Original Article

RELAPSED HUMAN BRUCELLOSIS AND RELATED RISK FACTORS

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ABSTRACT

Objectives: To determine risk factors of relapse among outpatient treated brucellosis.

Methodology: It is a Cohort study carried out from 2004 to 2006, in an Infectious Diseases Clinic in Ahvaz, Iran. We studied 115 patients with brucellosis. The diagnostic criteria were the finding of ≥1/80 (Wright) with a 2 mercaptoethanol (2 ME) ≥1/40, in association with compatible clinical findings (back pain, sweating and fever). Treatment by standard drug regimen against brucellosis was established according to National Program against Brucellosis (NPB) in Iran. The patients were evaluated at the end of treatment and after two, four and six months by clinical and serological examinations. Patients with relapse and patients without relapse were placed separately in two groups. The data in the two groups were statistically compared with SPSS, 11.5 by chi square test.

Results: Of the 115 patients, 12 were excluded because of study's exclusion criteria. Of 93 studied patients 17 (18.3%) had relapse, results in whom are as fallow: 12 (70.6%) had longer than three months duration of the symptoms prior to diagnosis (P=0.0001). 100% of relapses were in male (P=0.003). Aging was associated with increased risk of relapses (P=0.04). Eleven patients (64.6%) had lymphopenia (P<0.0001). Increased in ESR and CRP values were observed in 16(94.1%) and 17(100%), respectively (P<0.0001).No significant difference in antimicrobial regimen or high risk occupation was observed between two groups (P>0.05).

Conclusion: The present study showed that aging, gender, chronic infection and lymphopenia are risk factors for relapsing brucellosis.

KEY WORDS: Brucellosis, Relapse, Risk factors.

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INTRODUCTION

Brucellosis affects human populations in many developing countries including the Middle East and Latin America where it is still endemic.[1] It has been prevalent in Iran for years.[2] Patients suffering from this disease show unspecific signs and symptoms, most common of them are fever, night sweating and arthralgia.[3] A definite diagnosis requires the isolation of Brucellae from blood, bone marrow or other tissues.[4] However, cultural examinations are time-consuming, hazardous and not sensitive. Thus, clinicians often rely on the indirect proof of infection.[3] A variety of serological tests has been applied, but at least two serological tests have to be combined to confirm active infection. Usually, the standard tube agglutination (STA) test (Wright) is used first and 2mercaptoetanole (2ME) test will confirm its results (with 97.1% sensitivity and 100% specificity).[3,5]

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The primary goals of therapy for brucellosis are to improve the symptoms, reduce complications and prevent relapses.[4] The combination therapies recommended by the World Health Organization for treatment of brucellosis are doxycycline plus rifampicin or doxycycline plus streptomycin.[4,6] Although highly successful results have been obtained with these two regimens, therapeutic failure, relapses and chronic courses are characteristic for the disease.[3] Although risk factors for acquisition of infection such as ingestion of fresh cheese, animal skin contact and ingestion of undercooked meat or raw milk are described in previous reports.[1,2,4,8,9] but risk factors for relapse or treatment failure are not yet clearly understood. Relapses of brucellosis is belived depended on various factors: (i) differences in Brucella species. (ii) deppressed cell mediated immunity (CMI). (iii) locolized infection. (iv) long duration of time between appearance of symptoms and initiation of treatment.[4] Brucella melitensis with high tendency to produce tissue destruction and localized infection resulting in chronic infection and relapse is the most common isolated Brucella from the infected animals and human cases in Khuzestan.[2,5,10]

In limited reports antibiotic regimens, incomplete duration of therapy and CMI responses are discussed as possible risk factors for brucellosis relapses.[11-15] Knowing the risk factors of treatment failure and relapse helps us to manage brucellosis patients successfully and prevent severe complications like bone and joint involvement, neurobrucellosis and endocarditis.

The aim of this study was to determine risk factors for brucellosis relapse among outpatients brucellosis attending to infectious disease clinic.

METHODOLOGY

From 2004 to 2006, we studied a cohort of patients with brucellosis in the Infectious Diseases Clinic in Ahvaz, Iran. The diagnostic criteria were the finding of 1/80 or more standard tube agglutination titer (STAT) of antibodies to brucella (Wright) with a 2 mercaptoethanol (2 ME) >1/40, in association with compatible clinical findings such as: back pain, sweating and fever according to National Program against Brucellosis (NPB) in Iran.[5,6] After diagnosis of the brucellosis, patients were treated with doxycycline plus streptomycin or doxycycline plus rifampicin or co-trimoxasole plus rifampicin with a duration of 8 weeks according to NPB.[5,6] Doxycycline in a dose of 100mg twice per day, streptomycin;1g intramuscular per day for 21 days, rifampicin; 900mg per day in a single dose and co-trimoxasole; 960 mg twice per day. The patients were examined at the end of treatment and after two, four and six months, as well as at any intermediate time if relapse was suspected, and at each evaluation, Wright's seroagglutination and the 2ME were performed. Appropriate clinical response was defined by subsiding fever, back pain and sweating. Relapse was said to have occurred when the indicative clinical picture reappeared or the appearance of a new focal form highly suggestive of brucellosis (e.g., peripheral arthritis, sacroiliitis, orchiepididymitis, lymphocytic meningitis, endocarditis, etc.) and reduced titers of Wright and 2 ME after completion of therapy, increased again. Theraputic failure was definded by persisted fever, sweating or back pain at the end of treatment, with persistently high serological titers. For each patient information recorded in a medical chart included: sex, age, residency(rural, urban), duration of symptoms at admission, occupational expose, clinical symptoms (site of involvement e.g. sacroiliac, hip, knee, etc), laboratory results(CBC, ESR, CRP, Wright, 2ME), antibiotics administered and the duration of treatment, and the treatment outcome(complete recovery, failure). Any relapse during the follow-up was also noted.

Inclusion criteria: (i) active brucellosis, (ii) age above 18years.

Exclusion criteria: (i) incomplete treatment, (ii) incomplete follow up, (iii) pregnancy , (iv) febrile illness other than brucellosis, and (v) treatment failure.

At the end we placed patients in two groups.

Group 1 included complete recovered patients. Group 2 included relapsed patients.Varriables in each group compared with another group in spss 11.5 by using t and chi 2 tests. Differences with p value below 0.05 were considered significant.

RESULTS

Of the 115 patients included, 22 were excluded because of treatment failure, febrile illness suspected having other than brucellosis, incomplete treatment course and incomplete follow up. Of 93 studied patients 73 (78.5%) were men and 20(21.5%) were women. The mean age was 36.5 ± 12.4 years with range of 18 to 60 years. About 66.7% were living in urban area and 33.3% were in rural and remote mountain area without injecting equipment facilities. Fifty eight percent had high risk occupational exposure to *Brucellae* seventy six patients (81.7%) were completely recovered and in the other 17 (18.3%) it was a relapse. The duration of the symptoms prior to diagnosis was 9.7±20.4 weeks (range 1 to 102 weeks). In 42 patients (45.2%), the duration of the symptoms was between two weeks and one month; in 36(38.7%), it was between one and three months and in 15 (16.13%), it was longer than three months. Other results are shown in Table-I. There was a significant difference in gender between relapsed group and non relapsed group (p=0.003). No significant difference in residency (rural or urban area) was observed between two groups for relapse (p=0.211). There was also no difference in used standard antimicrobial regimen between relapsed and non relapsed patients (p=0.808). Increased in ESR and CRP values are associated with increased risk of relapse (p<0.0001).

There was no significant difference in high risk occupation between two groups (p=0.198). Decrease in lymphocyte count (lymphopenia) is associated with increased risk of relapse (p<0.0001). Increase in time between beginning of symptoms and initiation of treatment is associated with increased risk of relapse (P<0.0001).

DISCUSSION

In this study majority of patients with relapse had lymphopenia, long duration of time between appearance of symptoms and initiation of treatment, increased values of ESR and CRP reflecting chronic infection and inflamation and increased titers of anti brucella antibodies reflecting chronic intracellular infection. Low lymphocyte count reflects suppressed CMI. Brucella organisms are able to survive and even multiply within cells of mononuclear system, thus explaining the tendency of the disease to relapse.[4] Interleukin -12 produced by lymphocytes have direct effect on stimulating macrophages for destroying brucella.[4,15] So, we believe that lymphopenia(as a marker of suppressed CMI) in our patients may be considered as a risk factor for relapse. We couldn't find similar studies in literature review (PubMed), but related reports about immunity in brucellosis showed that CMI has main role in protection against brucellosis.[13,15]

Haghirizadeh and et al had reported that chronic and relapsed brucellosis are associated with diminished values in interleukin-12.[15] Dizer and et al showed that levamizole (as an immunomedulator) in addition to antibrucella drugs had a good effect on prevention of relapses.[12] These reports support our believes that decreased CMI due to lymphopenia is a risk factor for relapsing brucellosis. In our study relapses occurred in male more frequent than in female. This finding is consistent with Moreta's work.[11] Since, it is very difficult to differentiate reinfection from relapse in the region of study with a high rate of repeating exposure to brucella antigens in men due to their occupational status and behaviors, we think that sex is not a real risk factor. We found that aging is associated with increased risk of relapses in brucellosis. We suppose that aging is not an independent factor, indeed, decreased CMI in elderly may be the main reason for relapses. In the present study there was a long duration of time from appearance of symptoms of brucellosis to initiation of treatment. Majority of relapsed patients had more than two

Table-I: Risk factors for brucellosis relapse in Ahwaz, Iran, 2004-2007

Risk Factors		*Relapsed Group(n=17) N (percent)*	*No relapsed Group(n=76) N (percent)*	*P value*
Age (years):	18-25	0 (0.0)	21(27.6)	
	25-50	12 (70.7)	40(52.7)	
	> 50	5 (29.3)	15(19.7)	0.047 (S)
Sex:	Male	17(100)	55)72.4)	
	Female	0(0.0.)	21(27.6)	0.01 (S)
Occupation:	High Risk	10(58.8)	41(52.5)	
	Low/No Risk	7(41.2)	35(47.5)	0.79 (NS)
Residency:	Urban	10(58.8)	52(68.4)	
	Rural	7(41.2)	24(31.6)	0.57 (NS)
Anti bact. Regimen:	SDR	9(52.9)	41(52.5)	
	RD	5(29.4)	20(27.8)	
	RC	3(17.7)	15(19.7)	0.57 (NS)
Site of infection:	SI joint	13(76.5)	48(63.2)	
	Hip Joint	4(23.5)	14(18.4)	
	Other sites	0(0.0)	14(18.4)	0.15 (NS)
Lymphocyte count:	Normal	3(17.7)	65(85.5)	
	Lymphocytosis	3(17.7)	10(13.2)	
	Lymphopenia	11(64.6)	1(1.3)	0.0001 (S)
ESR:	Normal	1(5.9)	68(89.5)	
	>20	16(94.1)	8(10.5)	0.0001 (S)
CRP:	Negative	0(0.0)	12(15.8)	
	1+	0(0.0)	26(34.2)	
	2+	4(23.5)	33(43.4)	
	3+	13(76.5)	5(6.6)	0.0001 (S)
Wright:	1/80-1/320	10(58.8)	67(88.2)	
	>1/320	7(41.2)	9(11.8)	0.015 (S)
2ME:	1/20-1/160	13(76.5)	70(92.1)	
	>1/160	4(23.5)	6(7.9)	0.15 (NS)
Missed timefor treatment:	<1 month	0(0.0)	42(55.3)	
	1-2 months	5(29.3)	31(40.8)	
	>2 months	12(70.6)	3(3.9)	0.0001 (S)

Abbreviation: SDR; streptomycin+doxycycline+rifampicin, RD; rifampicin+doxycyclin, RC; rifampicin+ cotrimoxasole, SI; sacroiliac, ESR; erythrocyte sedimentation rate, CRP; Creative protein, 2ME; 2mercapto ethanol

months delay in treatment. This finding is different from the work of Moreta, et al (70% vs.16.6%).[11] This difference may be due to lack of laboratory facilities and medical services in the region of study especially in high risk population such as nomads and shepherds living in the resource limited area of Iran.

Limitations of the study: There are certain limitations in our study such as:

* Although, prospective study has strengths but, according to wide distribution of our patients and long duration of study, difficulties in follow up and compliance of patients may decrease accuracy of the work
* Differentiating real cure from treatment failure is difficult. There is no definite diagnostic criteria for this purpose.
* Conventional serological tests (Wright and 2ME) for detection of relapse are not sensitive tests in contrast to culture and PCR but, these two test were not available in area of study, so the rate of relapses may be over diagnosed.

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Conflict of interest: There is no Conflict of interest.

Ethical Consideration: This work has been approved by the ethical committees of Jondishapour University of Medical Science and the subjects were appropriately informed about the work

REFERENCES

1. Nimri LF. Diagnosis of recent and relapsed cases of human brucellosis by PCR assay. BMC Infect Dis 2003;28;3-5.
2. Iranian Disease Management Center. Information and Database of Communicable Disease in Iran (1977-2002).Tehran, Ministry of Health, 2004.
3. Al Dahouk S, Tomas H, Nockler K, Neubauer H, Frangoulidis D. Laboratory-based diagnosis of brucellosis-a review of the literature. Part II: serological tests for brucellosis. Clin Lab 2003;49(11-12):577-89.
4. Young EJ. Brucella species. In: Mandel GI, Bonnet JE, Dolin R. Principle and practice of infectious disease. 6th Ed. New York. Churchill Livingstone 2005;2669-74.
5. Ministry of health, CDC. National Programme against Brucellosis in Iran, 2002.
6. Tabatabaei SM, Zahraei M, Ahmadnia H, Ghotbi M, Rahimi F. Principles of Disease Prevention and Surveillance. 2nd Ed, Tehran, Roohghalam, 2006.
7. Solera J. Treatment of human brucellosis. J Med Liban 2000;48(4):255-63.
8. Alavi SM, Rafiei A, Nikkhoi AR. Seroepidemiological survey among nomads in Khuzestan, Iran. Iranian J Infect Dis Trop 2006;33;345-50.
9. Alavi SM, Rafiei A, Nikkhoi AR. The effect of lifestyle on brucellosis among nomads in Iran. Pak J Med Sci 2007;23(3):358-60.
10. Khuzestan Health Center. Annual Report on Health Activities. JUMS 2000;159-63.
11. Morata P, Queipo-Ortuño MI, Reguera JM, Garcia Ordonez MA, Pichardo C, Colmenero JD. Post treatment follow-Up of brucellosis by PCR assay. J Clin Microbiol 1999;37(12):4163-6.
12. Dizer U, Hayat L, Beker CM, Görenek L, Ozgüven V, Pahsa A. The effect of the doxycycline-rifampicin and levamisole combination on lymphocyte subgroups and functions of phagocyte cells in patients with chronic brucellosis. Chemotherapy 2005;51(1):27-31. Epub 2005;17.
13. Boura P, Skendros P, Kountouras J, Zacharioudaki E, Tsapas T. Effect of bacterial extracts on the immunologic profile in chronic relapsing brucellosis patients. Int J Immunopathol Pharmacol 1999;12(2):103-11.
14. Skendros P, Boura P, Chrisagis D, Raptopoulou-Gigi M. Diminished percentage of CD4+ T-lymphocytes expressing interleukine-2 receptor alpha in chronic brucellosis. J Infect 2007;54(2):192-7. Epub 2006 May 23.
15. Haghirizadeh Roudan M, Eslami MB, Alavi SM, Pezeshki M. A study on cytokines production in patients with brucellosis. Sci Med J AJUMS 2002;34:20-4.

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