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Title	The differences in the kinetic changes of lymphocyte populations and cytokine profiles between chickens genetically susceptible and resistant to Marek's disease
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infection.

The levels of TNF α and TNF α R mRNA were measured in sheep infected with BLV by using the reverse-transcriptase polymerase chain-reaction system. The levels of TNF α mRNA in sheep resistant to BLV expansion was higher than in susceptible sheep. On the other hand, the expressions of both TNF α R 1 and R 2 mRNA were detected in BLV-resistant sheep whereas only R 2 was expressed in susceptible sheep as well as KU-1,

a BLV tumor cell line. Furthermore, to investigate the effect of TNF α on the proliferative responses of lymphocytes, the proliferation assay was performed with PBMCs from BLV-resistant and susceptible sheep. Significantly higher lymphocyte proliferative responses were observed in BLV-susceptible than resistant sheep. These results suggest that clearance of BLV infection by BLV-resistant sheep was TNF α R 1 expression and not R2.

The differences in the kinetic changes of lymphocyte populations and cytokine profiles between chickens genetically susceptible and resistant to Marek's disease

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Genetic resistance or susceptibility to T cell lymphoma formation induced by Marek's disease virus (MDV) is associated with the major histocompatibility complex (MHC) haplotypes of chickens. It was hypothesized that in susceptible chickens the generation of activated CD 4⁺ T cells through intensive immune responses during the early phase of MDV infection provides a large pool of target cells for transformation. However, it remains unclear whether difference in the amounts of activated CD 4⁺ T cells can indeed determine the degree of resistance to MD *in vivo*. In addition, currently, only a few information is available for relationship between disease progression of Marek's disease (MD) and cytokine profiles in host chickens, though cytokine profiles have been reported to play an important role in the outcome of several infectious diseases. Thus, in this study, the differences in the kinetic changes of lymphocyte

populations and cytokine profiles were studied between genetically susceptible and resistant chickens during MDV infection.

The results obtained from the kinetic analysis of lymphocyte populations by flow cytometer were not in good agreement with the previous hypothesis. A transient increase in the number of CD 4⁺ cells was observed in both susceptible and resistant lines of chickens at 7 days post infection (pi), but no significant difference was shown in the degree of the increase between the two lines of chickens. Instead, more MDV was re-isolated from genetically susceptible chickens than resistant chickens. These results suggest that the differences in the numbers of infected cells and/or virus loads per cell, due to the differences in the numbers and/or affinities of virus receptors on the cell surface, can decide the degrees of susceptibility to MD during the early phase of infection. Since the numbers of

CD 4- and CD 8-positive cells were constantly lower in the peripheral blood of the susceptible line of chickens after 10 days pi, the severity of this continuous depression may also be a factor to determine susceptibility to MD.

The expression of cytokines, interleukin-2 (IL-2), interferon- γ (IFN- γ) and chicken myelomonocytic growth factor (cMGF) which is an avian homolog of mammalian interleukin-6, were also analyzed in both resistant and susceptible chickens by reverse-transcriptase polymerase chain reaction. The expression levels of IL-2 and IFN- γ appeared to be equal or suppressed compared to uninfected chickens followed by the enhancement

at 4 to 7 days pi., probably resulting from the immune suppression caused by MDV. Although the level of IFN- γ expression was higher in susceptible than in resistant chickens during MDV infection, the relationship between the level of IFN- γ expression and susceptibility to MD was not clarified in this study. No significant difference in the expression level of cMGF was observed between the two lines except for 4 days pi.

Thus, it would be necessary to study the differences between resistant and susceptible chickens for understanding the mechanism of lymphomagenesis.

Development of latex agglutination test
for the detection of *Echinococcus multilocularis*
coproantigens in the definitive hosts.

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In this study, the development of a simple and rapid latex agglutination test for the detection of *Echinococcus multilocularis* coproantigens in the definitive hosts was attempted. The optimal size of polybead carboxylated microspheres (latex beads), optimal monoclonal antibody and its reaction concentration for the sensitization of the latex beads were evaluated. It was determined that slide agglutination test was best performed using the latex beads (1.0 μm in diameter) sensitized with 50 $\mu\text{g}/\text{ml}$ of EmA 9 monoclonal antibody raised against somatic antigen of adult *E. multilocularis*. Different kinds of dilution buffer for the fecal samples were also evalu-

ated, and 1% Tween 20 in PBS was selected. The latex agglutination test (Test 1) was performed on 35 non-heated and 82 heated feces of wild foxes, resulting in 47% and 61% in sensitivity, and 94% and 86% in specificity, respectively. By heating fecal samples to inactivate parasite eggs, higher sensitivity was obtained but specificity was reduced. To improve the sensitivity of the coproantigen detection, a small amount of excretory/secretory antigen of adult *E. multilocularis* (EmES antigen) was added to dilution buffer of feces (Test 2). By addition of EmES antigen, the sensitivity increased to 82% and 91%, but the specificity decreased to 44% and 61%, respec-