

Research Note

Seroprevalence of Bovine Leukemia Virus (BLV) Infection in Pregnant Replacement Dairy Heifers in Saraburi Province, Thailand

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ABSTRACT

Seroprevalence of bovine leukemia virus (BLV) infection was studied in replacement dairy heifers. Blood samples were collected from 80 pregnant heifers raised in 8 dairy farms in Saraburi province, the central part of Thailand and serum samples were tested for antibodies against BLV infection using commercially available ELISA test kits. The results revealed that 26 (32.5%) pregnant heifers were positive reactors, which was higher than seroprevalence of BLV in lactating dairy cows previously reported in Thailand. This study provided an evidence of a high seroprevalence of BLV in replacement heifers raised in areas of Saraburi province. It was crucial to the dairy farmers to be aware of this disease in replacement heifer rearing. Although serological test does not provide information of the infection at the early stage and is not sensitive enough to detect every infected animals, elimination of seropositive animals and prohibition of introduction of seropositive animals are sufficient to promote BLV-free herds in Thailand.

Key words: bovine leukemia virus, replacement dairy heifer, seroprevalence

INTRODUCTION

Bovine leukemia virus (BLV) is an oncogenic lymphotropic retrovirus that causes persistent lymphocytosis and lymphosarcoma described as enzootic bovine leukosis (Miller *et al.*, 1969). The disease is classified into three stages; serologically positive without lymphocytosis; serologically positive with persistent lymphocytosis; and leukemia (Kabeya *et al.*, 2001). The disease may also have an asymptomatic course, in which the infected cattle remain as carriers without demonstrating any signs

of the disease (Trono *et al.*, 2001). The importance of BLV is based on the economic losses primarily due to condemnation of cattle with leukosis, reduction of milk production, and loss of trade opportunities with countries restricting the importation of BLV-infected cattle (Martin *et al.*, 2001). Bovine leukemia virus may be transmitted both vertically and horizontally; however, horizontal transmission is considered to be more important, usually occurring through poor management and manipulation (Martin *et al.*, 2001). Vertical transmission occurs when infected cows transmit the virus to the calf, either during

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pregnancy or through colostrum and milk (Martin *et al.*, 2001). Once infection has occurred, 80% to 90% of animals become BLV-seropositive. Of these infected cattle, 30% will develop persistent lymphocytosis, which occurs in cattle from 2 to 6 years of age and persists throughout life (Kaczmarczyk *et al.*, 2005).

Serological surveys have demonstrated the presence of seropositive cattle in Thailand. Bunyahotra *et al.* (1994) reported that average prevalence of BLV in dairy cattle in the Central part of Thailand was 13.39%, and the highest prevalence was observed in Lopburi Province. Wongkasemjit *et al.* (1994) also showed that average prevalence of BLV in dairy cattle in the Central region was 4.96%, and the prevalence of Saraburi was 10.2%. The serum samples of these two studies were tested using agar gel immunodiffusion assay. Arunvipas *et al.* (1997) studied BLV infection in dairy cattle using immunodot blotting assay and the results showed that 22.2% of 789 serum samples were BLV positive. The above studies were performed in dairy cattle aged over 2 years, with limited number of dairy heifers. Because replacement heifers play a key role on profitability of dairy farms and little is known about the prevalence of BLV infection in heifers in Thailand, the aim of this study was therefore to determine the seroprevalence of BLV infection in replacement heifers using commercially available ELISA test kit.

MATERIALS AND METHODS

Serum sampling

Blood samples were collected from 80

pregnant heifers raised in dairy farms in Saraburi Province. Blood samples without anticoagulant were allowed to clot at room temperature and were centrifuged at 1200×g for 15 min within 6 hours after collection. Serum samples were separated and stored at -20°C until analysis.

Serological analysis

Serum samples were analyzed for the presence of antibodies to BLV infection using the Bovine Leukemia Virus Antibody Test Kit (IDEXX HerdChek Anti-BLV; IDEXX Laboratories, Westbrook, USA) according to the manufacturer recommendation. The kit showed a high correlation with agar gel immunodiffusion test (kappa value of 0.998) and a total negative and positive agreement of 99.86 and 100% respectively. The positive test result was set at the cut-off point of serum to positive (S/P) ratio greater than 0.5.

Statistical analysis

Data were described using descriptive statistics (Patrie and Watson, 1999).

RESULTS AND DISCUSSION

The result of seroprevalence of BLV infection analyzed in 80 serum samples is demonstrated in Table 1. According to the cut-off point of S/P ratio, 26 (32.5%) pregnant heifers were BLV positive and 54 (67.5%) were BLV negative.

A high seroprevalence of BLV infection in dairy cattle also observed in other countries (Uysal *et al.*, 1998; Trono *et al.*, 2001).

Table 1 Seroprevalence of bovine leukemia virus infection in 80 replacement pregnant dairy heifers using Bovine Leukemia Virus Antibody Test Kit (IDEXX HerdChek Anti-BLV).

Test Result	Average S/P ratio	Number of heifers	Percentage
Negative	-0.003	54	67.5
Positive	2.604	26	32.5
Total		80	100

Seroprevalence observed in the present study was higher than the previous reports in Thailand (Bunyahotra *et al.*, 1994; Wongkasemjit *et al.*, 1994; Arunvipas *et al.*, 1997). Most serum samples from those earlier studies were collected from dairy cattle of ages ranging from 1 to 10 years, with predominantly over 2 - 5 years. The study of Arunvipas *et al.* (1997) used immunodot blotting assay to detect BLV antibody in serum samples, whereas the studies of Wongkasemkij *et al.* (1994) and Bunyahotra *et al.* (1994) used agar gel immunodiffusion assay. Our study used ELISA test, which provided at least the same quality of the test as agar gel immunodiffusion assay (Simard *et al.*, 2000). Although infection of BLV in these replacement heifers was high, clinical disease or lymphosarcoma was uncommon. However, atypical sporadic lymphosarcoma caused by BLV infection was reported (Dubreuil *et al.*, 1998; Hendrick, 2002; Nasir, 2005). Calves receiving colostrum from infected cows may remain seropositive to BLV for 6 months of age (Burridge *et al.*, 1982). All heifers in our study were pregnant and older than 20 months, meaning that our positive results from ELISA test were originated from natural infection.

In conclusion, this study provided an evidence of a high seroprevalence of BLV infection in areas of Saraburi province, despite a small number of samples. Although serological test does not provide evidence of the infection in its early stage and is not sensitive enough to detect every infected animals, elimination of seropositive animals and prohibition of introduction of seropositive animals are sufficient to promote BLV-free herds.

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