African Journal of Microbiology Research Vol. 5(23), pp. 3964-3967, 23 October, 2011
Available online http://www.academicjournals.org/ajmr
ISSN 1996-0808 ©2011 Academic Journals

Full Length Research Paper

Seroprevalence of Q fever in dairy cows in northeastern China

Hong-Bo NI[1,3], Si-Guo LIU[2], Hai-Fang JIANG[1], Chun-Ren WANG[1] and Ai-Dong QIAN[3]*

[1]College of Animal Science and Veterinary Medicine, Heilongjiang Bayi Agricultural University, Daqing, Heilongjiang Province 163319, P. R., China.
[2]Division of Bacterial Diseases, National Key Laboratory of Veterinary Biotechnology, Harbin Veterinary Research Institute of Chinese Academy of Agricultural Sciences, Harbin, Heilongjiang Province 150001, P. R., China.
[3]College of Animal Science and Technology, Jilin Agricultural University, Changchun, Jilin Province 130118, P. R., China.

Accepted 9 September, 2011

A total of 564 serum samples were randomly collected from 29 farms located in the nine main dairying districts in China's north-eastern Heilongjiang Province from 2009 to 2010, and the seroprevalence of antibodies against *Coxiella burnetii* was determined by indirect enzyme-linked immunosorbent assay (ELISA). The average seroprevalence of *C. burnetii* in dairy cattle was 10.1% (57/564). The seroprevalence of *C. burnetii* in aborting cows (11.6%) was higher than that in non-aborting cows (5.1%). Dairy cows aged 3 years had the highest seroprevalence (20.5%), followed by dairy cows aged 4 years (14.3%). Dairy cows in their first pregnancy had the highest seroprevalence (25.8%). There was an apparent association between *C. burnetii* seropositivity and abortion, age, and number of pregnancies ($P < 0.05$). The results of the present survey indicate that *C. burnetii* infection in dairy cows is widespread in Heilongjiang Province, and it may be one of the most major causes of dairy cow abortion.

Key words: Q fever, dairy cows, serologic survey, abortion, age, pregnancies.

INTRODUCTION

Q fever is a zoonotic disease caused by an obligate intracellular small gram-negative bacterium, *C. burnetii*. This disease is endemic throughout the world, occurring in various geographic regions and climatic zones (Woldehiwet et al., 2004).This bacterium which can infect a wide range of susceptible hosts including cows, sheep, and goats, pets, wild mammals, human and even non-mammalian species, such as domestic and wild birds, reptiles, and ticks. Infected females shed huge numbers of *Coxiella* into birth products (Arricau et al., 2003; Masala et al., 2004) and smaller numbers in urine, feces, and milk. This shedding may persist over several months, particularly in vaginal mucus, feces, and milk, even in those of females with normal parturition (Berri et al., 2005; Kim et al., 2005). *C. burnetii* may induce reproductive disorders such as abortion, stillbirth, and delivery of weak and nonviable neonates in ruminants. Metritis and infertility are frequently the main clinical signs of infection in cows. However, in animals, Q fever is rarely symptomatic, except for manifestations as reproductive disorders in females. The disease is transmitted to humans incidentally by inhalation of aerosols from infected cows and sheep (Arricau et al., 2003; Berri et al., 2005; Muskens et al., 2007). Although, person to person transmission is extremely rare (Kim et al., 2005; Maurin et al., 1999), humans infection with *C. burnetii* may lead to serious complications and even death in patients with acute disease, especially those with meningoencephalitis or myocarditis and more frequently in chronically infected patients with endocarditis. Q fever has been reported in almost every country except New Zealand (Hilbink et al., 1993; Masala et al., 2004). Among serologic tests for detection of antibodies against *C. burnetii*, ELISA and immunofluo-rescence assay (IFA) are commercially available. ELISA is preferable to IFA for serologic study

*Corresponding author. E-mail: qianaidong0115@163.com. Tel: +86-431-84533426. Fax: +86-431-84533426.

because it has higher sensitivity (Rousset et al., 2007).

In China, there were some reports about *C. burnetii* infection in humans and animals (Chai et al., 2010; Lai et al., 2008). But there is little information concerning the epidemiology of *C. burnetii* infection in northeast China. The aim of this survey was to investigate the presence of *C. burnetii* antibodies among the dairy cows in northeast China.

MATERIALS AND METHODS

Geographical location

Heilongjiang Province is situated in the northeastern part of mainland China between north latitudes of 43°25' to 53°33' and east longitudes of 121°11' to 135°05', sharing borders with Russia in the north and east, with Jilin province in the south, and with Inner Mongolian Autonomous Region in the west. It has 473,000 km^2 of land, a human population of approximately 38 million. Heilongjiang Province is divided into 13 districts (cities) with the city of Harbin as its capital. The study was conducted on 29 farms in 9 districts (Heihe, Mudanjiang, Harbin, Qiqihaer, Jiamusi, Shuangyashan, Jixi, Daqing and Suihua).

Collection of serum samples

A total of 564 serum samples were collected from 29 farms in the nine main cow-rearing districts (Table 1) between March 2007 and November 2008 in Heilongjiang province. The dairy cow populations represented local breeds (Chinese Holstein) as well as introduced breeds (American Holstein-Friesian and Australian Holstein-Friesian cows). All the blood samples were immediately transported on ice to the laboratory, and sera were separated immediately by centrifugation of blood at 1,500 x *g* for 15 min at room temperature and were kept at -20°C until the day of analysis. Biometric data for dairy cows including age, aborting or non-aborting, number of pregnancies, and animal husbandry practices were taken from the owners.

Detection of antibodies against *C. burnetii* by ELISA

Serum samples were tested for Q fever antibodies using the indirect ELISA kit (ID Screen® Q Fever Indirect ELISA kit, France), which was carried out following the protocol recommended by the manufacturer using Phase I + II purified antigens of *C. burnetii*. Sera were prepared at 1:10 dilution, and specific antibodies were measured using a peroxidase-labeled anti-ruminant immunoglobulin G (IgG) conjugate. Results were expressed as a percentage of the optical density reading of the test sample (value), calculated as value (S/P) = (S – N)/(P – N) x100, where S, N, and P are the OD of the test sample, the negative control, and the positive control, respectively. Sera were considered to be ELISA positive if they had a value of 50% or more, doubtful if the value was between 40% and 50%, and negative if the value was < 40%.

Statistical analysis

Statistical analysis of *C. burnetii* prevalence in different ages, aborting or non-aborting, different numbers of pregnancies, and different animal husbandry practices of dairy cows were performed by $\chi 2$-test with excel (Microsoft® Excel 2003). The differences were considered statistically significant if $P < 0.05$.

RESULTS AND DISCUSSION

Seroprevalence of *C. burnetii*

The sera of dairy cows were collected from aborting and non-aborting cows. The overall seroprevalence of *C. burnetii* in dairy cows is 10.6% (57/564) (Table 1). The seroprevalence of *C. burnetii* in dairy cows has been studied in many countries. In the present study, the seropositivity of *C. burnetii* was lower than that reported in some countries, for example, Danish, Netherlands, Dutch, and Spain (Agger et al., 2010; Guatteo et al., 2011; Muskens et al., 2011; Ruiz-Fons et al., 2010; Rodríguez et al., 2010), but was higher than that reported in some other countries, such as the Republic of Ireland and Russia (Ryan et al., 2010; Tokarevich et al., 2006).

Seroprevalence of *C. burnetii* in aborting and non-aborting cows

480 of the 564 dairy cows had a pregnancy history record for the analysis of the association of abortion and *C. burnetii* infection (Table 2). The seroprevalence of *C. burnetii* in aborting dairy cows (11.6%) was significantly higher than that in non-aborting dairy cows (5.1%), and the difference was statistically significant ($P < 0.05$). However, the overall prevalence of Q fever in dairy cows with the history of abortion was lower than that in India (Jones et al., 2010; McCaughey et al., 2009).

Association between seroprevalence and ages

528 of the 564 dairy cows had an age record for the analysis of the association between seroprevalence and ages (Table 3). The seroprevalence varied in different age groups, ranging from 2.8 to 20.5%, and there were statistically significant differences among the age groups ($P < 0.05$).

Association between seroprevalence and number of pregnancies

Of the 564 dairy cows, 523 had pregnancy record, the seroprevalence of *C. burnetii* infection in cows with different pregnancies is shown in Table 4. The seroprevalence of dairy cows that had given the first birth was the highest (25.8%), followed is cows with no record of pregnancy history (14.6%), and differences between different pregnancy groups were statistically significant ($P < 0.05$). It is not sure that about the unknown pregnancy history of 41 dairy cows were congenital infection or postnatal infection. It was reported that pregnant

Table 1. Seroprevalence of *Coxiella burnetii* in dairy cattle from Heilongjiang Province, China.

Geographical location	No. examined	No. positive	Percent positive (%)
Heihe	69	9	13.0
Mudanjiang	57	5	7.1
Harbin	54	4	7.4
Qiqihar	66	2	3.0
Jiamusi	61	2	3.9
Shuangyashan	63	9	14.3
Jixi	54	6	11.1
Daqing	76	11	14.5
Suihua	64	9	18.0
Total	564	57	10.1

Table 2. Seroprevalence of *Coxiella burnetii* infection in aborting and non-aborting dairy cows in Heilongjiang Province, north-eastern China.

Category	No. examined	No. positive	Percent positive (percent)
Aborting dairy cattle	363	42	11.6
Non-aborting dairy cattle	117	6	5.1
Unknown	84	4	4.8

Table 3. Seroprevalence of *Coxiella burnetii* infection in different ages of dairy cows in Heilongjiang Province, north-eastern China.

Age (year)	No. examined	No. positive	Positive percent (%)
1	24	1	4.2
2	58	3	5.2
3	73	15	20.5
4	91	13	14.3
5	87	6	6.9
6	67	4	9.0
7	84	8	9.5
8	40	3	7.5
>8	4	1	2.5
With no records	36	3	8.3

Table 4. Seroprevalence of *Coxiella burnetii* infection in dairy cows with different numbers of pregnancies in Heilongjiang Province, north-eastern China.

No. pregnancies	No. examined	No. positive	Positive percent (%)
0	54	2	3.7
1	66	17	25.8
2	63	9	14.3
3	68	5	7.4
4	77	5	6.5
5	66	3	4.5
6	72	4	5.6
7	31	2	6.5
>7	26	4	6.5
With no records	41	6	14.6

ruminants were highly susceptible to infection, and abortions occurred only at the first parturition after infection (Mohammad et al., 2009).

In this study, there was an apparent association between *C. burnetii* seropositivity and abortion, age, and number of pregnancies ($P < 0.05$). The results of the present survey indicate that *C. burnetii* infection in dairy cows is widespread in Heilongjiang Province, and it may be one of important pathogeny causing dairy cow abortion, further studies are needed.

ACKNOWLEDGEMENTS

This work was financially supported by the Department of Science and Technology of Jilin Province (Grant No.20100220), Education Department of Heilongjiang Province (1251G044), and. the State Key Laboratory of Veterinary Biotechnology (Grant No.SKLVBF200807).

REFERENCES

Agger JF, Christoffersen AB, Rattenborg E, Nielsen J, Agerholm JS (2010). Prevalence of Coxiella burnetii antibodies in Danish dairy herds. Acta. Vet. Scand., 52: 5.

Arricau Bouvery N, Souriau A, Lechopier P, Rodolakis A (2003). Experimental Coxiella burnetii infection in pregnant goats: excretion routes.Vet. Res., 34: 423-433.

Berri M, Crochet D, Santiago S, Rodolakis A (2005). Spread of Coxiella burnetii infection in a flock of sheep after an episode of Q fever. Vet. Rec., 157: 737-740.

Chai CL, Lu QY, Sun JM, Jiang LP, Ling F, Zhang LJ, Zheng SG, Zhang H, Ge JH (2010). Sero-epidemiologic investigation on tick-borne diseases of humans and domestic animals in Zhejiang province. Zhonghua Liu Xing Bing Xue Za Zhi (in Chinese), 31: 1144-1147.

Guatteo R, Seegers H, Taurel AF, Joly A, Beaudeau F (2011). Prevalence of *Coxiella burnetii* infection in domestic ruminants: A critical review. Vet. Microbiol., 149: 1-16.

Hilbink F, Penrose M, Kovacova E, Kazar J (1993). Q fever is absent from New Zealand. Int. J. Epidemiol., 22: 945-949.

Jones RM, Twomey DF, Hannon S, Errington J, Pritchard GC, Sawyer J (2010). Detection of *Coxiella burnetii* in placenta and abortion samples from British ruminants using real-time PCR. Vet. Rec., 167: 965-967.

Kim SG, Kim EH, Lafferty CJ, Dubovi E (2005). Coxiella burnetii in bulk tank milk samples, United States. Emerg. Infect. Dis., 11: 619-621.

Lai CH, Huang CK, Chin C, Chung HC, Huang WS, Lin CW, Hsu CY, Lin HH (2008). Acute Q fever: an emerging and endemic disease in southern Taiwan. Scand. J. Infect. Dis., 40: 105-110.

Masala G, Porcu R, Sanna G, Chessa G, Cillara G, Chisu V, Tola S (2004). Occurrence, distribution, and role in abortion of Coxiella burnetii in sheep and goats in Sardinia, Italy. Vet. Microbiol., 99: 301-305.

Maurin M, Raoult D (1999). Q Fever. Clin. Microbiol. Rev., 12: 518-553.

McCaughey C, Murray LJ, McKenna JP, Menzies FD, McCullough SJ, O'Neill HJ, Wyatt DE, Cardwell CR, Coyle PV (2009). *Coxiella burnetii* (Q fever) seroprevalence in cattle. Epidemiol. infect., 138: 21-27.

Mohammad K, Ehsanollah S (2009). An Update on a Serologic Survey of Q Fever in Domestic Animals in Iran. Am. J. Trop. Med. Hyg., 80: 1031-1032

Muskens J, van Engelen E, van Maanen C, Bartels C, Lam TJ (2011). Prevalence of *Coxiella burnetii* infection in Dutch dairy herds based on testing bulk tank milk and individual samples by PCR and ELISA. Vet. Rec., 168: 79.

Muskens J, Mars MH, Franken P (2007). Q fever: an overview. Tijdschr. Diergeneeskd., 132: 912-917.

Rodríguez NF, Carranza C, Bolaños M, Pérez-Arellano JL, Gutierrez C (2010). Seroprevalence of *Coxiella burnetii* in domestic ruminants in Gran Canaria Island, Spain. Transbound Emerg. Dis., 57: 66-67.

Rousset E, Durand B, Berri M, Dufour P, Prigent M, Russo P, Delcroix T, Touratier A, Rodolakis A, Aubert M (2007). Comparative diagnostic potential of three serological tests for abortive Q fever in goat herds. Vet. Microbiol., 124: 286-297.

Ruiz-Fons F, Astobiza I, Barandika JF, Hurtado A, Atxaerandio R, Juste RA, García-Pérez AL (2010). Seroepidemiological study of Q fever in domestic ruminants in semi-extensive grazing systems. BMC Vet. Res., 6: 3.

Ryan ED, Kirby M, Collins DM, Sayers R, Mee JF, Clegg T (2010). Prevalence of *Coxiella burnetii* (Q fever) antibodies in bovine serum and bulk-milk samples. Epidemiol. Infect., 15: 1-5.

Tokarevich NK, Freilykhman OA, Titova NM, Zheltakova IR, Ribakova NA, Vorobeychikov EV (2006). Anthropogenic Effects on Changing Q.Fever Epidemiology in Russia. Ann. N. Y. Acad. Sci., 1078: 120-123.

Woldehiwet Z (2004). Q fever (coxiellosis): Epidemiology and pathogenesis. Res. Vet. Sci., 77: 93-100.

Zhong Li, Tong Chen. Jie Huang (1996). Seroprevalence of Q Fever among different animals in Shandong Province of China. Chin. J. Zoonoses (in Chinese), 12: 12.