A seroprevalence and relationship survey of brucellosis between pregnant women and women with spontaneous abortion in Iran

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Abstract
Background: Brucellosis is one of the most prevalent diseases common between humans and animals. It is also called Malta fever, Undulant fever and Mediterranean fever. This disease is spread by consuming milk and its unpasteurized derivatives. Clinical symptoms of brucellosis in humans are fever, chills, headache, muscular pain, tiredness, loss of appetite, joint pain, weight loss, constipation, sore throat, and dry cough. The present study aimed at surveying the seroprevalence of brucellosis in pregnant women and those women who suffered from spontaneous abortion.

Methods: This case-control study was conducted in Sanandaj (Iran) in 2016 and included 2 groups of pregnant women: one group included 160 pregnant women and the other included 160 women who suffered from spontaneous abortion. Then, the participants were asked to fill out the questionnaire. After receiving permission from an obstetrician, a 10-cc blood sample was taken from each person to be used in the Rose Bengal, Wright, 2ME, and Coombs tests. Independent samples t test and Chi-square test were used to analyze the data and compare the groups.

Results: Mean±SD age of women in the case group was 30.9±7.3 years, while it was 27.74±5.41 years in control women. The Rose Bengal, Wright, and 2ME prevalence for both groups was negative, but the Coombs and Wright tests score was 33 (20.6%) in pregnant women and it was 27 (16.9%) in women who experienced spontaneous abortion. No meaningful relationship was observed between spontaneous abortion and brucellosis (p=0.39).

Conclusion: Even though the present study did not find a meaningful relationship between spontaneous abortion and brucellosis (p=0.39), high brucella seroprevalence rates between both groups of women indicated that screening tests should be considered before gestation as an appropriate therapeutic strategy.

Keywords: Brucellosis, Seroprevalence, Pregnant women, Spontaneous abortion

Introduction
Brucellosis is one of the most prevalent diseases recognized in humans and animals. It is also referred to as Malta fever, Undulant fever, and Mediterranean fever (1). This disease can be transmitted by consumption of raw milk and dairy products (2). Clinical symptoms of brucellosis in humans are fever, chills, headache, muscle pain, fatigue, loss of appetite, joint pain, weight loss, constipation, sore throat, and dry cough (3). Complications of the
gastrointestinal tract from a brucellosis infection are minor such as loss of appetite, nausea, vomiting, abdominal pain, diarrhea, or constipation (4, 5). The liver is the largest member of the reticuloendothelial system and may also be involved in brucellosis (6). In a small number of patients, the symptoms continue for a long time after the fever period has ended. If the presence of the symptoms lasts longer than 1 year, the disease is referred to as chronic brucellosis. Chronic brucellosis is usually caused by persistent infection of the deep tissues such as purulent matter in bones, joints, liver, spleen, or kidneys (7). In a study conducted to determine the prevalence of brucellosis infection among women, employing the brucellosis serological method, brucellosis infection was found in 5.8% of the study sample (8). In another study, the prevalence rate of brucellosis in pregnant women and in women who had abortion was 15.18% and 27.2%, respectively (9).

Considering the high prevalence of brucellosis in Sanandaj (Iran) and a high consumption of raw milk, the present study aimed at surveying the seroprevalence of brucellosis in pregnant women and those women who suffered from spontaneous abortion.

**Methods**

The present case-control study was conducted in Sanandaj, Kurdistan province (Iran) in 2016. This study included 2 groups: the first group consisted of blood samples of 160 women who had spontaneous abortion at the gestational age of 11 to 20 weeks, and the second group included the blood samples of 160 women with normal pregnancy and no signs of abortion at the gestational age of 35 to 37 weeks. The inclusion criteria of this study were lack of antibiotic consumption, consumption of raw milk, and lack of vaginal infection. However, women with diabetes, endocrine disorders, and hypertension as well as those who had an abortion based on trauma or anatomical abnormalities were excluded. Demographic data relevant to their husbands were obtained including age, education, occupation, and no signs of abortion at the gestational age of 35 to 37 weeks. The inclusion criteria of this study were lack of antibiotic consumption, consumption of raw milk, and lack of vaginal infection. However, women with diabetes, endocrine disorders, and hypertension as well as those who had an abortion based on trauma or anatomical abnormalities were excluded. Demographic data relevant to their husbands were obtained including age, education, location, number of childbirth, genital infection, urinary infection, smoking prior and during pregnancy, and urinary tract infection (UTI). At the beginning of the study, the signed consent forms were taken from the participants. Following this step and obtaining permission from an obstetrician, a 10-cc blood sample was taken from each person to be used in the Rose Bengal, Wright, 2ME, and Coombs Tests Kit (Pasteur Institute, Iran). In addition to asking the participants about their first day of most recent menstrual, ultrasound scan tests were performed to estimate the gestation age. To remove the threat of chromosomal abnormalities and likelihood of genetic miscarriage, fetal health assessment tests including nuchal translucency (NT) and double tests such as pregnancy associated plasma protein A (PAPP-A) and free ßHCG were conducted at 11 and 13 weeks of gestation. Furthermore, the confirmatory triple tests were performed at 15 and 17 weeks of gestation to eliminate probable neural tube and chromosomal anomalies. Data were analyzed by Stata 12 software. The One-Sample Kolmogorov-Smirnov test was used to determine the normal age distribution. Results are given as mean ± SD, and independent samples t test was used to compare the mean differences. To compare the qualitative variables, Chi-square and Fisher’s exact tests were used. Significance level was set at p > 0.05.

**Results**

The present study revealed that the result of Rose Bengal test was negative in all samples; moreover, Wright and 2ME prevalence was negative for both groups. However, the prevalence of Abundance Coombs Wright in pregnant women and in women who had spontaneous abortion was 33 (20.6%) and 27 (16.9%), respectively. The age range of women in the case group was 30.9 ± 7.3 years, while it was 27.74 ± 5.41 years in control women. The frequency of vaginal infection in pregnant women was 26 (16.2%),

<table>
<thead>
<tr>
<th>Variables</th>
<th>Cases (Spontaneous abortion)</th>
<th>Controls (Pregnant woman)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean±SD age (year)</td>
<td>n=160</td>
<td>n=160</td>
<td></td>
</tr>
<tr>
<td>&lt;25</td>
<td>(30.9 ±7.3)</td>
<td>(27.74 ± 5.41)</td>
<td>0.3</td>
</tr>
<tr>
<td>25-35</td>
<td>41(25.6%)</td>
<td>43(26.8%)</td>
<td></td>
</tr>
<tr>
<td>&gt;35</td>
<td>74(46.2%)</td>
<td>85(53.1%)</td>
<td></td>
</tr>
<tr>
<td>Education</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Illiterate</td>
<td>8(5%)</td>
<td>10(6.2%)</td>
<td>0.47</td>
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<tr>
<td>Less than high school diploma</td>
<td>129(80.6%)</td>
<td>120(75%)</td>
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<tr>
<td>Academic education</td>
<td>23(14.4%)</td>
<td>30(18.8%)</td>
<td></td>
</tr>
<tr>
<td>Spouse</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Less than high school diploma</td>
<td>125(78.1%)</td>
<td>116(72.5%)</td>
<td>0.37</td>
</tr>
<tr>
<td>Academic education</td>
<td>33(20.6%)</td>
<td>43(26.9%)</td>
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</tr>
<tr>
<td>Occupation</td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>Housekeeper</td>
<td>148(92.5%)</td>
<td>149 (93.1%)</td>
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<tr>
<td>Employee</td>
<td>12 (7.5%)</td>
<td>11(6.9%)</td>
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<tr>
<td>Free</td>
<td>124(77.5%)</td>
<td>95 (59.4%)</td>
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<tr>
<td>Occupation Spouse</td>
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<tr>
<td>Employee</td>
<td>36(22.5%)</td>
<td>65(40.6%)</td>
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<tr>
<td>&lt;=1</td>
<td>58(36.2%)</td>
<td>84(52.5%)</td>
<td></td>
</tr>
<tr>
<td>Number of children</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>&gt;=2</td>
<td>102(63.8%)</td>
<td>76(47.5%)</td>
<td></td>
</tr>
<tr>
<td>Smoking</td>
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<tr>
<td>No</td>
<td>1(0.6%)</td>
<td>1 (0.6%)</td>
<td>0.99</td>
</tr>
<tr>
<td>History of Vaginal infection</td>
<td>28 (17.5%)</td>
<td>26 (16.2%)</td>
<td>0.76</td>
</tr>
<tr>
<td>History of urinary infection</td>
<td>16(10%)</td>
<td>25 (15.6%)</td>
<td>0.13</td>
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<tr>
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<td>5(3.1%)</td>
<td>5 (3.1%)</td>
<td>0.99</td>
</tr>
<tr>
<td>Location</td>
<td></td>
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</tr>
<tr>
<td>City</td>
<td>48(30%)</td>
<td>46(28.8%)</td>
<td>0.80</td>
</tr>
<tr>
<td>Village</td>
<td>112(70%)</td>
<td>114(71.2%)</td>
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<tr>
<td>Coombs Wright</td>
<td>27(16.9%)</td>
<td>33(20.6%)</td>
<td>0.39</td>
</tr>
</tbody>
</table>

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Discussion

Abortion is a preterm birth without a live fetus, and studies have shown that many factors play a role in the incidence of abortion such as fetal and maternal factors (10). For years, researchers have thought that abortion occurs due to microbial factors, and efforts in this regard have been completed and the results have been significant (10, 11). Brucellosis is a bacteria that causes abortion in animals, but its role in humans has not been established because animal placentas contain erythritol, which is a growth factor for brucellosis (12, 13). Brucellosis in humans may lead to financial and physical problems, thus, early diagnosis and proper treatment can prevent it from being chronic (14, 15). In the present study, the obtained results revealed that Rose Bengal test result was negative in all samples, and the prevalence of Wright and 2ME was also negative. The result of Abundance Coombs Wright test was 20.6% in pregnant women and 16.9% in women who had spontaneous abortion. Furthermore, it was found that the prevalence of chronic brucellosis is increasing among women. Although the study revealed no significant relationship between spontaneous abortion and brucellosis, its prevalence rate in women with spontaneous abortions can be verified.

Ali S et al. (2016) found that the prevalence rate of brucellosis to be 5.8% in pregnant women, using the serological methods (8). This study when compared to our study revealed that brucellosis frequency is less common because the participants all consume local dairy. In another study conducted by Elshamy et al. (2008), a case group consisting of 55 pregnant women with brucellosis was compared with a control group of 395 women who had infection. The analysis of the results of the (enzyme-linked immunosorbent assay) ELISA test (antibody titer for brucellosis) revealed that in the case group, 15 patients (27.2%) suffered from abortion, while 60 patients (15.18%) experienced a miscarriage in the control group. Thus, It was concluded that there might be a relationship between brucellosis and abortion (9). This is similar to our study in that no significant relationship was found between spontaneous abortion and brucellosis; however, our study found that the prevalence of brucellosis among women is high.

Hassan-Janny et al. (2011) investigated the rate of prevalence of brucellosis by employing a serology method in 19 pregnant women who were infected by brucellosis. After performing the test, it was revealed that 10 (55%) participants had a spontaneous abortion. Moreover, from the 19 women suffering from brucellosis, who were treated with co-trimoxazole and rifampin, 5 (31%) experienced abortion and 9 (69%) had a normal vaginal delivery. Hence, they concluded that a screening test is vital for pregnant women (14). This study and our study suggest that a brucellosis screening program for pregnant women and for those with spontaneous abortion is necessary in brucellosis endemic regions.

Abo-Shehada et al. (2011) conducted a case-control study with 445 women who had an abortion and 445 women who did not have an abortion. In that study, the incidence of brucellosis was tested employing serology, i.e., the Rose Bengal method; and the results revealed that the rate of prevalence was 1.8% and 1% in the patient and control groups, respectively. Furthermore, no significant relationship was found between brucellosis and abortion. The low prevalence of brucellosis in both groups, compared to our study, might have been due to the type of screening study (15). Some factors may be transmitted in bacteria that cause vaginal infection during pregnancy. Several studies have been conducted on the relationship between vaginal infections and spontaneous abortions (16-18).

Even though the present study did not find a significant relationship between spontaneous abortion and brucellosis (p=0.39), high brucella seroprevalence rates between both groups of women indicate that screening tests should be considered before gestation as an appropriate therapeutic strategy.

Conclusion

This study did not find a significant relationship between spontaneous abortion and brucellosis. However, the high brucella seroprevalence rates between both groups of women indicate that screening tests should be considered before gestation as an appropriate therapeutic strategy.

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Conflict of Interests

The authors declare that they have no competing interests.

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