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Serologic survey for *Coxiella burnetii* phase II antibodies among slaughterhouse workers in Kerman, southeast of Iran

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PEER REVIEW

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Comments

This result suggests the importance of Q fever as an occupational risk for slaughterhouse workers. Q fever has been considered primarily an occupational zoonotic disease for abattoir workers, sheep shearers, livestock farmers, and especially veterinarians because of their direct contact with potentially infected animals.

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ABSTRACT

Objective: To determine the presence of antibodies against phase II among slaughterhouse workers in Kerman, southeast of Iran.

Methods: The antibody titers of the serum samples were measured by enzyme-linked immuno sorbent assay using phase II *Coxiella burnetii* as the antigen [kit (Virion\Serion, Wurzburg, Germany) according to the manufacturer's protocol].

Results: The positive rate of IgG antibody was 68% in the slaughterhouse workers.

Conclusions: Our findings suggest that slaughterhouse workers in Kerman area have a higher risk of infection and should consider potential infection with *Coxiella burnetii*.

KEYWORDS

Q fever, *Coxiella burnetii*, Slaughterhouse workers, Kerman, Iran

1. Introduction

Q fever is a zoonotic disease, caused by the obligate intracellular bacterium *Coxiella burnetii* (*C. burnetii*) and is known as an occupational disease of veterinarians, farmers and abattoir workers[1]. The main sources of *C. burnetii* shedding into the environment are manure, urine, milk, and especially birth materials such as amnion fluid and placenta material. Humans contract infection mainly through inhalation of the infectious aerosols that generated by infected animals or animal products[1,2]. Q fever has been considered primarily an occupational zoonotic disease for abattoir workers, sheep shearers, livestock farmers, and especially veterinarians because of their direct contact with potentially infected animals[3].

Symptomatic acute Q fever mainly presents as fever and headache, hepatitis, or pneumonia chronic Q fever that may develop within several months or years after an acute infection in 10% to 20% of all acute Q

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fever cases[4]. Chronic Q fever generally presents as a culture-negative endocarditis or vascular infection with a high case fatality[5]. *C. burnetii* has two distinct antigenic presentation and/or phases: the virulent phase I variant and the avirulent phase II variant. In the body, *C. burnetii*is controlled by the T-cell dependent immune system, resulting in the production of specific antibodies. Acute Q fever is characterized by high levels of anti-phase II Ab(s) whereas chronic Q fever is characterized by increasing titers of anti-phase I Ab(s). The diagnosis of Q fever is usually made through serological testing[1].Few serological studies have been carried out in human populations in Iran. In a study, Q fever serology examined in 75 febrile patients in Kerman (Eastern Iran) that 24% and 36% of the patients had phase I antibodies and phase II antibodies, respectively[6]. Also in a previous study conducted in Zahedan, Southeastern Iran were positive among 105 febrile patients, 35.2% and 34.3% febrile patients had a positive serology test for acute Q fever and past infection[7].

Although cases of Q fever have been documented in Iran, the knowledge of the disease in Iran is not yet known. Q fever is a demonstrated occupational hazard to those employed in zoological professions, but the risk to slaughterhouse workers has not yet been quantified.

The aim of the present study was to determine the presence of antibodies against phase II among slaughterhouse workers in Kerman, southeast of Iran. This is the first study that examined the phase II antibodies for *C. burnetii* among slaughterhouse workers.

2. Materials and methods

2.1. Study area

Kerman is located at 30°17′13″N and 57°04′09″E southeast of Iran. The mean elevation of the city is about 1 755 m above sea level. Based on climate, soil, and other geographical conditions, Kerman has different vegetation and agricultural type. Density of livestock animals in this area per square kilometer is 33 livestock animals in 1 km^2; however, this density in cultivable lands is 505 livestock animals in 8 km^2.

2.2. Collection of samples

This study was conducted to determine the presence of antibodies against phase II among slaughterhouse workers in south eastof Iran. Serum specimens were collected from 75 slaughterhouse worker using simple random sampling from September 2010 to March 2011.

The slaughterhouse workers in this study came into daily contact with livestock, and did not report any specific symptoms of Q fever. Age and years of occupational experience were recorded as risk factors in this study. Moreover, among the workers, all of them were male.

The samples were transported to the laboratory of the Veterinary Faculty of Shahid Bahonar University of Kerman on ice and centrifuged at 1 500 μg for 10 min at room temperature. Serum was separated from the samples and stored at −20 °C until analysis by enzyme-linked immuno sorbent assay (ELISA).

2.3. Serological tests

Serum samples were tested for antibodies against phase II *C. burnetii* using a commercially available indirect ELISA test kit (Virion\Serion, Wurzburg, Germany) according to the manufacturer′s protocol.

The plates were read at 405 nm by an ELISA reader (Anthos 2020, Wals, Austria). Optical density cut-off values and control sera were checked. For phase II, antibody activities in IU/mL were calculated by a standard curve which was incorporated in the kit using the manufacturer′s guidelines as follows: <20 IU/mL, negative; 20-30 IU/mL, doubt; >30 IU/mL, positive.

2.4. Statistical analysis

The computer software, SPSS Version for SPSS 15.0 Windows was used for statistical analysis. To compare relative frequency of infection between different groups, *Chi*-square test was used. Differences were considered significant when $P<0.05$.

3. Results

The mean age was 38 years with a range from 20 to 50 years. The characteristics of the subjects in this study are summarized in Table 1. IgG phase II antibodies were detected in 51 of the 75 participants (68%). There was no significant correlation between years of occupational experience and the titers of IgG phase II antibodies (Table 1). Seropositive cases were higher in 31 to 40 years old group, but there were no significant differences in age among the four groups (Table 1).

Table1

The rate of phase II antibodies against *C. burnetii* in different age groups and years of occupational experience.

Parameters		Seropositive		Seronegative		*P*
		No.	Percent (%)	No.	Percent (%)	
Age	20-30	12	66.6	6	33.3	*P*>0.05
	31-40	23	76.6	7	23.3	*P*>0.05
	41-50	10	62.5	6	37.5	*P*>0.05
	>50	6	54.5	5	45.4	*P*>0.05
Occupational experience (years)	1-5	11	68.7	5	31.2	*P*>0.05
	6-10	14	70.0	6	30.0	*P*>0.05
	11-20	16	76.1	5	23.8	*P*>0.05
	>20	8	44.4	10	55.5	*P*>0.05

4. Discussion

In this study, we tested samples from healthy slaughterhouse workers with ELISA test and 68% were positive for IgG phase II antibodies. This survey clearly demonstrates that *C. burnetii* phase II antibody is more prevalent in slaughterhouse workers. This indicates that slaughterhouse workers may have a higher risk of exposure to *C. burnetii* through their occupational contact with animals.

C. burnetii infection is a public health problem in many countries, such as European countries, Canada, Japan, Turkey and Australia. Therefore, epidemiologic and clinical features are known from research studies. There is not any data about seroprevalence of *C. burnetiiin* slaughterhouse workers in Iran. Moreover, there are few studies in human population in some part of Iran. In a study in Kerman (southeast of Iran), Q fever serology examined in 75 febrile patients, using the ELISA method that 24% and 36% of the patients had phase I antibodies and phase II antibodies, respectively[6]. Also, in a previous study conducted in Zahedan, Southeastern Iran, 35.2% and 34.3% febrile patients had a positive serology test (IFA test) for acute Q fever and past infection[7]. The results of this study was much higher than described in Iranian febrile patients and several international seroepidemical studies among livestock veterinarians[3,8]. In the Slovak republic, *C. burnetii* phase II antibodies were detected in 63% of employees (of the Veterinary University)[8].

The veterinarians and patients in the previous survey may have had less contact with animals than the slaughterhouse workers in our study. However, it is difficult to compare the results of this survey with previous serological studies in Iran and other countries because different criteria were used. Moreover, seropositive rates differ from one region to another and vary in different countries probably because of different epidemiologic and climatic environments. In addition, the use of a different laboratory test and cut-offs, differences in study population and different infection rates of livestock over time could be possible explanations for other seroprevalence estimates.

A study conducted in Turkey estimated a prevalence of 13.5% for Q fever in otherwise healthy people[9]. Q fever was mainly observed in people over 30 years of age, hunters, and slaughterhouse workers. In this study, seropositive cases were higher in 31 to 40 years of age group (*P*>0.05). In other studies, seroprevalence to *C. burnetii* tended to increase with age. Higher prevalences at a more advanced age could be explained by longer exposure of older people[10].

The years of occupational experience was one of risk factors in this study. Seropositive cases were higher in worker between 11 to 20 years of occupational experience in the slaughterhouse workers (*P*>0.05). Slaughterhouse workers have a high risk of getting *C. burnetii* seropositive because of long term contact with potentially infected livestock. In other studies, contact with livestock is described as an important risk factor for seropositivity[3,8,11], and exposure to goats was the most important risk factor associated with *C. burnetii* infection in southeast of Iran because goats are extensively raised in close contact with humans in Southeast Iran[6]. A considerable number of Q fever outbreaks have been observed where goats were the source of infection in humans[1]. In recent studies in southern parts of Iran, the higher Q fever seroprevalence has been reported in goats (65.78%) than in cattle (10.75%) and sheep (29.42%)[6,12,13]. And high contact with goat flocks suggests that these flocks may be a significant risk factor for the apparent human Q fever seropositivity.

Further studies are needed to investigate the risk factors for Q fever in Iran which can suggest ways to prevent and reduce transmission. Several actions have been proposed to prevent and reduce the animal and environmental contamination. The vaccination of animals in infected flocks, as well as in uninfected ones close to them, with an efficient vaccine can prevent abortions and shedding of the bacteria[1], but the target animal populations need to be well defined for this to be effective.

This study is the first study that examined the seroprevalence for *C. burnetii* among slaughterhouse workers. Based on the above results, it can be concluded that the major cause of high apparent seropositivity to Q fever in this study area may be due to contact with livestock and livestock products. This suggests the importance of Q fever as an occupational risk for slaughterhouse workers. Since clinical Q fever illness was not self reported, further researches are recommended to study the health implications of seropositivity. Moreover, more investigations on the other reservoirs and other human populations (especially at risk population) are necessary to make epidemiologic feature of coxiellosis

in different parts of the province and the country. Overall, this study contributes to the knowledge and the awareness of Q fever as a risk for slaughterhouse workers in order to contribute to its prevention.

Conflict of interest statement

We declare that we have no conflict of interest.

Acknowledgements

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Comments

Background

The disease is caused by *C. burnetii*, a strictly intracellular, Gram-negative bacterium. Many species of mammals, birds, and ticks are reservoirs of *C. burnetii* in nature. *C. burnetii* infection is most often latent in animals, with persistent shedding of bacteria into the environment.

Research frontiers

The present study determined the presence of antibodies against phase II among slaughterhouse workers in Kerman, southeast of Iran. This is the first study that examined the phase II antibodies for *C. burnetii* among slaughterhouse workers.

Related reports

The results of this study was much higher than described in Iranian febrile patients and several international seroepidemical studies among livestock veterinarian. In the Slovak republic, *C. burnetii* phase II antibodies were detected in 63% of employees (of the Veterinary University).

Innovations and breakthroughs

This is the first study that examined the phase II antibodies for *C. burnetiia* mong slaughterhouse workers.

Applications

We can get information about Q fever as a risk for slaughterhouse workers in order to contribute to its prevention.

Peer review

This result suggests the importance of Q fever as an occupational risk for slaughterhouse workers. Q fever has been considered primarily an occupational zoonotic disease for abattoir workers, sheep shearers, livestock farmers, and especially veterinarians because of their direct contact with potentially infected animals.

References

[1] Angelakis E, Raoult D. Q Fever. *Vet Microbiol* 2010; **140**(3-4): 297-309.

[2] Wallensten A, Moore P, Webster H, Johnson C, Van der Burgt G, Pritchard G, et al. Q fever outbreak in Cheltenham, United Kingdom, in 2007 and the use of dispersion modelling to investigate the possibility of airborne spread. *Euro Surveill* 2010; **15**(12). pii: 19521

[3] Whitney EA, Massung RF, Candee AJ, Ailes EC, Myers LM, Patterson NE, et al. Seroepidemiologic and occupational risk survey for *Coxiella burnetii* antibodies among US veterinarians. *Clin Infect Dis* 2009; **48**: 550-557.

[4] Sukocheva OA, Marmion BP, Storm PA, Lockhart M, Turra M, Graves S. Long-term persistence after acute Q fever of non-infective *Coxiella burnetii* cell components, including antigens. *QJM* 2010; **103**: 847-863.

[5] Parker NR, Barralet JH, Bell AM. Q fever. *Lancet* 2006; **367**: 679-688.

[6] Khalili M, Shahabi-Nejad N, Golchin M. Q fever serology in febrile patients in Southeast Iran. *Trans R Soc Trop Med Hyg* 2010; **104**: 623-624.

[7] Metanat M, Sepehri Rad N, Alavi-Naini R, Shahreki S, Sharifi-Mood B, Akhavan A, et al. Acute Q fever among febrile patients in Zahedan, Southeastern Iran. *Turk J Med Sci* 2014; **44**: 99-103.

[8] Dorko E, Kalinova Z, Weissova T, Pilipcinec E. Seroprevalence of antibodies to *Coxiella burnetii* among employees of the Veterinari University in Kosice, Eastern Slovakia. *Ann Agric Environ Med* 2008; **15**: 119-124.

[9] Gozalan A, Rolain JM, Ertek M, Angelakis E, Coplu N, Basbulut EA, et al. Seroprevalence of Q fever in a district located in the west Black Sea region of Turkey. *Eur J Clin Microbiol Infect Dis* 2010; **29**: 465-469.

[10] Cardeñosa N, Sanfeliu I, Font B, Muñoz T, Nogueras MM, Segura F. Seroprevalence of human infection by *Coxiella burnetii* in Barcelona (northeast of Spain). *Am J Trop Med Hyg* 2006; **75**(1): 33-35.

[11] Chang CC, Lin PS, Hou MY, Lin CC, Hung MN, Wu TM, et al. Identification of risk factors of *Coxiella burnetii* (Q fever) infection in veterinary-associated populations in Southern Taiwan. *Zoonoses Public Health* 2010; **57**: e95-101.

[12] Khalili M, Sakhaee E. An update on a serologic survey of Q fever in domestic animals in Iran. *Am J Trop Med Hyg* 2009; **80**: 1031-1032.

[13] Sakhaee E, Khalili M. The first serologic study of Q fever in sheep in Iran. *Trop Anim Health Prod* 2010; **42**: 1561-1564.