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## STUDIES OF INHIBITORY FACTORS FOR MITOGEN-INDUCED BLASTOGENESIS OF LYMPHOCYTES IN SERA FROM CATTLE WITH LYMPHOMA

Terukazu ODAWARA

*Department of Epizootiology  
Faculty of Veterinary Medicine  
Hokkaido University, Sapporo 060, Japan*

*In vitro* mitogen-induced blastogenesis of lymphocytes from bovine leukemia virus (BLV)-free healthy cows were inhibited by sera from cows with enzootic bovine leukosis (EBL). Properties of the inhibitory factors and mechanisms of the factors to mitogen-induced blastogenesis of lymphocytes were examined. The activity of the factors in sera from EBL cows was expressed as the incorporation of <sup>3</sup>H-thymidine in cultured lymphocytes with mitogen and serum from EBL cows. The results were summarized as follows:

1) An addition of sera from cows with EBL to cultured lymphocytes from normal cows inhibited the blastogenic response to mitogens. Individual sera from EBL cows varied in their inhibitory activity even causing the stimulation index to decrease from the level found in normal sera. Heat inactivation at 56°C for 30 minutes did not affect the inhibitory activity. The pool of inhibitory sera showed the same inhibitory activity level as the individual sera.

2) The inhibitory effect was reversible when lymphocytes were preincubated with the inhibitory serum, followed by washing and reculturing with serum-free medium.

3) The inhibition of mitogen-induced blastogenesis by serum could not be overcome by addition of increasing amounts of phytohemagglutinin (PHA), indicating that this inhibitory effect might not be caused by inhibition of binding of PHA receptor of lymphocytes nor inactivation of PHA by factors.

4) Inhibitory factor of sera from cows with EBL to PHA-induced blastogenesis of lymphocytes may act on the lymphocytes in the G<sub>0</sub> to G<sub>1</sub> phases but not on those in the S phase.

5) Circulating immune complex (CIC) levels of sera from BLV-negative healthy cows and EBL cows were quantified by the polyethylene glycol turbidity method. Sera from EBL cows or those from EBL cows with glomerulo-nephritis showed significantly higher levels of CIC than that of normal control sera ( $p < 0.05$ ). The CIC of sera from EBL cows showed a slightly inhibitory effect to mitogen-induced blastogenesis of lymphocytes.

6) Separation and partial purification of sera from EBL cows showed that the inhibitory effect of mitogen-induced blastogenesis of lymphocytes consisted of several factors, and the highest inhibitory activity was associated in a high molecular region,  $\beta$ -lipoprotein, of serum protein.