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with various hepatic diseases and leukosis. Cattle with leukosis showed high serum ADA activity with or without BLVab. There was no correlation between ADA activity and the histopathological findings of degeneration and the degree of fibrosis in the diseased livers. However, serum ADA activity was significantly elevated accompanied by increase of mononuclear cells infiltration.

In conclusion, the elevation of serum ADA activity in bovine liver diseases appeared to be a result of invading mononuclear cells as well as disruption of hepatocellular integrity. The results of this study suggest that the measurement of serum ADA activity might be a useful aid in the diagnosis of bovine hepatic diseases and leukosis.

#### Clinical application of ultrasonographic examination to the canine neonatal brain

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Ultrasonography (USG) is widely used in veterinary medicine as a diagnostic imaging technique. However, little intracranial USG and no pulsed-wave Doppler analysis has been reported in veterinary medicine. Therefore, to obtain ultrasonographic images of brain abnormalities in neonatal dogs, the following experiments were done using 12 neonatal and 2 adult dogs.

Intracranial USG, using the B-mode, color-flow Doppler mode and pulsed-wave Doppler mode was performed on normal neonatal dogs from the day of birth to 50 days of age and on 2 normal adult dogs. Intracranial USG was also performed on 4 dogs with experimentally induced intracranial disease and 2 puppies with mild ventriculomegaly. Pulsed-wave Doppler imaging was performed on the rostral cerebral artery to evaluate blood flow velocity and the resistive index (RI).

It was possible to image the brains of neonates up to approximately 40 days of age. The changes noted in the brain with age were the size of the brain, the formation of sulci and the curve of the rostral cerebral artery. Compared

to the adult brain, the neonatal brain had a less clear distinction between gray matter and white matter with smoother sulci. Blood flow velocity increased as puppies grew older and it was higher than that of adults at 15 or 20 days after birth. The RI was not stable within 24 hours after birth; however, it was stable between 0.65 and 0.70 from 1 day to 30 days old. Then the RI tended to decrease gradually to around 0.50, which was the average level of the normal adults. In the dogs with experimentally induced hemorrhage in the brain parenchyma and ventricle, the hemorrhagic lesions were imaged as hyperechoic areas. One day after intraventricular hemorrhage, mild enlargement of the ventricle and a hyperechoic mass suspected to be a clot were visualized. In puppies with spontaneous mild ventriculomegaly, enlargement of the ventricle and the 3rd ventricle were observed. USG of the brains from puppies with canine herpetic encephalitis, which were fixed in buffered formalin placed in a water bath, revealed enlargement of the lateral ventricle and hyperechoic lesions that resulted from intraventricular hemorrhage.

In this study, the serial changes of intracranial ultrasonographic appearance and the Doppler spectrum of intracranial blood flow were investigated in neonatal dogs. It was suggested that intracranial USG in neonatal dogs provides useful

information on various brain abnormalities. Although further study is needed, pulsed-wave Doppler spectral analysis appeared to be worthy of clinical application.

Study on development of serodiagnosis for Borna disease virus infection using recombinant antigen

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Borna disease virus (BDV) is a pathogen which causes Borna disease in sheep and horses in Central Europe. Recently, a seroepidemiological survey of BDV infection suggested that this virus is distributed in wide areas among different species of animals. However, there is no standard method for serodiagnosis of BDV infection. Thus, in this study, two BDV proteins, p40 and p24, synthesized by means of a baculovirus expression system were used to develop methods of serodiagnosis for BDV infection in *Rattus norvegicus* in the field and in horses.

1. The antigenicity of the two recombinant BDV proteins, rBACp40 and rBACp24, was verified by using rabbit antisera raised against each of the two BDV proteins synthesized in *E. coli* as GST fusion proteins.

2. IFA antibodies to rBACp40 and rBACp24 were detected in sera from rats infected with BDV. In WB analysis, the infected rat sera showed a specific reaction to rBACp40 but not to rBACp24. These results suggest that rBACp40 is better than rBACp24 for serodiagnosis assay.

3. Sera from 30 field rats were examined by ELISA and WB with rBACp40. A specific antibody to BDV was not detected in these assays.

4. Horse sera from Hokkaido were examined by IFA, PG-IFA, and WB. Of 80 sera, 21 (26.2%) and 17 (21.3%) were positive in IFA and PG-IFA, respectively, when MDCK/BDV was used as an antigen. However, none of 21 sera randomly selected from the 80 sera were positive in WB analysis with rBACp40.