the different brain areas in samples obtained from bovine not was found statistically significant differences between areas, but in samples of horses was found a greater presence of these cells in the brain stem (p = 0.0266). We could also observe that the meningeal and perivascular environments were where we find more immunostaining for B lymphocytes. The detection of B lymphocytes in CNS samples of cattle and horses was more pronounced in meningeal and perivascular environments, suggesting that these cells would be entering the CNS by breaking the blood brain barrier, however, the induction of specific antibodies for viral clearance is delayed, occurring only when the animal already have in severe neuronal damage. Although we have detected B cells *in situ* in the samples studied, these cells were in small amounts mainly in samples of horses. The collaboration intercellular between CD4 + T lymphocytes and B lymphocytes for activation of these cells and consequently induction of specific antibodies to the virus may be impaired because CD4 + T lymphocytes when entering the CNS may undergo apoptosis through its association with infected neurons that up expressing FASL and bind to CD4 + T cells expressing FAS occurring so the death of these immune cells essential for protection against rabies virus. These findings are important for understanding how the immune response is manifested in these animal species and also to improve understanding of the pathogenesis of rabies in cattle and horses.

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PT.016
EFFICACY OF RECOMBINANT ADENOVIRUS EXPRESSING G PROTEIN OF RABIES IN MICE

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Background: Since rabies case occurred again in 1993, a number of animal rabies had been reported up to 2011. Even though animal rabies cases seemed to be decreased, the continuous outbreak was identified at some counties of Gangwon Provinces of Korea. Although national mass vaccination program with live and inactivated vaccines to domestic and pet animals has blocked dog-to-dog transmission, most of rabies cases are related with to animal bitten by rabid raccoon dogs and rabies in wild animals are not eradicated. A safe and effective vaccine is needed for the immunization of wild animals and dogs. Human adenoviruses have been studied as viral vector. In this study, we constructed three kinds of recombinant adenovirus expressing rabies proteins and checked efficacy of the constructs in mice. **Material and Methods**: Rabies virus (RABV) circulating in Korea was isolated using neuroblastoma cell (NG108-15) in 2009. The RABV designated as KRVB0910 strain was propagated in the NG108-15 cells for the cloning of genes. In order to analysis the glycoprotein (G) and nucleocapsid (N) genes of the strain, the G and N genes were amplified with three kinds of primers and cloned into pENTR/D-TOPO cloning vector respectively. After cloning three genes (Nfull, Gfull,G-TMCD), each plasmids containing the genes were transfected into TOP10 competent cells. The purified plasmids were mixed with pAd/CMV/V5-DEST gateway vector and the mixtures had reaction with LR Clonase II enzyme to catalyze the LR recombination reaction. After confirming the expression clones, the clones were digested with Pac I to expose the ITRs and transfected into the 293A cell lines to construct recombinant adenovirus (reAdV) expressing N and G genes of RABV. The 293A cells transfected with the clones showed specific cytophatic effect. For 6 days after inoculation, the cells were stained with monoclonal antibodies and FITC conjugated goat anti human IgG+IgM and examined by fluorescent microscopy. To check efficacy of three kinds of reAdVs, the reAdVs containing 108.0 TCIID50/ml was inoculated into 4 weeks old Balb/C mice. Survival rate and change of body weight of the mice were checked for 17 days after challenge. **Results**: We successfully reconstructed three kinds of reAdVs (Nfull, Gfull, G-TMCD) in 293A cells. The titer of reAdVs ranged from 107.7 to 108.0 TCID50/ml. Four groups of mice (Gfull, G-TMCD, Nfull+Gfull, Nfull+G-TMCD) were inoculated with 0.2 ml reAdV and half of mice in each group were challenged with CVSN2c strain intramuscularly 21 days after inoculation. All mice did not show any typical rabies symptoms and showed complete protection. On the other hand, half of mice in three groups (Gfull, G-TMCD, G-TMCD+Nfull) did not show complete protection against challenge by intracranial (IC) route. However, the one group inoculated with Nfull+Gfull reAdVs revealed 100% survival rate. These data demonstrated the potential of the reAdV as a safe rabies vaccine. **Conclusion**: We constructed three kinds of reAdVs in 293A cells. The combination of two kinds of reAdVs (Nfull+Gfull) may be a useful tool in search of rabies vaccine candidate for animals and further study related to oral vaccination of dogs and raccoon dogs is needed in the near future.

PT.017
INTERACTION OF RABIES VIRUS GLYCOPROTEIN FRAGMENTS WITH THE NICOTINIC ACETYLCHOLINE RECEPTOR

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The rabies virus glycoprotein (RVG) interacts with Torpedo and muscle nicotinic acetylcholine receptors (nAChR). The field of Ligand Gated Ion Channels, such as the nicotinic receptors, has benefited greatly over the last decade due to the discovery of non-membrane bound Acetylcholine Binding Proteins (AChBP). Since nicotinic acetylcholine receptors and the ACHBP share significant sequence and structural homology in the neurotoxin binding domain, the AChBP could provide a useful model for studying the molecular basis of the RVG/nAChR interaction. In this study we investigated the interaction between RVG neurotoxin like peptide fragments and the AChBP. Surface Plasmon resonance (SPR) was used to assess binding kinetics to the AChPB. Electrophysiology experiments were used to compare these results to interactions between these RVG fragments and human nicotinic acetylcholine receptor subtypes. RVG fragments were shown to bind with micromolar affinity to the Lymnaea AChBP. SPR permits determination of on and off rates for binding of all 6 fragments. Our data show slow on rates (ka= 100-300 1/M•s) with off rates (kd = 0.01-0.004 1/ M•s) corresponding to binding with a dissociate rate (Kd of 25.4-60.3 micromolar). Voltage clamp electrophysiology data obtained using Xenopus oocytes shows similar Ki values for inhibition of acetylcholine induced responses on alpha4/beta2 nAChR.

PT.018
RABIES IN IRAN

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Rabies is a disease caused by a virus, Lyssavirus rabies that affects the nervous system and usually results in death unless treated quickly. Rabies is found in mammals in all regions of the world. The disease infects domestic and wild animals, and is spread to people through close contact with infected saliva via bites or scratches. Dogs are the main host and transmitter of rabies but bats,