

Selected epidemiological features of human brucellosis in Yazd, Islamic Republic of Iran: 1993–1998

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ملاحم وبائية منتقاة لداء البروسيلات البشرية في يزد، جمهورية إيران الإسلامية: 1993–1998

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الخلاصة: يظلُّ داء البروسيلات من المشكلات الصحية الهامة في البلدان التي تكون فيها مكافحة الأمراض الحيوانية المصدر غير كافية. فخلال الفترة بين 1993–1998 تم تحليل المصل وإجراء الزرع من 792 من المرضى الذين يشتبه في إصابتهم بداء البروسيلات ولديهم سوابق من الحمى والنوافض والتعرق الليلي بالإضافة إلى ضعف ووعكة وصداع ممن راجعوا مستشفى للإحالة في يزد في جمهورية إيران الإسلامية. وقد استُقصيت الحالات باختبار التراص في الأنبوب واختبار 2-مركابتوإيثانول واستُكملت استمارة لكل حالة. وقد وصل عيار اختبار التراص في الأنبوب إلى ما يساوي أو يزيد على 160:1 لدى 745 مريضاً (94.1٪) في حين كان اختبار المركابتوإيثانول إيجابياً لدى 42 حالة (5.3٪). ومن بين الحالات الـ 745 المثبتة كان هناك 460 حالة تعود للفترة 1996–1997، وكان أعلى معدل للانتشار في الصيف 39.5٪، وكان أكثر شيوعاً لدى الذكور من الإناث. وكان أعلى معدل للانتشار بين من تتراوح أعمارهم 10–19 عاماً (27.7٪) وكان لدى معظم المرضى سوابق تناول جبن أو لبن أو مشتقات لبن حاملة للعدوى (98٪).

ABSTRACT Brucellosis is a significant health problem in countries where control of zoonoses is inadequate. During 1993–98, we analysed sera and cultures from 792 suspected brucellosis patients who presented with histories of fever, chills, night sweating, weakness, malaise and headache to the referral hospital in Yazd. Cases were investigated by tube agglutination test (TAT) and 2-mercaptoethanol test (2-MET) and a questionnaire was completed for each. TAT titre was $\geq 1:160$ for 745 patients (94.1%) and 2-MET was positive for 42 (5.3%). Of 745 confirmed cases, 460 were from 1996–1997. Prevalence was highest in summer (39.5%) and more common males than among females. Prevalence was highest among those aged 10–19 years (27.7%). Most patients had a history of infected cheese, milk and milk product consumption (98%).

Quelques caractéristiques épidémiologiques de la brucellose humaine à Yazd (République islamique d'Iran) : 1993-1998

RÉSUMÉ La brucellose demeure un important problème de santé dans les pays où les mesures de lutte contre les zoonoses sont insuffisantes. Pendant la période 1993-1998, nous avons analysé les sérums et les cultures de 792 patients suspects de brucellose qui se sont présentés à l'hôpital de recours à Yazd avec des antécédents de fièvre, de frissons, de sueurs nocturnes, de faiblesse, de malaises et de céphalées. Les cas ont été examinés en réalisant l'épreuve d'agglutination en tube et l'épreuve d'agglutination en présence de mercapto-2 éthanol ; un questionnaire a été rempli pour chacun des patients. Le titre pour l'épreuve d'agglutination en tube était supérieur ou égal à 1:160 pour 745 patients (94,1 %) et l'examen au mercaptoéthanol était positif pour 42 patients (5,3 %). Sur les 745 cas confirmés, 460 remontaient à 1996-1997. La prévalence était la plus élevée pendant l'été (39,5 %) et la fréquence était plus importante chez les hommes que chez les femmes. La prévalence était plus forte chez les sujets âgés de 10 à 19 ans (27,7 %). La plupart des patients avaient consommé des fromages, du lait et des produits laitiers infectés (98 %).

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Introduction

Brucellosis is a major health and economic problem in many areas of the world [1,2]. It is mainly a contagious disease of domestic animals such as sheep, goats, cows, camels and dogs. Humans are commonly infected through ingestion of raw milk, cheese and meat or through direct contact with infected animals [2–6]. The disease is transmitted from animals to humans by three routes: direct contact of infected tissues, blood or lymph with conjunctive or broken skin; ingestion of contaminated meat or dairy products; and inhalation of infectious aerosols. Brucellosis occurs on all continents and affects about 500 000 individuals annually worldwide [6,7].

Diagnosis of human brucellosis relies on serological tests, such as the tube agglutination test (TAT), Coombs test, and enzyme-linked immunosorbent assay (ELISA). A single titre of 1:160 or greater for TAT is considered significant. In a modification of assay, the use of 2-mercaptoethanol (2-MET) in the assay disulfide bonds in IgM allow measurement of only IgG. IgG antibody typically appears within weeks of infection and, in the absence of infection, usually persists. After cure, IgG may be present for as long as 1 year. In the chronic stage of the disease, *Brucella* spp. antibodies persist for many years after infection; in most cases only 2-MET sensitive agglutinins persist [8–11].

This study had 2 main aims: first, to obtain data about epidemiological features of human brucellosis in Yazd, Islamic Republic of Iran; second, to describe the characteristics and exposure to risk factors for brucellosis among cases.

Methods

The city of Yazd in central Islamic Republic of Iran has an estimated population of ap-

proximately 388 171 (201 194 males and 186 977 females). During the study period (1993–1998), all untreated suspected brucellosis patients (792 cases) with history of fever, chills, night sweating, weakness, malaise and headache were referred to Nicopour hospital, the Yazd referral hospital, by Yazd health centres and physicians. A questionnaire was used to collect information about patients and also exposure to risk factors, and a blood sample was collected from each case.

For isolation and identification of *Brucella* spp., biphasic blood culture medium (Hemolin, Biomerieux, France), incubated in an atmosphere of 5%–10% carbon dioxide for 30 days, was used [12].

Serum specimens were analysed in 2 phases, using suspension of *B. abortus* and *B. melitensis* (Wellcome Laboratories, UK). In the first phase, all specimens were analysed by the TAT. A titre of 1:160 or greater was taken as an index of seropositivity [8,10]. In the second phase, *Brucella* spp. antibody of patients (IgG) was investigated by 2-MET. The serum dilutions were prepared in 0.85% NaCl containing 0.05 mol/L MET. Then agglutination reactions was read after 48 h incubation at 37 °C [11,13].

The collected data and the results of laboratory tests were analysed by SPSS, version 6 and chi-squared test to determine variables that were significantly associated with seropositivity to *Brucella* spp.

Results

Brucellosis was more common among men than among women (Table 1). There were no significant differences between sexes ($\chi^2_1 = 1.7, P < 0.1$). The average annual rate of human brucellosis (TAT positive) was 124 cases (Table 2). The highest average annual rate of brucellosis was among age

Table 1 Frequency distribution of patients with brucellosis by sex

Sex	1993		1994		1995		1996		1997		1998		Total		Average annual rate	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Male	69	62.2	24	51.1	38	56.7	124	55.9	137	57.6	37	61.7	429	57.6	71	57.3
Female	42	37.8	23	48.9	29	43.2	98	44.1	101	42.4	23	38.3	316	42.4	53	42.7
Total	111		47		67		222		238		60		745		124	

Table 2 Frequency distribution of patients with brucellosis by age group

Age group (years)	1993		1994		1995		1996		1997		1998		Total		Average annual rate	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
<10	15	13.5	6	12.8	2	3.0	24	10.8	20	8.4	9	15.0	76	10.2	13	10.5
10-19	31	27.9	12	25.5	22	32.8	77	34.7	52	21.8	12	20.0	206	27.7	34	27.4
20-29	21	18.9	9	19.1	14	20.9	38	17.1	47	19.7	9	15.0	138	18.5	23	18.5
30-39	21	18.9	8	17.0	8	11.9	32	14.4	37	15.5	16	26.7	122	16.4	20	16.1
40-49	10	9.0	5	10.6	7	10.4	19	8.6	42	17.6	6	10.0	89	11.9	15	12.1
>50	13	11.7	7	14.9	14	20.9	32	14.4	40	16.8	8	13.3	114	15.3	19	15.3
Total	111		47		67		222		238		60		745		124	

group 10–19 years (27.4%) and the lowest was among those under 10 years (10.5%). Results of statistical analysis showed significant differences between age groups of patients ($\chi^2=13.28$, $P < 0.01$).

The number of TAT positive cases (antibody titre $\geq 1/160$) and TAT negative cases (antibody titre $< 1/160$) were 745 (429 males and 316 females) and 47 respectively (Table 3). Of the total TAT positive cases, 42 were 2-MET positive. The average annual number of individuals with a titre $\geq 1:160$ was 124 cases (Table 3).

Consumption of cheese, milk and milk products was reported by 730 brucellosis patients (98%) and unknown risk factors or contact with animal were reported by 15 cases (2%) (Figure 1). The average annual rate of brucellosis was lowest in winter (16.1%) and peaked in summer (39.5%) (Table 4).

The most common presenting symptoms and physical findings with active brucellosis were fever (89%), chills (63%), weakness and malaise (57%), sweating (61%), headache (51%), backache (47%), lymphadenopathy (19%), splenomegaly (28%) and arthritis (18%). Mild anaemia, leucopenia and relative lymphocytosis were common. This information was collected from questionnaires, medical records and clinical examinations. The percentage of patients with brucellosis on the basis of their occupation was 9% farmer, 25.5% housewife, 10.1% worker, 6.8% employee, 30.5% student and 18.1% others.

Discussion

Brucellosis is diagnosed by culture method, serological tests and clinical findings. In the presence of appropriate signs and symptoms, a presumptive diagnosis of

Table 3 Frequency distribution of patients with brucellosis by *Brucella* spp. antibody titre

TAT titre	1993		1994		1995		1996		1997		1998		Total		Average annual rate	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
1/160	24	21.6	7	14.9	16	23.9	46	20.7	53	22.3	16	26.7	162	21.7	27	21.8
1/320	34	30.6	14	29.8	19	28.4	56	25.2	66	27.7	23	38.3	212	28.5	35	28.2
1/640	37	33.3	18	38.3	17	25.4	67	30.1	55	23.1	9	15.0	203	27.2	34	27.4
1/1280	15	13.5	7	14.9	13	19.4	50	22.5	54	22.7	11	18.3	150	20.1	25	20.2
1/2560	1	0.9	1	2.1	2	3.0	3	1.4	10	4.2	1	1.7	18	2.4	3	2.4
Total	111		47		67		222		238		60		745		124	

TAT = tube agglutination test.

brucellosis is usually defined serologically as a TAT of 1:160 or greater [8,13]. Of the 745 TAT positive cases, 42 cases were 2-MET positive.

Brucellosis has been brought under control in the industrialized countries through rigorous diagnostic and control procedures at the animal production level, and through elimination of *Brucella* spp. in livestock and proper pasteurization of milk. Therefore, there are very few reports of indigenously acquired human cases of brucellosis; nonetheless acute imported human infections still occur, generally linked to the consumption of unpasteurized cheese or milk [14-17]. Data from developing countries of the Mediterranean, particularly the Middle East, report prevalence ranging from 8% in Jordan [18], to 12% in Lebanon and Kuwait [19,20]. Even higher seroprevalence rates have been reported in sub-Saharan countries, such as 18% in

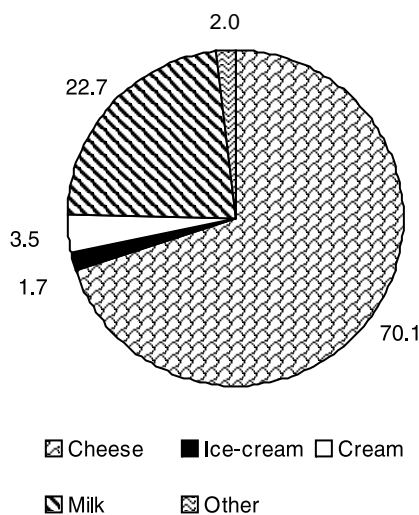


Figure 1 Distribution of patients with brucellosis by self-reported cheese, milk and milk product consumption and other risk factors (%)

Table 4 Frequency distribution of patients with brucellosis by season of the year

Season	1993		1994		1995		1996		1997		1998		Total		Average annual rate	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Spring	32	28.8	8	17.0	14	20.9	15	6.8	66	27.7	18	30.0	153	20.5	25	20.2
Summer	34	30.6	20	42.6	22	32.8	87	39.2	103	43.3	25	41.7	291	39.1	49	39.5
Autumn	32	28.8	10	21.3	23	34.3	55	24.8	52	21.8	8	13.3	180	24.2	30	24.2
Winter	13	11.7	9	19.1	8	11.9	65	29.3	17	7.1	9	15.0	121	16.2	20	16.1
Total	111		47		67		222		238		60		745		124	

Uganda [21] and 13% in Nigeria [22]. According to the results of this study most patients had a history of infected cheese and milk consumption. Prevalence of brucellosis in the Yazd population was much lower than in the above-mentioned reports (31.9 cases per 100 000 population every year; 35.3/100 000 males and 28.3/100 000 females). The prevalence of brucellosis increases with age; this has been observed in the Islamic Republic of Iran, Jordan, Lebanon, and Kuwait [18,19,23–25]. Our findings show that the highest prevalence of the disease was in 1997, particularly among ages 10–19 years (21.8%). Consumption of infected milk, milk products and contact with imported animals with brucellosis were the most important sources of infection.

In an analysis of 104 cases of brucellosis in Saudi Arabia and 1288 cases in United States, the most common symptoms and physical findings reported were fever, chills, weakness, malaise, sweating, backache, headache, lymphadenopathy, splenomegaly and arthritis [26,27]. These results are comparable to those for our subjects.

The prevalence of brucellosis was least in winter (16.2%) and peaked in summer

(39.5%) in our study. In Kuwait and some other countries, however, most cases occurred during the spring and early summer [24,28].

Control of brucellosis requires elimination of infected animals and vaccination of healthy ones in order to reduce the risk for those in regular contact with animals and to have brucellosis-free animal products. Human brucellosis acquired from milk is preventable, and requires making pasteurization of milk and dairy products obligatory. Nevertheless, public health education is important in preventing the transmission of brucellosis from animals to humans.

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