

Bovine Leukemia Virus Infection in Taiwan: Epidemiological Study

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(Received 18 September 1990/Accepted 17 January 1991)

ABSTRACT. We conducted a seroepidemiological survey for antibodies to bovine leukemia virus (BLV), by using agar gel immunodiffusion technique, in dairy cows, water buffaloes, and yellow cattle throughout Taiwan. The positive reactors were 8.4% (376/4,459) in 1985 and 5.8% (1,277/22,190) in 1986, in 15 prefectures and 7 cities. Relatively high infection rate appeared in the northern and southern areas of Taiwan. Positive reactors increased gradually with age. The incidence of positive antibodies was 2 to 3 times higher in pasture-style farms than in housed-style farms. Among the 6,313 imported cattle, 302 (4.8%) showed positive reaction. Between 1985 and 1987, 5 cattle showed enzootic bovine leukosis among 351 sero-positive reactors in four highly positive prefectures. Survey of 134 water buffaloes and yellow cattle showed no positive reactors. This survey demonstrated that BLV-infection has increased over the years and spread throughout Taiwan.—**KEY WORDS:** bovine leukemia virus, enzootic bovine leukosis, epizootiological survey, Taiwan.

J. Vet. Med. Sci. 53(3): 395–398, 1991

Bovine leukemia virus (BLV), an exogenous retrovirus, is capable of causing enzootic bovine leukosis (EBL) [7]. Clinical signs of BLV-infection include lymphocytosis and lymphoid tumors, following a long period of incubation, during which the presence of antiviral antibodies in the serum is the only common feature [15]. Several serological methods have been designed and used for detection of infected animals. Among them agar gel immunodiffusion (AGID) technique gained wide acceptance [8,13], because of its simplicity and reliability. Serological surveys provide the basis for an eradication policy.

Seroepizootiological surveys using AGID test have described the spread of BLV-infection in many countries [2, 3, 5, 11, 17]. In Taiwan, Wang and his associates [19] conducted the first serological survey in 1979–1980 and showed an infection rate of 1.9%. These BLV-positive cattle appeared in farms located in the northern and southern areas of Taiwan. However, the incidence of BLV-infection among clinically normal cattle has not been studied in detail. In this report, we summarize the results of the second survey in 1985–1989 on distribution of BLV-infected cattle using gp and p antigens and the incidence of clinical manifestation in Taiwan.

MATERIALS AND METHODS

Sera: We collected random serum samples from 29,469 dairy cattle of 1–12 years old in 6 cities and 15 prefectures in 1985 and 1986. A total of 4,390 serum samples were also collected from 6 dairy farms with

high infection rate (Farms A and B in Taoyuan and Hsintsu Prefecture in northern Taiwan; Farms C, D, E and AF in Tainan Kaohsiung Prefecture and Kaohsiung City in southern Taiwan). Records on importation and relocation were available for these 6 farms. To determine the source of BLV-infection, we also randomly selected draft cattle (134 water buffaloes and 142 yellow cattle) ranches in nearby BLV-infected farms in 5 prefectures in 1986. These samples were obtained by the regional livestock disease control centers.

Herd style: Farms A, B, and C were pasture-style farms. All other farms raised their cattle with modern management, in which dairy cattle were housed in stall throughout the year.

AGID test: We carried out AGID tests with gp and p antigens to BLV as described previously [13, 20].

RESULTS

Table 1 shows the results of the present survey for antibodies against BLV. The overall positive reactors were 8.4% (376/4,459) in 1985 and 5.8% (1,277/22,190) in 1986. During the one year observation period in 1986, the percent of positive reactors in 5 cities and 15 prefectures varied from 0 to 60.4%. High positive rates were observed in Taoyuan and Hsintsu Prefecture in the north, and Tainan and Kaohsiung Prefecture and Kaohsiung City in the south. The infection rates showed above were higher than the rate (1.9%) reported in the 1979–1980 survey [19]. Figure 1 shows map of

Table 1. Comparison of results of the first (1979) and the second (1985–1986) serological surveys on BLV antibodies in cattle in Taiwan

Locations	1979 ^{a)}		1985		1986	
	Reacted/Tested(%)		Reacted/Tested(%)		Reacted/Tested(%)	
Prefectures						
Taipei	—		3/552	(0.5)	92/1,301	(7.1)
Taoyuan	11/255	(4.3)	89/512	(17.4)	15/1,242	(1.2) ^{b)}
Hsintsu	—		31/116	(26.7)	36/251	(14.3) ^{b)}
Miaoli	—		34/597	(5.7)	18/1,185	(1.5) ^{b)}
Taichung	0/39	(0)	—		39/906	(4.3)
Changhwa	0/19	(0)	—		44/1,057	(4.2)
Nantu	—		18/300	(6.0)	27/618	(4.4)
Yunlin	0/42	(0)	10/500	(2.0)	46/2,485	(1.9)
Chiayi	0/74	(0)	8/60	(13.3)	135/1,961	(6.9)
Tainan	0/48	(0)	18/450	(4.0)	197/4,061	(4.9)
Kaohsiung	—		—		187/2,678	(7.0)
Pintung	—		4/100	(4.0)	238/2,719	(8.8)
Yilan	—		0/50	(0)	0/7	(0)
Hualian	0/48	(0)	2/80	(2.5)	4/132	(3.0)
Taitung	0/102	(0)	12/278	(4.3)	9/358	(2.5)
Cities						
Keelung	0/18	(0)	—		—	
Taipei	0/193	(0)	2/49	(4.1)	11/170	(6.5)
Hsintsu	8/42	(19.0)	112/254	(44.1)	84/139	(60.4)
Taichung	0/34	(0)	—		4/141	(2.8)
Chiayi	—		6/104	(5.8)	—	
Tainan	0/5	(0)	12/325	(3.7)	39/509	(7.7)
Kaohsiung	1/115	(0.9)	15/132	(11.4)	52/270	(19.3)
Total	20/1,034 (1.9)		376/4,459 (8.4)		1,277/22,190 (5.8)	

a) Cited from Wang and Chang [19].

b) Some of the antibody-positive cattle were eliminated previously.

examined areas in Taiwan.

Table 2 shows the results in 6 farms with high rate of infection during the period between 1979 and 1987. Antibody positive reactors in Farm A ranged from 18.1 to 46.4%, and Farm B from 18.4 to 54.2%. The rate of infection rose by 2 to 3 fold during 9 years. In Tainan, the infection rate reached 59.5% in 1986 in Farm C, and 30.3% in 1987 in Farm D. In Farm D, 140 of the BLV-positive cattle were eliminated since 1988, thereby infection rate reduced from 23.8% in 1988 to 3.7% in 1989. In Kaohsiung City, BLV incidence in Farm AF increased from 11.8% in 1979 to 19.3% in 1986. During 2 years observation period, the rates of conversion from BLV-negative to positive in the two pasture-style farms were 12.0% (11/92) in Farm A and 15.8% (16/101) in Farm B. On the other hand, it was only 5.0% (4/80) in the housed-style Farm D. The rate of seroconversion differed by 2 to 3 fold in the two farming styles.

Infection rates were analyzed according to age groups for the year 1986 as shown in Table 3. A relatively few reactors (6.4%) were found in cattle

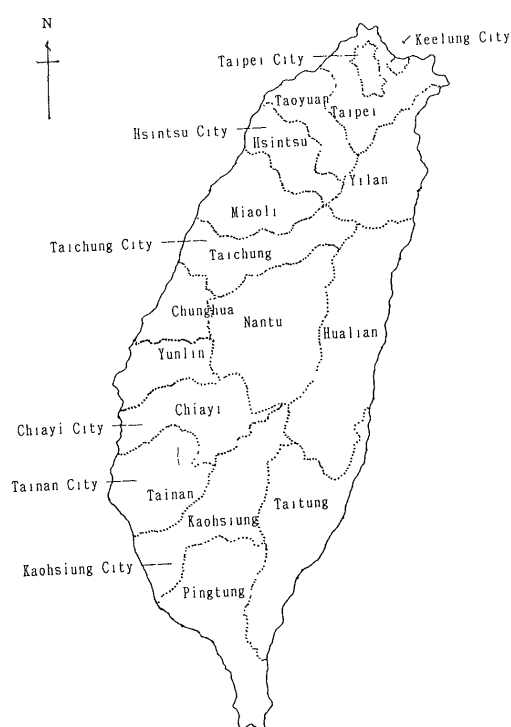


Fig. 1. Map of examined areas in Taiwan.

less than 2 years old, but more than 40% (41.4–48.7%) of 5 years and older cattle were infected.

Among 22,190 serum samples obtained in 1986, 1,277 (5.8%) showed seropositive. The percent BLV-positive was 6.1% (975/15,877) in the domestic-produced and 4.8% (302/6,313) in the imported cattle. Eighty percent of the imported BLV-positive cattle were aged between 1 to 4 years. The infection rates varied with countries imported; U.S.A. 5.1% (249/4,930), Japan 4.8% (21/436), Australia 4.1% (31/760), and New Zealand 0.5% (1/187). None of the sera obtained from draft cattle (134 water buffaloes and 142 yellow cattle) in 5 southern and eastern prefectures were positive for BLV antibodies.

Occurrence of EBL was reported in 5 out of the 256 BLV-positive cattle during 2 years observation period in the northern (Taoyuan, Hsinchu, and Miaoli Prefecture) and southern (Kaohsiung City) areas. The infection rate in these areas ranged from 11.8 to 44.1%. Of the 5 cattle of 7 to 12 years of age, two showed enlargement of superficial lymph nodes, one with visceral lymph nodes, and two with eyeball protrusion and tumor in the orbit. No EBL cattle was reported in the low infection (0.5–2%) regions, Taipei, Yunlin, and Hualian Prefecture.

DISCUSSION

Our survey showed the wide-spread existence of BLV-infection and the increasing trend over the years in Taiwan. The infection rate was higher than the rate (1.9%) previously reported in the

1979–1980 survey [19]. Furthermore, the prevalence of EBL seems to be correlated with the rate of BLV-infection [2, 11].

Style of farming affects the rate of infection and the rate of transition from BLV-negative to positive as evidenced by the 2 to 3 fold difference between cattle raised in pastures and those kept in stalls in this and other reports [6, 12, 14]. The difference indicates the importance of blood-sucking Tabanid flies in the transmission of BLV [1, 10]. We previously have found 3 varieties of Tabanid flies in pasture-style farms [4].

Although Romero *et al.* [16] demonstrated the experimental susceptibility of buffalo to BLV, our survey showed no natural infection of BLV in 134 buffaloes. Both water buffaloes and yellow cattle showed BLV-negative in the 1979–1980 [19] and the present surveys, despite grazed around infected farms. This fact implicates imported dairy cattle as the source of BLV-infection in Taiwan. We suspect other domestic animals may also be infected with BLV. In Tainan, one in 267 milk goat tested showed positive reaction to BLV antigens, in 1987. Further

Table 3. Relationship between BLV-infection and ages in farms A, B, C, E, and AF

Age (yr)	Reacted/Tested (%)
1–2	16/250 (6.4)
3–4	42/302 (13.9)
5–6	98/237 (41.4)
7–8	101/208 (48.6)
>8	113/232 (48.7)
Total	369/1,229 (30.0)

Table 2. Results of antibody tests in cattle of the areas with high infection rate

Area	Farm	1979	1982	1984	1986	1987	1988	1989
Northern:								
Taoyuan	A	49/271 ^{a)} (18.1)	38/220 (17.3)	76/223 (34.1)	69/174 (39.7)	26/56 (46.4)	–	–
Hsintsu	B	33/179 (18.4)	55/106 (51.9)	78/198 (39.4)	112/254 (44.1)	122/225 (54.2)	–	–
Southern:								
Tainan	C	–	–	–	44/74 (59.5)	–	–	–
	D	–	–	–	–	89/294 (30.3)	160/674 ^{b)} (23.8)	20/537 ^{b)} (3.7)
Kaohsiung	E	–	–	–	92/457 (20.1)	–	–	–
	AF	12/102 (11.8)	–	8/76 (10.5)	52/270 (19.3)	–	–	–

a) Numbers are Reacted/Tested (%).

b) One hundred and forty BLV-positive cattle were eliminated in previous years.

investigations are suggested to find out the relationships between feeding style, management, and genetical factors.

The reasons why the wide-spread BLV-infection in Taiwan might be caused by a) importation of infected cattle from foreign countries without testing and b) frequent trade and relocation of these infected animals in Taiwan. Importation of cattle can be traced back to 1960 in four prefectures where the infection rate was high. The rate of BLV-infection increased gradually since 1960. We found the high prevalence of BLV-infection in farms where the imported cattle had introduced. However, in the BLV-free farms in these areas, there was no record of introduction of imported cattle. Furthermore, no BLV-infected cattle can be detected in native cattle in these areas. Therefore, the first occurrence of BLV-infection may be caused by introduction through importation as suggested in other countries [9, 18] and then widely distributed in Taiwan by frequent trades. The present survey suggests the importance of quarantine procedures for cattle importation.

ACKNOWLEDGEMENTS. This investigation was supported by the Council of Agriculture. We thank Prof. S. Namioka and Prof. M. Onuma, of the Faculty of Veterinary Medicine, Hokkaido University; and Prof. H. Koyama of the Department of Veterinary Microbiology, Kitasato University, Japan; Prof. C. H. Chang of the Department of Veterinary Medicine, National Taiwan University; and Dr. T. C. Lin, Chief of Animal Health Division, Council of Agriculture, Taiwan, for their encouragement. We also wish to express our appreciation to Livestock Disease Control Centers in Taiwan for collecting serum samples.

REFERENCES

1. Bech-Nielsen, S., Piper, C. G., and Ferrer, J. F. 1978. Natural mode of transmission of the bovine leukemia virus; Role of blood-sucking insects. *Am. J. Vet. Res.* 39: 1089-1092.
2. BurrIDGE, M. J., PuhR, D. M., and Hennemann, J. M. 1981. Prevalence of bovine leukemia virus infection in Florida. *J. Am. Vet. Med. Assoc.* 179: 704-707.
3. Burton, R. W., Hoare, R. J., and Grewal, A. S. 1986. Bovine leukosis virus: antibody in cattle in New South Wales. *Aust. Vet. J.* 63: 380-381.
4. Chang, H. C. and Wang, C. T. 1983. Studies on the disease control of cattle: Production of bovine leukemia virus antigen and seroepizootiological survey on bovine leukemia virus infection in Taiwan. pp. 11-17. In: Coun. Agr. Res. Proj. No. 72 (Chiu, S. Y. ed.), Council of Agriculture, Taipei, Taiwan.
5. Dufour, B. 1989. Epidemiological situation of enzootic bovine leukosis in France at the end of 1988. pp. 31-41. In: G. D. S. Inform. San. No. 95. Paris, France.
6. Kaaden, O. R., Neth, R., and Frenzel, B. 1978. Sequential studies of bovine leukemia virus antibody development in dairy cattle over a four-year period. *Ann. Res. Vet.* 9: 771-776.
7. Miller, J. M., Miller, L. D., Olson, C., and Gillette, K. G., 1969. Virus-like particles in phytohemagglutinin stimulated lymphocyte culture with reference to bovine lymphosarcoma. *J. Natl. Cancer Inst.* 43: 1297-1305.
8. Miller, J. M. and Olson, C. 1972. Precipitating antibody to an internal antigen of the C-type virus associated with bovine lymphosarcoma. *J. Natl. Cancer Inst.* 49: 1459-1462.
9. Ministry of Agriculture, Fisheries and Food, U. K. 1979. Enzootic Bovine Leucosis—a preliminary survey. pp. 8-11. In: Animal Disease Report, No. 3. Central Vet. Lab., New Haw, Surrey, U. K.
10. Ohshima, K., Okada, K., Numakunai, S., Yoneyama, Y., Sato, S., and Takahashi, K. 1981. Evidence on horizontal transmission of bovine leukemia virus due to blood-sucking Tabanid flies. *Jpn. J. Vet. Sci.* 43: 79-81.
11. Onuma, M., Honma, T., Mikami, T., Yoshikawa, H., and Yoshikawa, T. 1978. Survey for antibodies to bovine leukemia virus in dairy and beef cattle in Japan. *Jpn. J. Vet. Sci.* 40: 691-696.
12. Onuma, M., Ishihara, K., Ohtani, T., Honma, T., Mikami, T., and Izawa, H. 1979. Seroepizootiological survey on antibodies against bovine leukemia virus in Japanese black cattle. *Jpn. J. Vet. Sci.* 41: 601-605.
13. Onuma, M., Olson, C., Baumgartener, L. E., and Pearson, L. D. 1975. An ether-sensitive antigen associated with bovine leukemia virus infection. *J. Natl. Cancer Inst.* 55: 1155-1158.
14. Onuma, M., Watarai, S., Ishijo, S., Ishihara, K., Ohtani, T., Soneda, M., Mikami, T., Izawa, H., and Konishi, T. 1980. Natural transmission of bovine leukemia virus among cattle. *Microbiol. Immunol.* 24: 1121-1125.
15. Paul, P. S., Pomeroy, K. A., Castro, A. E., Johnson, D. M., Muscoplat, C. C., and Sorensen, D. K. 1977. Detection of bovine leukemia virus in B-lymphocytes by the syncytia induction assay. *J. Natl. Cancer Inst.* 59: 1269-1272.
16. Romero, C. H., Aguiar, A. A., Zanolchi, H. G., Abaracon, D., Rowe, C. A., and Silva, A. G. 1981. Susceptibility of the water buffalo (*Bubalus bubalis*) to enzootic bovine leukosis virus. *Pesquisa Vet. Brasil* 1: 137-140.
17. Singh, V. P., Bansal, M. P., and Singh, K. P. 1987. Seroepidemiological studies of bovine leukemia virus infection in Indian cross-breed Zebu cattle. *Rev. Sci. Tech. Off. Int. Epizooties* 6: 225-231.
18. Steck, F., Kupferschmied, H., Leemann, W., Kaderli, R., Bommeli, W., Gafner, P., and Spurri, H. K. 1979. Serological investigation on the occurrence of bovine leukosis in Switzerland. *Schweiz. Arch. Tierheilkd.* 121: 439-450.
19. Wang, C. T. and Chang, C. H. 1982. Seroepizootiological Survey on bovine leukemia virus infection in Taiwan. pp. 88-92. In: 3rd Cong. Fed. Asian Vet. Assoc. Reports (Lee, S. W. ed.), Korean Vet. Med. Assoc., Seoul, Korea.
20. Wang, C. T. and Chang, C. H. 1988. Agar gel Immuno-diffusion: An evaluation of the titer for bovine leukemia virus antigen. *J. Chinese Soc. Vet. Sci.* 14: 101-112.