livestock is set up next to the room and tent of their owners and as soon as new domestic animals are born, young children embrace and stroke them. In some regions of Iran, congestion of plants, high rainfall, lack of sunlight, as well as unhealthy practices in process of providing milk and meat cause the spread of brucellosis. Another common cause of brucellosis in Iran is the consumption of unpasteurized milk from animals infected by *Brucella*, especially colostrum or concentrated milk, which is consumed raw immediately after delivery as a complete meal. Unpasteurized dairy products are still extensively bought in villages and even in cities, and this is another factor in the spread of this disease even among educated people in the community. In our study, we observed a significant relationship between positive brucellosis serology and contact with livestock, use of masks and gloves in contact with the livestock, animal husbandry, farming and previous infection of brucellosis.

In a study by Hasanjani Roshan et al, 55.9% of the patients were male, 75.8% were from rural environments and 45.9% had a history of unpasteurized dairy consumption. There was a significant association between the incidence of brucellosis and occupation and consumption of dairy products as two important risk factors.[25] In our study, there was no significant relationship between unpasteurized dairy consumption and brucellosis, which might be due to the fact that our study was performed on the general population in Famenin. Also, in a study by Eini et al, 39.7% had contact with livestock and 49.6% consumed unpasteurized dairy.[22] These studies indicate that unpasteurized dairy consumption and exposure to livestock are among the major risk factors for brucellosis.

The risk of brucellosis transmission can be reduced by practicing hygiene, avoiding contact with the tissues of affected animals, or avoiding non-pasteurized milk. During abortion and shortly after that, by releasing highly infectious uterine secretions, livestock with brucellosis could contaminate the environment, grounds and pastures and can spread the infection to other animals and humans. Also respiratory transmission to humans has been reported through contaminated suspended particles in air and dust of stalls.[6] In our study, we observed a significant relationship between contact with livestock and the occurrence of brucellosis. Also, people who deal with animals are more likely to acquire the brucellosis infection.[6,26] In this study, there was a significant relationship between high risk jobs (animal husbandry, agriculture) and positive brucellosis serology. Subjects with positive Wright serology had significantly more frequent contact with livestock, but there was no significant relationship between positive

brucellosis serology and dairy consumption. In a study by Nematollahi et al, the rate of brucellosis recurrence was higher in females, as well as in subjects older than 50 years, and those with a history of consuming unpasteurized dairy products, but not in those people who did not have history of contact with animals.[4]

One of the main strengths of this study was that it is the first study of its kind to enroll a large sample size of participants in urban and rural areas in Iran. Second, we have conducted this large seroprevalence study using survey approach in the general population, in contrast to other studies which were done on patients. Accordingly, our study provides external validity and generalizability of seroprevalence findings to general population for the first time. However, the weaknesses of the study were non-registration of clinical manifestations and the examination of participants with signs and symptoms of brucellosis. Therefore, in Famenin population, the actual rate of people with brucellosis should be higher than the reported annual incidence. Yet, we know that the best way to finally win the battle against human brucellosis is control and subsequent eradication of animal brucellosis. In near future, we are going to extend our study to the seroprevalence of brucellosis among livestock in Famenin.

In conclusion, based on the results of this study, the seroprevalence of brucellosis in the population of Famenin was 6.6% based on positive Wright test and 2.4% based on positive 2ME test. The chance of being positive for the Wright test was 3.74 times higher in people with a history of brucellosis than those who did not have any history of disease. Contact with livestock, livestock husbandry, farming, and rural residence were risk factors for higher possibility of brucellosis. The results of our study will be used as basic data for the Famenin Brucellosis Cohort Study, which is needed for preventive interventions and measures against brucellosis in Famenin.

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