Prevalence of Coxiella burnetii in Bulk Tank Milk Samples from Dairy Cattle in West and Northwest of Iran

Arvin Shajeie (PhD)
Department of Pathobiology, Faculty of Veterinary Medicine, Ferdowsi University, Mashhad, Iran

Mohammad Khalili (PhD)
Research Center of Tropical and Infectious Disease, Kerman University of Medical Sciences, Kerman, Iran

Zeinab Abiri(PhD)
Department of Pathobiology, Faculty of Veterinary Medicine, Shahid Bahonar University of Kerman, Kerman, Iran

Sajad Akbari (MSc)
Department of Pathobiology, Faculty of Veterinary Medicine, Shahid Bahonar University of Kerman, Kerman, Iran

Corresponding author: Mohammad Khalili

Tel: + 983433202903

Email: mdkhalili1@yahoo.com

Address: Kerman University of Medical Sciences, Kerman, Iran

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ABSTRACT

Background and Objectives: Q fever is a zoonotic disease caused by an obligate gram-negative intracellular pathogen called Coxiella burnetii. The aim of this study was to determine the seroprevalence of anti-C. burnetii antibodies in bulk tank milk (BTM) samples of dairy cattle in west and northwest of Iran.

Methods: Overall, 71 BTM samples (covering nearly 700 dairy cattle) were collected in autumn 2013. A commercial Q fever antibody ELISA Test Kit (Liebefeld-Bern, Switzerland) was used to identify the presence of antibodies against inactivated phase 1 and phase 2 C. burnetii antigens.

Results: The results of ELISA test showed that 17 BTM samples (23.9%) were positive for the presence of anti-C. burnetii antibodies .

Conclusion: This study is the first to evaluate presence of anti-C. burnetii antibodies in BTM samples from dairy cattle herds in west and northwest of Iran. The high prevalence of this pathogen highlights the need for pasteurization of raw milk and raising awareness in consumers of dairy products in these regions.

Keywords: Q fever, Cattle, Iran, Coxiella.

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INTRODUCTION

Q fever is a zoonotic disease caused by an obligate gram-negative intracellular pathogen Coxiella burnetii. The disease is endemic in every part of the world except for New Zealand (1, 2). C. burnetii has been detected in different domestic animals such as cattle, sheep and goats, which are considered as main reservoirs of the pathogen (3, 4). Although C. burnetii infection in cattle may be asymptomatic, it could cause several reproductive disorders in livestock such as abortion, stillbirth, premature deliveries and delivery of weak and unviable newborns. In addition, incidence of metritis and infertility due to C. burnetii infection is more frequent in bovinae compared to other ruminants (4, 5). Moreover, infection of these animals with C. burnetii decreases efficiency of milk production (6). In humans, acute Q fever causes self-limiting flu-like symptoms, while chronic Q fever is generally associated with endocarditis and hepatitis (7). The main route of infection for humans is inhalation of aerosols contaminated with C. burnetii shed from infected animals. The pathogen could be shed to the environments through birth products, urine, faeces and milk of cattle suffering from miscarriage. Ingestion of C. burnetii-contaminated milk or milk products may result in serological conversion, which is accompanied by clinical manifestations in some cases (5). Several cheap and fast serological techniques are widely used in epidemiological studies (8). The European Union Food Safety Authority and Panel on Animal Health and Welfare have recommended diagnosis of Q fever by enzyme-linked immunosorbent assay (ELISA). Testing milk samples using ELISA could be a suitable choice for screening lactating dairy cows for detection of C. burnetii (8). The aim of this study was to investigate the presence of antibodies against C. burnetii in cow milk Samples collected from bulk-tank milk (BTM) of dairy cattle in west and northwest of Iran.

MATERIAL AND METHODS

Overall, 71 BTM samples were randomly collected in autumn 2013, from more than 30 different traditional dairy farms. Of all samples, 34 were collected from cattle herds in Kurdistan Province (western Iran), and the remaining 37 collected from herds in West Azerbaijan Province (northwest of Iran). The sample size was determined based on accessibility to herds. First, 10 ml of milk from each BTM was collected for analysis. After centrifugation, fat was discarded and non-fat-containing phase was kept in freezer for later experiments. Highly sensitive and specific CHEKIT Q fever antibody ELISA test kit (IDEXX, Liebefeld-Bern, Switzerland) and complement fixation test (98% correlation between the two methods) were used to evaluate the presence of antibodies against inactivated phase 1 and phase 2 C. burnetii antigens in the collected milk samples. Later, optical density of the samples was measured. According to the manufacturer's instructions, $S/P \geq 40\%$, $S/P < 30\%$ and results in the interval $30\% \leq S/P < 40\%$ were considered as positive, negative and intermediate, respectively. Data was analysed by SPSS software (version 18, IBM Company, United States). Chi-square test was used to calculate two-tailed P-values for independent variables and the linear trend in stratified data.

RESULTS

Of 71 samples, 17 BTM samples (23.9%) were positive in the ELISA test (Table1).

The presence of anti- C. burnetii antibodies had no significant association with number of cows and daily milk production. However, there was a statistically significant relationship between type of farm and rate of positive samples ($P<0.03$).

Table 1- Presence of antibodies against *C. burnetii* in BTM samples

IDEXX	BTM samples	
	Kurdistan Province	West Azerbaijan Province
Positive (S/P >40%)	7 (20.5%)	10 (27%)
Intermediate (30% ≤ S/P < 40%)	-	-
Negative (S/P < 30%)	27 (79.5%)	27 (73%)

DISCUSSION

This study investigated the presence of antibodies against C. burnet in milk sample collected from cattle herds in the Kurdistan Province and West Azerbaijan Province using ELISA. The results showed that 23.9% of samples contained anti-C. burnetii antibodies. In Turkey and Iraq, the prevalence of C. burnetii in cows and small ruminants (goat and sheep) has been reported to be 12.4% and 16%, respectively (9,10). Previous studies have also investigated the prevalence of C. burnetii contamination in various provinces of Iran including Kerman (45.4%), Chaharmahal and Bakhtiari (6.2%) and Isfahan (3.2%) (11, 12). Our study is the first to evaluate the presence of anti-C. burnetii antibodies in BTM samples from dairy cattle herds in the west and northwest of Iran. Based on the results of the present study and previous studies, it can be concluded that Iran is a hyperendemic area for Q fever. It should be noted that C. burnetii is able to produce spore-like form that are resistant to adverse environmental conditions. C. burnetii has also been detected in breathable dusts from arid regions of Iraq and Kuwait (13). Therefore, presence of C. burnetii in the west of Iran might be related to the dust storms hitting Iran from Iraq and Kuwait. The presence of anti-C. burnetii antibodies in BTM may be attributed to various factors including housing systems, herd size, lack of appropriate farm management, etc. Thus, proper management of farms and careful monitoring of cattle housing systems may be beneficial approaches for preventing spread of contamination between herds (14).

CONCLUSION

Our study shows that 23.9% of BTM samples from the Kurdistan and West Azerbaijan Provinces are contaminated with C. burnetii. Since the presence of anti-C burnetii antibodies indicates past infection with the pathogen, the high prevalence of this pathogen highlights the need for pasteurization of raw milk and raising awareness in consumers of dairy products in these two provinces.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

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