Seroepidemiologic Studies on Babesia equi and Babesia caballi Infections in Horses in Jilin Province of China

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(Received 25 December 2002/Accepted 15 May 2003)

ABSTRACT. The prevalence of equine piroplasmosis caused by Babesia equi and Babesia caballi in northeast China has remained unknown, although the People’s Republic of China is recognized as an endemic country for the diseases. In the present study, we investigated the prevalence of equine piroplasmosis in Jilin province, a part of northeast China. A total of 111 serum samples were taken from horses in eastern Jilin, and examined for diagnosis of Babesia equi and Babesia caballi infections by the enzyme-linked immunosorbent assays with recombinant antigens, equi merozoite antigen-1 and P48, respectively. Of the 111 samples, 38 (34%) and 36 (32%) samples were sero-positive for B. equi and B. caballi infection, respectively. In addition, 14 (12%) samples were sero-positive for both B. equi and B. caballi infections. These results indicate that equine piroplasmosis is widespread and therefore a cause for serious concern in northeast China.

KEY WORDS: Babesia caballi, Babesia equi, ELISA.

Equine piroplasmosis is caused by two tick-borne haemoprotozoan parasites, Babesia equi and Babesia caballi. The diseases are endemic in most tropical and subtropical areas of the world as well as in some temperate zones [1]. Both B. equi and B. caballi are usually detected in blood smears only during the acute stage of the infections, and animals recovered from the diseases become carriers of the parasites. Therefore, the detection of specific antibodies to parasites is the useful way to diagnose the parasite carriers or previously exposed horses. The complement fixation test (CFT) and indirect fluorescent antibody test (IFAT) are commonly used for detecting B. equi and B. caballi infections. However, these serological tests are generally restricted by antibody detection limits and cross reactivity [1, 4]. Previously, we have developed enzyme-linked immunosorbent assays (ELISAs) using recombinant antigens, and demonstrated that the ELISAs can be used as an alternative to CFT and IFAT for diagnosis of B. equi and B. caballi infections, respectively [2, 3, 5, 6]. In the present study, we investigated the prevalence of equine piroplasmosis in Jilin province of the People’s Republic of China by the ELISAs using recombinant antigens.

The ELISA with recombinant equi merozoite antigen-1 (EMA-1) expressed in insect cells by baculovirus for diagnosis of B. equi infection in horses was performed as described previously [5]. Briefly, recombinant EMA-1 was purified from culture medium of baculovirus AcEMA-1-infected Sf9 cells, and used as an ELISA antigen for detection of antibody to B. equi in horses. The ELISA titer was expressed as the reciprocal of the maximum dilution that showed an optical density value at 415 nm equal to or greater than 0.1, which is cut off value in absorbance between values for the EMA-1 antigen and control antigen (lacZ). The ELISA using recombinant P48 protein expressed in Escherichia coli by pGEX vector for diagnosis of B. caballi infection in horses was carried out as described elsewhere [2, 3]. Briefly, recombinant P48 protein was purified using glutathione Sepharose 4B beads, and used as an ELISA antigen for detection of antibody to B. caballi in horses. The ELISA titer was expressed as the reciprocal of the maximum dilution that showed an optical density value at 415 nm equal to or greater than 0.2, which is the difference in absorbance between values for the P48 antigen and control antigen (GST). A total of 111 serum samples were taken from horses in three villages in eastern area of Jilin province, China, on July of 2002. No apparent clinical signs were observed on all the sampled horses by macroscopic examination.

Jilin province lies in northeast China, shares borders with Inner Mongolia province and Heilongjiang province to the north, Liaoning province to the west, North Korea to the south, and Russia to the east (Fig. 1). The climate is predominantly dry and continental, with warm summers and cold winters. The average temperature is 22.0°C in summer (July) and 16°C below zero in winter (January). To date, there is no report about prevalence of equine piroplasmosis in Jilin province, although the serious prevalence of the diseases is reported recently in Xinjiang province, northwest China [6].

As shown in Tables 1 and 2, of the 111 samples, 38 (34%) and 36 (32%) samples were positive for B. equi infection and B. caballi infection by the ELISAs, respectively. The ELISA antibody titers to both B. equi and B. caballi are
The equine piroplasmosis is considered to be endemic in northeast China, Jilin province, which has a distance about 5,000-kilometer from Xinjiang province. There are only a few previous reports on the prevalence of equine piroplasmosis in China [6, 7]. Recently, we have reported that the prevalence of equine piroplasmosis in northwest China, Xinjiang province was serious [7]. In the present study, we have investigated the prevalence of equine piroplasmosis in northeast China, Jilin province, which has a distance about 5,000-kilometer from Xinjiang province. Our data may be considered as important information that would contribute to understand the prevalence of equine piroplasmosis not only in northeast China, but also in countries of neighboring the Jilin province.

The equine piroplasmosis is considered to be endemic because all 111 horses were locally, and showed no clinical signs. Since these horses are constantly under exposure of B. equi and B. caballi infections, they may acquire comparatively high immunity against both diseases.

Controlling the possible tick vectors is considered as an easy way to reduce the infection rate and to improve the quality of horse populations in endemic areas. The tick vectors for equine piroplasmosis in Jilin province are remained unclear to date, therefore there is a need to identify the potential tick vectors could involve in the transmission of both B. equi and B. caballi in horses in Jilin province.

In the present study, the recombinant antigens, EMA-1 and P48, were expressed by the genes from B. equi USDA strain and B. caballi USDA strain, respectively, both isolated in the U.S.A. It is not clear whether or not the recombinant antigens reacted with anti-EMA-1 or anti-P48 antibody completely in horses in Jilin province. Therefore, there is a need to identify the potential tick vectors could involve in the transmission of both B. equi and B. caballi in horses in Jilin province.

The present study, the recombinant antigens, EMA-1 and P48, were expressed by the genes from B. equi USDA strain and B. caballi USDA strain, respectively, both isolated in the U.S.A. It is not clear whether or not the recombinant antigens reacted with anti-EMA-1 or anti-P48 antibody completely in horses in Jilin province infected with B. equi or B. caballi, respectively, as any genetic information on the Jilin isolates is lacking. Possible genetic diversity of the EMA-1 and P48 genes between the USDA strains and Jilin isolates may affect the detection levels of equine piroplasmosis in Jilin province. Therefore, there is a need to compare the EMA-1 and P48 genes of the USDA strains to those of the Jilin isolates in the future.

This study was supported in part by the grants from regional Research Institute of Agriculture in the Pacific, College of Bioresource Sciences, Nihon University and from the Ministry of Education, Culture, Sports, Science and Technology of Japan.

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