Epidemiological, Risk Factors, Clinical, and Laboratory Features of Brucellosis in the Southwest of Iran within 2009–2015

Mahmood Nabavi, Hossein Hatami1, Hedayatollah Jamaliarand1
Department of Medicine, Faculty of Medicine, Shahid Beheshti University of Medical Sciences, Tehran, Iran.
1Department of Public Health, School of Public Health, Shahid Beheshti University of Medical Sciences, Tehran, Iran.

Abstract
Background: Brucellosis is a serious disease affecting many individuals in the world and in Iran. The aim of this study was to investigate the epidemiological and clinical features of patients diagnosed with brucellosis in Kohgilouyeh and Boyerahmad province, southwest of Iran.
Methods: This retrospective descriptive-analytic study included all individuals diagnosed with brucellosis during 2009–2015. The clinical and epidemiological information were gathered from recorded data available in the health center of Kohgilouyeh and Boyerahmad province, the southwest of Iran.
Results: The total number of patients diagnosed with brucellosis during the study period was 658, of whom, 339 (51.5%) were males. A total of 541 (82.2%) patients resided in rural and tribal areas, and the others lived in urban regions. The mean age of diagnosis was 39.59 ± 17.28 years and the most prevalent age groups were 31–50 (277, 42.1%) and 11–30 (178, 27.1%)-year olds. The most affected groups were housekeeper women (229, 34.8%) and ranchers (152, 23.1%). The relationship between jobs and disease was significant (P < 0.001). Transmission through either suspected dairy products (582, 88.4%) or close contact with infected livestock (537, 81.6%) comprised the most common routes of brucellosis dissemination. The clinical presentation was acute in the majority (581, 88.3%) of the patients. The highest titer for both wright and Coombs Wright tests was 1:320. The most frequently administrated drugs were doxycycline and streptomycin (183, 27.8%). The most commonly observed clinical symptom was bone pain (477, 72.5%). Arthritis (12, 1.8%) comprised the most frequent drug-associated complication. Either relapse or treatment failure was recorded collectively in four (0.6%) patients.
Conclusions: Due to the prevalence of the disease in the nomadic areas, timely detection and control of the disease is essential. Furthermore, livestock vaccination along with educating farmers and physicians about brucellosis can be helpful.

Keywords: Brucellosis, clinical, epidemiology, Iran, risk factors

Introduction
Brucellosis is a common infectious disease affecting both humans and animals. The disease may represent with fever, fatigue, sweating, arthritis, and enlargement of the liver and spleen.[1] Nevertheless, the disease has a wide range of clinical manifestations including musculoskeletal, digestive, urogenital, hematological, cardiovascular, respiratory, and central nervous system symptoms.[2] Brucellosis is a major concern for both health and financial facilities in many parts of the world including the Mediterranean, Middle East, as well as countries surrounding Persian Gulf.[3] Accordingly, brucellosis is also one of the most common infectious diseases in many parts of Iran.[4]

According to the data reported by the Iran Ministry of Health [Figures 1 and 2], brucellosis incidence has increased from 12,248 cases in 2010 to 20,117 in 2014, showing an increment of 70% in this period. The reasons behind this raise are controversial and multifactorial. A major factor may be reduction in domestic production of brucellosis vaccine in the Razi Vaccine and Serum Research Institute. In addition, reduced or halt of vaccine supply from international companies due to political sanctions can also be a potential reason. This notion is supported by declination of brucellosis incidence during 2015–2016 when vaccination was resumed as sufficient products were available.

Vaccination and surveillance of brucellosis among animals are necessary to prevent the transmission of the disease to humans.[5,4] In addition to this, appropriate and accurate epidemiological and clinical data are required to implement effective preventive measures.
and therapeutic policies for brucellosis.\[7] Brucellosis has been a serious health-care concern in Iran with multiple reports on its severe complications.\[8-10] Some risk factors of brucellosis in Iranian population have been unprotected animal contact and using nonpasteurized dairy products.\[10] There is incomplete knowledge about the epidemiological and clinical aspects of brucellosis, especially among rural and tribal populations of Iran. Accordingly, we found no previous reports on brucellosis epidemiology in Kohgilouyeh and Boyerahmad province in the southwest of Iran except for one epidemiological report conducted within 2009–2013.\[11] There were significant differences in the geographic distribution of brucellosis, with the incidence rates being highest in most of the cities in the west and north-west of the country.\[12] Here, we aimed to provide a comprehensive view on the epidemiological and clinical features of patients with brucellosis in this area during 2009–2015. This study can be complementary to the previously published data.

Methods

In this retrospective descriptive-analytic study, all the cases of brucellosis whose data were available at the Kohgilouyeh and Boyerahmad (Yasouj) health center were included. The demographic, clinical, and laboratory information of the patients were acquired. Our study was conducted according to the ethical consideration of the Declaration of Helsinki 2000 revision.

Diagnosis of brucellosis

Brucellosis was diagnosed based on the relevant clinical picture (i.e., intermittent or persistent fever, fatigue, anorexia, and weight loss) along with confirmatory laboratory results including the detection of agglutinating antibodies (wright, Coombs wright, and 2-mercaptoethanol [2-ME] tests) and/or a positive blood culture test. The thresholds of the serological tests for being considered as positive were ≥1:80 and ≥1:40 for wrights and 2-ME tests, respectively. Coombs wright test was considered positive if agglutination was observed in three dilutions prior to the observed wright titer (e.g., for a wright titer of 1:80, a Coombs wright of 1:40 would be positive). A blood culture was rendered as a confirmatory test in case of positivity for any brucellosis strains.\[13,14] Based on these criteria, 658 patients were diagnosed with brucellosis during the time course of the study.

Inclusion and exclusion criteria

The patients diagnosed with brucellosis according to the previously mentioned criteria during 2009–2015 who were residents of Kohgilouyeh and Boyerahmad province were included. They all had been registered in our local system of reporting and management of diseases.

Statistical analysis

The data were analyzed using SPSS 19 software. Descriptive statistics (means and frequencies) were used to present the population’s characteristics. The Chi-square and independent sample Student t-test were applied for seeking any relationship or significant differences between different groups. A two-tailed P value of less than 0.05 was considered as statistically significant.

Results

Out of 658 patients diagnosed with brucellosis during 2009–2015, 339 (51.5%) and 319 (48.5%) were males and females, respectively. A total of 541 (82.2%) cases were residents of rural and nomadic areas, whereas 117 (17.8%) patients lived in urban areas. The mean age of brucellosis diagnosis was 39.59 ± 17.28 years which was significantly different between men (40.81 ± 18.33 years) and women (38.30 ± 16.30 years) (P < 0.001, Table 1). The relationship between jobs and disease was significant (P < 0.001). The most prevalent titer for wright test was 1:320 (24%). The results of Coombs wright, 2-ME, and blood culture tests have been presented in Table 2.

The time-lapse from diagnosis to cure was 0–3 months for 581 (88%) cases (corresponding to acute brucellosis). In additions, this period was 3–12 months for 64 (9.7%) patients (representing subacute brucellosis). For 13 (2%) patients, the period exceeded from 1 year, suggesting chronic brucellosis. Due to the variable clinical picture of the disease, final diagnosis was delayed in a number of patients until confirmatory laboratory results received. A family history of brucellosis was recorded in 40 (11.8%) and 38 (11.9%) of male and female patients, respectively (P = 0.36) [Table 3].

There were 45 different combinations of administrated therapeutic regimens. The most commonly prescribed drugs were doxycycline and rifampin [Table 4]. Furthermore, rifampin was the first-line drug in relapsed patients. The majority of the patients (84.6%) did not experience any adverse drug reactions. Others, however, encountered arthritis (9, 1.4%), encephalitis (1, 0.2%), arthritis and spondylitis (2, 0.3%), and other reactions (6, 0.9%).

Figure 1: The incidence rate of brucellosis in Iranians from 1979 to 2015. Horizontal axis denotes the time period in years and the vertical axis shows the incidence rate per 100,000 population (adapted from Iranian Health Ministry report; http://www.health.gov.ir)
The peak incidence of brucellosis was in spring and summer (460, 70%, Figure 3).

**Discussion**

In the present report, the clinical and epidemiological aspects of brucellosis were assessed in Kohgilouyeh and Boyerahmad province, the southwest of Iran during 2009–2015. The general knowledge and public awareness about brucellosis are low among Iranians.\[15\] Brucellosis is a common infectious disease in many regions of the world. The incidence of brucellosis has increased in developing countries during recent years.\[6\] Accordingly, the incidence rate of brucellosis has increased in recent years in Iran with the peak incidence in the northwest and northeast regions.\[12\] In fact, the countries with the highest incidence rate of brucellosis in the Middle East are Saudi Arabia, Iran, Palestine, Syria, Jordan, and Oman.\[16\] Accordingly, we here observed a respectively high prevalence of brucellosis in our province.

Out of 658 patients identified during 2009–2015, males constituted 51.5% of the cases. This ratio was similar to previous studies performed in Iran (49.5–54.2%),\[11,17\] Turkey (55.7%),\[18\] and Saudi Arabia (66.5%).\[19\] On the contrary, women constituted the majority of patients with brucellosis in some other reports (59%).\[20\] The mean age of brucellosis diagnosis was 39.59 ± 17.28 years in our study, which was similar to previous studies in
Table 1: Epidemiological and clinical findings in 658 patients with brucellosis in Kohgilouyeh and Boyerahmad province, Southwest of Iran

| Variables                             | Male (n) | Male (%)   | Female (n) | Female (%) | P     |
|---------------------------------------|----------|------------|------------|------------|-------|
| Age of diagnosis (mean±SD)            | 40.81±18.33 | 38.30±16.30 | <0.001     |            |       |
| Occupation                            |          |            |            |            |       |
| Rancher                               | 152 (23.1%) |            |            |            | <0.001|
| Housekeeper                           | 229 (34.8%) |            |            |            |       |
| Farmer and rancher                    | 95 (14.4%) |            |            |            |       |
| Housewife and rancher                 | 14 (2.1%) |            |            |            |       |
| Student                               | 51 (7.8%) |            |            |            |       |
| Collegian                             | 2 (0.3%) |            |            |            |       |
| Farmer                                | 5 (0.8%) |            |            |            |       |
| Self-employment                       | 6 (0.9%) |            |            |            |       |
| Employee                              | 16 (2.4%) |            |            |            |       |
| Manual worker                         | 17 (2.6%) |            |            |            |       |
| Children                              | 21 (3.2%) |            |            |            |       |
| Other                                 | 50 (7.6%) |            |            |            |       |
| Clinical symptoms                     |          |            |            |            |       |
| Yes Fever                             | 434 (66) |            |            |            | 0.001 |
| No                                    | 224 (34) |            |            |            |       |
| Yes Fatigue and anorexia              | 275 (41.8) |            |            |            | 0.672 |
| No                                    | 383 (58.2) |            |            |            |       |
| Yes Backache                          | 366 (55.6) |            |            |            | 0.383 |
| No                                    | 291 (44.2) |            |            |            |       |
| Yes Musculoskeletal and bone pain     | 477 (72.5) |            |            |            | 0.04  |
| No                                    | 181 (27.5) |            |            |            |       |
| Yes Weight loss                       | 180 (27.4) |            |            |            | 0.761 |
| No                                    | 478 (72.6) |            |            |            |       |
| Yes Depression                        | 5 (0.8) |            |            |            | 0.703 |
| No                                    | 653 (99.2) |            |            |            |       |
| Yes Weakness and lethargy             | 61 (9.3) |            |            |            | 0.878 |
| No                                    | 597 (90.7) |            |            |            |       |
| Yes Adenopathy, hepatosplenomegaly    | 2 (0.3) |            |            |            | 0.966 |
| No                                    | 656 (99.7) |            |            |            |       |
| Local climate                         |          |            |            |            |       |
| Cool                                  | 232 (35.3) |            |            |            | 0.287 |
| Tropical                              | 426 (64.7) |            |            |            |       |

Figure 3: The seasonal incidence of brucellosis in Kohgilouyeh and Boyerahmad province, southwest of Iran

There were also studies in which the mean age of brucellosis diagnosis was higher compared to our study.\(^\text{[2,18,19]}\) In this study, brucellosis was more prevalent in the 31–50-year-old age group. In comparison, the age groups of 15–20\(^\text{[21]}\) and 40–49 years have been noted as the most common age groups affected with brucellosis in precedent reports.\(^\text{[18]}\) Overall, brucellosis is a disease affecting both genders and a wide range of age groups necessitating preventive measures to be implemented throughout the populations.

Similar to a study by Moosazadeh \textit{et al.},\(^\text{[10]}\) we found that brucellosis was more common among housewives, ranchers, and farmers. The disease was also more prevalent in the tropical regions of the province compared to the cold regions. Furthermore, brucellosis incidence was reported in all the seasons; however, it was more prevalent in spring and summer corresponding to livestock breeding and lactation periods. This pattern of distribution has also been described in previous studies in Iran and overseas.\(^\text{[17,21]}\) It is recommended for at-risk individuals to consider appropriate cautious measures when contacting with livestock, especially in breeding and lactation seasons.
The most prevalent clinical manifestations of brucellosis were fever, anorexia, weight loss, low back, and muscle pains. These were also common clinical features reported in a previous study. In addition, anemia has been described as a common clinical finding in patients with brucellosis in precedent studies from Iran and other countries. Due to the high clinical variability of brucellosis, the final diagnosis may be delayed if appropriate and timely confirmatory laboratory evidences are not warranted. In a number of our patients, the final diagnosis and therefore therapeutic measures were delayed due to the late development of clinical symptoms and procrastinating in obtaining confirmatory laboratory tests. Therefore, physicians should be prompted to order appropriate tests in suspicious individuals.

The consumption of unpasteurized dairy products as well as close contact with infected livestock were the most common routes for brucellosis transmission in our study. This was similar to previous reports in Iran and Turkey. A family history of brucellosis was identified in 18.5% of our cases. However, this ratio was higher in other studies reaching as high as 40% in some reports. The most common tests utilized to diagnose brucellosis are serological tests (i.e., Wright, Coombs Wright, and 2-ME). In this study, Wright test retrieved positive results in 24% of the patients with the most prevalent titer as 1:320. Furthermore, the majority of our patients represented 1:320 and 1:80 titers for Coombs Wright and 2-ME tests, respectively. In comparison, positive Wright test was reported in 3.3% of patients with brucellosis in a study performed in Fars province of Iran. In a study in Turkey, 98.8% of 1016 patients with brucellosis showed positive results for the standard tube agglutination Coombs test. In another recent report in Georgia, positive Wright test was found in 41% of patients with brucellosis. In comparison, we observed positive STA Coombs, 2-ME, and Wright tests in 97 (17%), 363 (55.1%), and 597 (90.7%) of the cases.

In this study, 88%, 9.7%, and 2% of the patients achieved cures within 0–3, 3–13, and >12 months, respectively. These ratios were representatives of acute, subacute, and chronic brucellosis, respectively. Compared to this, 53.6% and 21.5% of patients with brucellosis showed acute and chronic brucellosis, respectively. Compared to this, 53.6% and 21.5% of patients with brucellosis showed acute and chronic presentations, respectively, in a study performed in China. The outcomes of therapeutic strategies in brucellosis are variable based on the clinical presentations. The selection of appropriate therapeutic regimes is important for nailing effective management of brucellosis. We here observed that doxycycline–streptomycin regime was more effective than doxycycline–rifampin regarding lower adverse effects (29% vs. 10%, respectively) which is in line with a previous report from Spain.

Although 72% of Iranian populations reside in urban areas, the majority of our patients with brucellosis were residents of rural and tribal regions. This finding was in accordance with the observations of other researchers. A
Table 4: Therapeutic protocols in 658 patients with brucellosis in Kohgilouyeh and Boyerahmad province, Southwest of Iran

| Therapeutic combinations          | Percentage $n=658$ (%) | Duration (weeks) |
|----------------------------------|------------------------|------------------|
| Doxycycline + rifampin           | 183 (27.8)             | 2-6              |
| Doxycycline + streptomycin       | 84 (13.1)              | 1-6              |
| Doxycycline + streptomycin + rifampin | 54 (8.2)          | 2-6              |
| Gentamicin + cotrimoxazole       | 49 (7.4)               | 2-6              |
| Gentamicin                       | 32 (4.9)               | 1-6              |
| Cotrimoxazole + rifampin         | 28 (4.3)               | 2-6              |
| Rifampin                         | 23 (3.5)               | 2-6              |
| Doxycycline + gentamicin + rifampin | 21 (3.2)        | 2-6              |
| Tetracycline + rifampin          | 17 (2.6)               | 2-6              |
| Gentamicin + rifampin            | 16 (2.4)               | 1-4              |
| Streptomycin                     | 12 (1.8)               | 1-3              |
| Streptomycin + rifampin          | 10 (1.5)               | 3-6              |
| Others                           | 129 (19.3)             | 1-8              |

considerable ratio of rural and tribal households does animal husbandry for a living, which can partly explain the higher rates of brucellosis in these populations. Educating these at-risk individuals can be a suitable strategy to manage and prevent brucellosis transmission from livestock to humans.

Conclusions

Implementing managemental policies regarding brucellosis requires a comprehensive epidemiological view on the disease. Regarding the prevalence of the disease among young age groups, timely diagnosis of brucellosis is crucial in this active group of societies. Regarding the severe complications and the complex diagnostic and therapeutic aspects of brucellosis, preventive measures are by far more cost-effective in this condition. In addition to vaccination of livestock, teaching farmers to avoid contacting with infected livestock and not to consume nonpasteurized dairy products can be helpful. It is highly recommended to implement a systematic comprehensive health education and promotion model in order to boost general education about brucellosis.

Acknowledgments

The authors wish to thank all the officials and experts of communicable and zoonotic diseases of health centers in Yasuj city who helped us in carrying out this research. This study was a result of MPH course thesis.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

Received: 19 Jan 18 Accepted: 23 Nov 18 Published: 12 Jun 19

References

1. Yaman Y, Gözmen S, Özkaya AK, Oymak Y, Apa H, Vergin C, et al. Secondary hemophagocytic lymphohistiocytosis in children with brucellosis: Report of three cases. J Infect Dev Ctries 2015;9:1172-6.

2. Okur M, Erbey F, Bektas MS, Kaya A, Doğan M, Acar MN, et al. Retrospective clinical and laboratory evaluation of children with brucellosis. Pediatr Int 2012;54:215-8.

3. El-Koumi MA, Afify M, Al-Zahrani SH. A prospective study of brucellosis in children: Relative frequency of pancytopenia. Mediterr J Hematol Infect Dis 2013;5:e2013011.

4. Hadadi A, Rasoulinejad M, Haji Abdolbaghi M, Mohraz M, Khashayar P. Clinical profile and management of brucellosis in Tehran–Iran. Acta Clin Belg 2009;64:11-5.

5. Krcakil I, Abdullahayev R, Asadov K, Ismayilova R, Baghirova M, Ustun N, et al. Human brucellosis trends: Re-emergence and prospects for control using a one health approach in Azerbaijan (1983–2009). Zoonoses Public Health 2016;63:294-302.

6. Bosilovsky M, J Rodriguez-Morales A. Brucellosis and its particularities in children travelers. Recent Pat Antiinfect Drug Discov 2014;9:164-72.

7. Murtur B, Amarnath S, Shinde R. Review of clinical and laboratory features of human brucellosis. Indian J Med Microbiol 2007;25:188-202.

8. Aghaali M, Mohebi S, Heydari H. Prevalence of asymptomatic brucellosis in children 7 to 12 years old. Interdiscip Perspect Infect Infect Dis 2015;2015:187369.

9. Mahmoodabadi SM, Barkhordari A, Nabizadeh M, Ayatollahi J. The effect of health education on knowledge, attitude and practice (KAP) of high school students’ towards brucellosis in Yazd. World Appl Sci J 2008;5:522-4.

10. Moosazadeh M, Nikaeen R, Abedi G, Kheradmand M, Safiri S. Epidemiological and clinical features of people with Malta fever in Iran: A systematic review and meta-analysis. Osong Public Health Res Perspect 2016;7:157-67.

11. Shahriari S, Ghatee M, Haghdoost A, Tabody Z, Khajeh Kazemi R, Parisae Z, et al. Demographic and Epidemiological Study of Brucellosis in the Kohgilooeye and Boyerahmad Province, 2009-2013. Armaghanne Danesh 2015;20:149-60.

12. Pakzad R, Pakzad I, Safiri S, Shirzadi MR, Mohammadpour M, Behroozi A, et al. Spatiotemporal analysis of brucellosis incidence in Iran from 2011 to 2014 using GIS. Int J Infect Dis 2018;67:129-36.

13. Golshani M, Buozari S. A review of Brucellosis in Iran: Epidemiology, Risk Factors, Diagnosis, Control, and Prevention. Iran Biomed J 2017;21:349-59.

14. Haja M, Rahbar M. Isolation of brucella from blood culture of hospitalized brucellosis patients. Arch Clin Infect Dis 2006;1:59-62.

15. Bayram N, Özkinay F, Onay H, Yilmaz‑Çiftdogan D, Tufan S, Vardar F. Mannose‑binding lectin gene codon 54 polymorphism susceptible to brucellosis in Turkish children. Turk J Pediatr 2012;54:234-38.

16. Refai M. Incidence and control of brucellosis in the Near East region. Vet Microbiol 2002;90:81-110.

17. Roushan MH, Gangi S, Ahmadi S. Comparison of the efficacy of doxycycline plus co-trimoxazole versus doxycycline plus rifampin in brucellosis. Swiss Med Wkly 2004;134:564-8.

18. Al-Tawfiq JA, AbuKhamsin A. A 24-year study of the epidemiology of human brucellosis in a health-care system in
19. Kayaaslan B, Bastug A, Aydin E, Akinci E, But A, Aslaner H, et al. A long-term survey of brucellosis: Is there any marker to predict the complicated cases? Infect Dis 2016;48:215-21.

20. Honarvar B, Moghadami M, Lankarani K, Davarpanah M, Ataolahi M, Farbod A, et al. Brucellosis as a neglected disease in a neglected population: A seroepidemiological study of migratory nomads in the Fars province of Iran. Epidemiol Infect 2017;145:491-7.

21. Dastjerdi MZ, Nobari RF, Ramazanpour J. Epidemiological features of human brucellosis in central Iran, 2006–2011. Public Health 2012;126:1058-62.

22. Solera J, Rodriguez-Zapata M, Geijo P, Largo J, Paulino J, Sáez L, et al. Doxycycline-rifampin versus doxycycline-streptomycin in treatment of human brucellosis due to Brucella melitensis.

23. Buzgan T, Karahocagil MK, Irmak H, Baran AI, Karsen H, Evirgen O, et al. Clinical manifestations and complications in 1028 cases of brucellosis: A retrospective evaluation and review of the literature. Int J Infect Dis 2010;14:e469-78.

24. Akhvlediani T, Bautista CT, Garuchava N, Sanodze L, Kokaia N, Malania L, et al. Epidemiological and clinical features of brucellosis in the country of Georgia. PLoS One 2017;12:e0170376.

25. Jia B, Zhang F, Lu Y, Zhang W, Li J, Zhang Y, et al. The clinical features of 590 patients with brucellosis in Xinjiang, China with the emphasis on the treatment of complications. PLoS Negl Trop Dis 2017;11:e0005577.