

Serosurvey of Q fever in active reproductive rams in northern Palestine

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Abstract

This investigation of Q-fever or Coxiellosis was undertaken to study the presence and the infection rate of *C. burnetii* infection in reproductively active rams in the Northern Palestine, where most of the sheep herds are located. In all, 2806 samples which collected from active rams during the reproductive season from herds in five cities (Jenin, Jericho, Nablus, Tulkarm and Tubas). Sera were tested by ID Screen[®] Q Fever Indirect Multi-species enzyme-linked immunosorbent assay (ELISA) for detection of *C. burnetii* immunoglobulin G (IgG). A total of 28.1 % of ram sera were positive for *C. burnetii* IgG. There was no significant difference between the four location cities studied However, a significantly low difference was observed in the Tubas city ($P < 0.05$). Highest infection rate was detected in rams of Tulkarm (29.8%), followed by Nablus (29.5%), Jericho (28.4%), Jenin (28.2%) and Tubas (16.1%). Moreover, at the farm level, 73.3% had at least one seropositive animal. It had been concluded that a high infection rate were detected in rams of Northern Palestine, at both individual and herd level. Therefore, Q fever could be responsible for considerable numbers of ovine abortions in Palestine, as well as of public health significance, Hereby control programs should be advised.

Keywords: Q fever, *Coxiella burnetii*, Serosurvey, Ram, Palestine

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معدل انتشار حمى كيو في الكباش التناسلية النشطة في شمال فلسطين

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الخلاصة

أجريت هذه الدراسة لاكتشاف وجود ومعدل انتشار خمج *C. burnetii* في كباش نشطة تناسليا في شمال فلسطين، حيث تقع معظم قطعان الأغنام. تم جمع ٢٨٠٦ عينة من الكباش النشطة تناسليا من قطعان في خمس مدن (جنين وأريحا ونابلس وطولكرم وطوباس). إذ تم فحص المصل باستخدام اختبار الاليزا للكشف عن الاجسام المضادة الخاصة بالعامل المسبب *C. burnetii*. اظهرت النتائج ان ٢٨,١% من مصل الدم الحيوانات المفحوصة كانت مصابة بالمرض وبدون فرق معنوي بين أربع من المدن المشمولة بالدراسة. فضلا عن ذلك، لوحظ انخفاض طفيف في مدينة طوباس ($P < 0.05$). أعلى نسبة انتشار للمرض سجلت في كباش طولكرم (٢٩,٨%) تبعتها نابلس (٢٩,٥%) وأريحا (٢٨,٤%) جنين (٢٨,٢%) وطوباس (١٦,١%). على مستوى المزرعة، كان ٧٣,٣% من المزارع مصابة بالمرض. استنتج من هذه الدراسة ان مرض حمى كيو كشفت في شمال فلسطين بنسب عالية وقد تكون احد السباب المؤدية الى مشاكل تناسلية كالإجهاض او مشاكل انتقالها الى البشر عليه ينصح بوضع خطط للوقاية بشكل مبكر.

Introduction

Q-fever or Coxiellosis is an important notifiable zoonotic disease caused by an intracellular microorganism

Coxiella burnetii. The name came after a (query fever) describe a febrile illness originally observed in abattoir workers in Australia (1). The disease is worldwide distributed among wild, pet and farm animals (2). Dairy

animals are the primary animal reservoirs of the infectious agent (3). Susceptible animals, as well as human, get the infection via inhalation of the contaminated aerosol or through direct contact with feces, urine or infected uterine fluid. The venereal transmission of *C. burnetii* is well documented in both human and animals (2,4). The infection in animals is usually subclinical and may be associated with general signs include anorexia and fever. Infected animals keep shedding of *C. burnetii* for long period. However, it could also shed in milk which plays an important epidemiological role in the transmission of disease to human.

Dairy animals are of importance significant for economic in Palestine. With approximately 670,000 heads, sheep consider the most abundant farm animal, followed by goats (204,000) and cattle (25,000) (5). In sheep, the infection are mostly subclinical; however, the main clinical findings in ewes are placentitis, late pregnancy abortions and stillbirths (2,6,7). The incidence of sheep's abortion is very high in Palestine without specific clinical signs and known etiology. A significant number of these abortions occurring in mid to late gestation (8). The subclinical state of the disease, as well as the long period shedding of untreated infected animals, enhances the risk of human infection. The clinical presentation in people varies from asymptomatic self-limiting disease or mildly symptomatic influenza-like illness. Q fever may be a fatal disease due to pneumonia, hepatitis, and endocarditis particularly in immunocompromised hosts and in pregnant women (3,9).

Isolation of *C. burnetii* is hazardous and requires level-3 biosafety (10). Serological methods provide valuable tools for diagnosis of Q fever in humans and animals, facilitate epidemiological studies and measures to control the disease (10). Enzyme-linked immunosorbent assay (ELISA) has high sensitivity and good specificity in detection antibodies, particularly animal shedders (11,12). Cases of Q fever has been reported in human and animals in the neighboring countries (13,14). However, no case has been reported in Palestine. This might be related to lack of diagnosis and records keeping. This study presents for the first time an investigation into the presence and the infection rate of *C. burnetii* in Palestine sheep. Prevalence data of Q fever infection in domestic animals are important to support risk assessments and decisions on the preventive measures regarding public and animal health. This will enable the detection of the potential changes in seroprevalence of the disease for the future studies.

Materials and methods

Area of the study, Sample collection and preparation

This survey was performed in 2016 to determine the infection rate of *C. burnetii* in the Palestinian sheep population. The study was carried out on Jenin, Jericho,

Nablus, Tulkarm and Tubas cities in Palestine. The number of farms sampled on each city was 47, 238, 46, 11 and 11, respectively. Numbers of rams tested depending on the farm size (5-15 ram); all rams in the farm were tested. The targeted regions were chosen as they were the highest sheep population. The samples were taken from active reproductive rams during the reproductive season (late June and early April). All samples were collected in plain tubes and sent to the diagnostic laboratory at the Palestinian Livestock Development Center for analysis.

Laboratory examinations

Antibodies to *C. burnetii* antigens were detected by ID Screen[®] Q fever indirect Multi-species kit, (France). Following the manufacturer's instruction.

Statistical analysis

Data were analyzed with statistical software SPSS version 20th (SPSS Inc, USA). The Chi-square test was used to determine the association between the infection rate of Q fever antibodies and the city. *P* value < 0.05 was considered statistically significant (15).

Results

Out of 2806 blood samples serologically investigated in this study, 789 (28.1%) were found *C. burnetii* seropositive, however 2017 (71.9 %) were found seronegative. No significant statistical difference between four cities have been detected. The prevalence rates were found high Jenin (28.2%), followed by Nablus (29.5%), Tulkarm (29.8%), and Jericho (28.4%). However, there the prevalence in Tubas city was (16.1%), which was significantly lower than the other cities ($P < 0.05$) Table 1.

Moreover, at the farm level, among the 353 farms studied, 259 (73.3 %) had at least one seropositive animal. The highest rate was found in the farms of Jenin (91.5%), followed by Nablus (86.9%), Tulkarm (81.8%), Tubas (72.7%) and Jericho (66.8%). Table 2.

Discussion

C. burnetii infection is one of the most common causes of sheep abortion worldwide (16,17). The sexual transmission of *C. burnetii* is well known in both human and animals (2,4). Prevalence studies have been conducted in several countries, confirmed that *C. burnetii* is widely distributed in sheep. Serological methods, particularly ELISA provides safe and valuable tool for diagnosis of Q fever in humans and animals, facilitate epidemiological studies and application control and prevention measures of the disease (10,11). The present study is the first epidemiological investigation of ovine *Coxiellosis*, estimates at the individual and the farm level the prevalence

in a representative number of randomly selected active reproductive rams in Palestine. Currently, vaccination against sheep *C. burnetii* is not practiced in Palestine. So,

the detected antibodies in this survey imply a natural response to exposure to the microorganism.

Table 1: Infection rate of Q fever in rams of northern Palestine

	Jericho	Jenin	Nablus	Tubas	Tulkarm	Total
Positive/overall (Infection rate %)	377/1327 28.4a	174/617 28.2a	187/634 29.5a	20/124 16.1b	31/104 29.8a	789/2806 28.1

Pearson Chi-Square =9.62. Each subscript letter denotes a subset of infected farm categories whose column proportions do not differ significantly from each other at the.05 levels.

Table 2: Infection rate of Q fever, according to farm level in north Palestine

	Jericho	Jenin	Nablus	Tubas	Tulkarm	Total
Positive/overall (Infection rate %)	159/238 66.8 ^b	43/47 91.5 ^b	40/46 86.9 ^b	8/11 72.7 ^a	9/11 81.8 ^a	259/353 73.3

Pearson Chi-Square = 21.76. Each subscript letter denotes a subset of infected farm categories whose column proportions do not differ significantly from each other at the.05 levels.

Results of this epidemiological study revealed a variation of infection rate between the studied cities. A significantly lower infection rate was detected in Tubas city compared to other cities. However, this low prevalence rate of specific antibodies for *C. burnetii* cannot be explained by changes in rearing type and geographical origin. The overall infection rate was 28.1 % at individual level, which was higher than reported by Asadi et al. (18) in Northern Iran, where 19.5% sheep were seropositive (18). In Turkey, seroprevalence analysis showed that 20% of sheep were seropositive and 81% of the flocks surveyed revealed at least one seropositive animal (19). Seroprevalence analysis in Egypt indicated that 22.5 % of sheep were positive to *C. burnetii* (20). A large Q-fever outbreaks have been observed in Israel, where farm animals have been shown to be the source of infection in humans (21).

Seroprevalence reports included both male and female sheep did not show a significant correlation between *C. burnetii* infection among gender (22,23). The ratio of reproductive ram to ewes is 1:20. In the male, Q fever did not have obvious clinical representation. Targeting of rams during active reproduction seasons provide an extended view of the infection rate in sheep and sheep farms. In addition, this approach is important for control the sexual transmission of the disease.

Q fever has a high prevalence among sheep in Palestine compared to the other countries. Identification of the associated risk factors of the disease is required for the improvement of control measures. In the present study; the risk factors for Q fever in sheep and sheep flocks have not been investigated. Unfortunately, this is not available among traditional farms in Palestine. The lacking of animals record keeping is a major issue. Previous studies in

other areas mentioned several risk factors for farms and animals to acquire a *C. burnetii* infection. Domestic pets, such as cats, dogs and domestic birds are known to be a source of infection. In addition, ticks are believed to play crucial role in the transmission of the agent from infected wild vertebrates to farm animals (24). In a study in the Canada, the flock size, density and hygiene were the strong associated risk factors (25). The rearing system in Palestine mostly relies in mixing between sheep, goat and or other Q fever susceptible farm animals (5). In addition, the free movement of the animal flocks, poor farms fencing and buying without adequate quarantine might be additional factors for spreading of the infection among sheep. A number of disease control measures could be used to control the disease, including measures to increase diagnostic accuracy and general awareness. Precautions to reduce spillover of infection in farm animals include screening animals prior to reproductive season, vaccination and eradication program (26).

The findings of this study confirmed the high prevalence of Q fever among sheep in the investigated regions. Further investigations are required to characterize the epidemiology of Q-fever infection in human and animals In addition, the Identification of the associated risk factors of the Q fever is essential for the improvement of control measures.

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Authors' contributions

N. Jalboush was responsible for regional blood collection and for the diagnostic work (ELISA) and helped drafting the manuscript. I. Alzuheir carried out the analysis of data and for drafting the manuscript. Authors read and approved the final manuscript.

Conflict of interest

None of the authors of this paper have a financial or personal relationship with other people or organizations that could inappropriately influence or bias the content of the paper.

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