

## Short communication

## The recent prevalence of bovine leukemia virus (BLV) infection among Japanese cattle

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## ABSTRACT

A seroepidemiological survey of bovine leukemia virus (BLV) infection was conducted in Japan in 2007 using an enzyme-linked immunosorbent assay (ELISA) and an agar gel immunodiffusion (AGID) test. A total of 5420 cattle (dairy, 3966; breeding beef, 797; fattening beef, 657) from 209 farms in seven prefectures in Japan were tested. The overall prevalence of BLV infection was 28.6%. The prevalence of BLV infection in dairy cattle (34.7%) was higher than for both fattening beef cattle (7.9%) and breeding beef cattle (16.3%). Age-specific prevalence showed that BLV prevalence increased with age in all types of cattle and was notably different between dairy and beef cattle under 1 year of age. Among 207 farms, 141 herds (68.1%) had one or more positive animals. The proportion of these positive farms was significantly higher among dairy farms (79.1%) than among beef breeding farms (39.5%) and beef fattening farms (51.9%) ( $P < 0.001$ ). Dairy farms (40.5%) also showed a significantly higher within-herd prevalence than beef breeding (27.4%) and fattening (14.9%) farms ( $P = 0.001$ ). This study indicated that BLV is more widely spread in dairy cattle than in beef breeding cattle in Japan. Given the prevalence of BLV infection in dairy and beef cattle was 8- and 1.7-fold higher, respectively, than rates previously found in 1980–1982, BLV appears to be spreading particularly among the dairy cattle population during the last two decades. Further investigation is required to determine the risk factors necessary to control BLV infection that take into account the different farming practices that exist between dairy and beef sectors.

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## 1. Introduction

Bovine leukemia virus (BLV) is the causative agent of enzootic bovine leukosis (EBL), a neoplasm of lymphatic tissue in bovine species (Kettmann et al., 1994). BLV is classified into the genus Deltaretrovirus in the family Retroviridae (Hunter et al., 2000). The majority of infected animals remain healthy with no apparent negative

economic effects, but some BLV carriers develop a form of the disease known as persistent lymphocytosis (PL) and a low percentage of BLV-infected animals develop lymphoid tumors (Kettmann et al., 1994). BLV infection has a worldwide distribution (Burny et al., 1980; Schwartz and Levy, 1994) and seroepidemiological studies have indicated higher prevalence in some countries (Bause et al., 1978; Cockerell et al., 1992). EBL was successfully eradicated in some countries through national control programs in Europe in recent years (Acaite et al., 2007; Nuotio et al., 2003).

EBL is specified as a notifiable disease and has been subject to passive surveillance in Japan since 1998. There were 838 outbreaks of EBL on 677 farms in 2007, whereas there were only 159 outbreaks on 157 farms in 2000

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**Table 1**  
Size of tested farms by farm type.

	Farm		
	Dairy	Breeding	Fattening
Number of herd	139	43	27
Mean herd size	57.8	81.3	537.7
Range	4–290	6–1050	16–5800
Standard error	3.9	26.1	214.8
Median	46	30	200

according to the animal hygiene statistics of Japan (MAFF, 2008). However, a nationwide serological survey of antibodies against BLV has not been performed since 1982. The objective of this study was to estimate the recent prevalence of BLV infection in Japan. Moreover, the herd prevalence and within-herd prevalence in each sector (dairy and beef) were also investigated.

## 2. Materials and methods

### 2.1. Study area and population

The present study was performed on both dairy and beef farms in seven Japanese prefectures. These prefectures were selected to cover a wide geographical area in Japan.

Blood samples were obtained from farms on a voluntary basis from June to December 2007. On each farm, 30 cattle were randomly selected using the random number generator in Microsoft Excel 2003 (Microsoft, Redmond, WA, USA). When the herd size was less than 30, all cattle were sampled. Under these criteria, a total of 5420 cattle (3966 on dairy farms, 797 on breeding farms and 657 on fattening farms) from 209 farms (139 dairy, 43 breeding beef and 27 fattening beef) were subjected to testing. The sizes of tested farms are shown by farm type in Table 1.

### 2.2. Serological tests

Indirect enzyme-linked immunosorbent assay (ELISA) was performed on commercially available microplates for the detection of the anti-BLV gp51 antibody according to the manufacturer's instructions (Institut Pourquier, Montpellier, France). The sensitivity and specificity of the ELISA test were 99.0 and 99.6%, respectively (Acaite et al., 2007). Optical density (OD) values were determined with an Immuno Mini NJ-2300 ELISA reader (InterMed, Tokyo, Japan). Both positive and negative control samples were provided in the kit. When false-positive results were suspected by field veterinarians, samples were re-tested by agar gel immunodiffusion (AGID) test. The procedure for the AGID test was previously described (Kono et al., 1982).

### 2.3. Statistical analysis

Animal prevalence (the proportion of positive animals among the tested animals), herd prevalence (the proportion of farms with one or more positive animals among tested herds) and within-herd prevalence (the proportion

**Table 2**  
Seroprevalence of BLV among dairy and beef cattle.

Cattle	Total	Positive	Prevalence (%)	95% Confidence interval (CI)
Dairy	3966	1375	34.7	33.2–36.2
Beef	1454	173	11.9	10.2–13.6
Breeding	797	120	15.1	12.6–17.5
Fattening	657	53	8.1	6.0–10.1
Total	5420	1548	28.6	27.4–29.8

of positive animals on each seropositive farm) were statistically examined. For within-herd prevalence, only herds with 10 or more cattle were analyzed.

The chi-square test was used to compare the animal and herd prevalence between dairy and beef cattle. The Mann–Whitney *U*-test was used to evaluate the effect of herd size between positive and negative farms. The Kruskal–Wallis test was used for the within-herd prevalence to test the difference among farm types (dairy, breeding beef and fattening beef).

## 3. Results

The data presented in Table 2 show that 28.6% (95% confidence interval (CI): 27.4–29.8%) of cattle were positive among a total of 5420 cattle tested. The prevalence of BLV infection among animals was 34.7% (95% CI: 33.2–36.2%) in dairy cattle and 11.9% (95% CI: 10.2–13.6%) in beef cattle, whereas in breeding and fattening beef cattle prevalence was 15.1% (95% CI: 12.6–17.5%) and 8.1% (95% CI: 6.0–10.1%), respectively. The animal prevalence in dairy cattle was 2- and 4-fold higher than for beef breeding and fattening cattle, respectively ( $P < 0.0001$ ).

Age-specific prevalence by each type of cattle is shown in Fig. 1. Approximately 16% of dairy cattle had already been infected with BLV when they were less than 1 year old. On the other hand, breeding and fattening beef cattle under 1 year old typically showed a lower prevalence. The prevalence in adult cattle (>2 years old) on dairy farms was 37.5% (95% CI: 36.7–38.3%) and significantly higher than that in cattle less than 2 years old (19.3%, 95% CI: 17.7–20.8%). The prevalence in adult (>2 years old) and young breeding cattle (<2 years old) was 18.4% (95% CI: 16.8–19.9%) and 4.5% (95% CI: 2.9–6.0%), respectively.

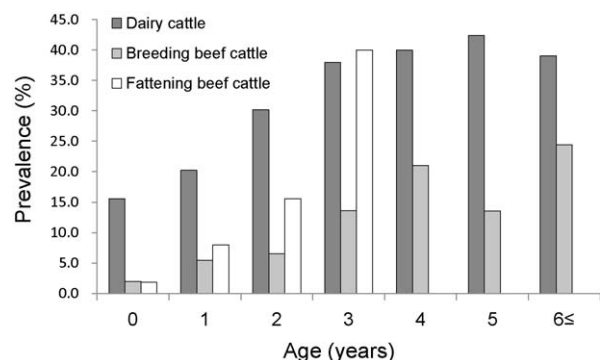


Fig. 1. Age-specific BLV prevalence by cattle type.

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