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A BOVINE LEUKEMIA VIRUS (BLV) VACCINE :
INDUCTION OF CELL-MEDIATED IMMUNITY

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It is known that cell-mediated immunity (CMI) plays an important role in the protection against bovine leukemia virus (BLV) infection, and several experimental vaccines have been reported using the BLV envelope (BLV*env*) protein. In this study, induction of humoral and cell-mediated immunity, and the protective effect of the BLV*env* protein expressed by a recombinant baculovirus against BLV challenge were investigated in sheep. Protective effects of vaccination with synthetic multiple antigenic peptides (MAP) of T-helper (Th), T-cytotoxic (Tc) and B cell (B) epitopes of the BLV*env* protein reported previously, were also studied. Moreover, the effect of mannan-coated liposomes (M-Lip) containing the synthetic peptide was tested in terms of the induction of CMI in mice.

Spleen cells prepared from Balb/c mice immunized with M-Lip containing the BLV*env* 98–117 peptide (Th epitope) showed a high lymphocyte proliferative reaction after stimulation with either the 98–117 peptide or BLV virion. The culture supernatant contained high titers of interferon-gamma and interleukin (IL)-2, but not IL-4. These results indicate that vaccination with M-Lip containing the 98–117 peptide induces the activation of Th 1 cells, which are important to induce CMI. In addition, the splenic lymphocytes, which showed a high proliferative reaction, have a specific cytotoxic activity against a syngeneic cell line, A-31, primed with the peptide.

Two sheep immunized with the BLV*env* protein, expressed by a recombinant baculovirus, produced anti-BLV virion antibodies, but these antibodies did not show any neutralizing activity *in vitro*. Lymphocytes from these two sheep showed no proliferative response in the lymphocyte blastogenesis test. No protection against the BLV challenge was observed in these two sheep. Three sheep immunized with a mixture of peptides corresponding to Th-, Tc-, and B cell epitopes produced antibodies against the B cell epitope and these antibodies inhibited syncytium formation by BLV *in vitro*. Two of three sheep showed a high lymphoproliferative response. One of these sheep (No. 4) showed complete protection against the BLV challenge, whereas, the other sheep (No. 3) did not show complete protection, but showed specific CTL activity and the rapid clearance of BLV. However, despite producing antibodies, one sheep (No. 5), showed no lymphoproliferative response, and was not protected against the BLV challenge.

Together, these results suggest that M-Lip has effective adjuvant activity in the induction of CMI, and that synthetic peptides can be used for vaccination against BLV

infection. Thus, it is expected that the combination of M-Lip with either the synthetic peptides or highly purified recombinant BLV*env* protein could result in the development of a more effective BLV vaccine.