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Investigations and considerations on the epizootiology of *Echinococcus multilocularis* in Hokkaido, Japan.

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An epizootiological study was designed to clarify the current status of *Echinococcus multilocularis* in Hokkaido with an objective of providing a baseline data for establishment of control measure against the parasites.

1. Evaluation of a coproantigen detection assay : The validity of a coproantigen detection assay using a murine monoclonal antibody, EmA9, for diagnosing adult *E. multilocularis* infection in the definitive hosts was evaluated by comparison with the result of autopsy. A total of 76 red foxes (*Vulpes vulpes*) shot in Sapporo and environs were autopsied, and the intestinal tracts were examined for *E. multilocularis*. Of which 42 were infected with the cestodes. The ELISA could detect 95.2% (40/42) of the worm positives and showed no false-positives. Two false-negatives were the cases harboring extremely few worms (1 and 4 worms) whereas 3 cases with similar intensities (2, 4 and 6 worms) were diagnosed as positives. In addition, there was strong and significant positive correlation between ELISA OD values and worm intensities ($r=0.779, n=42, P<0.001$).
2. *E. multilocularis* infection among domestic carnivores : 1) *E. multilocularis* infection in stray dogs captured within Sapporo during October 1996 to March 1997 was investigated by the coproantigen detection assay and fecal egg examination. All of the 112 samples showed negative results. The results suggested limited contact between dogs and potential intermediate hosts of *Echinococcus* and *Taenia* species in the urban area. 2) Farm dogs and cats bred in an area endemic for echinococcosis, Koshimizu, a rural town of eastern Hokkaido, were investigated. Taking the 1996 and 1997 survey together, a single (0.4%, 1/227) and 4 cats feces (at least from 2 individuals, 14 samples examined) were found to be positives for *E. multilocularis* coproantigen, but no taeniid eggs were recovered. A questionnaire survey was also conducted on the animal owners to collect information about animal managemental practices. The results revealed 5.2% of dogs and 61.8% of cats showed some access to rodents. Especially in dogs, the way of keeping was associated significantly to contact with rodents (odds ratio=1.786, 95% confidence interval=1.134-2.807).
3. *E. multilocularis* infection among wild foxes : 1) The epizootic state of *E. multilocularis* in urban and suburban of Sapporo was evaluated. During the 1997 to 1998, 17 fox den sites were found in urban area of Sapporo, and 94 fecal samples were collected around the dens. Of the samples examined, 33 (35.1%) were *E. multilocularis* coproantigen positives and 24 (25.5%) were taeniid egg positives.

A relatively high prevalence was consistently detected in the study period, indicating the establishment of urban cycle in the city. 2) A quarterly monitoring for the *E. multilocularis* prevalence among a local fox population was carried out in Koshimizu from April 1997 to January 1998. Thirty-six breeding dens of fox families were identified in the area and 534 fecal samples were collected within 500 m around the dens. Seasonally, whereas the prevalence of coproantigen positives showed no fluctuation (51.6-66.7%), the mean coproantigen OD values of positives rose in summer and winter. Similar variation was found in the egg positive rate in which higher prevalence was observed in summer and winter (31.3% and 38.7%, respectively) than spring and autumn (13.3% and 13.5%, respectively). The data implies the infection pressure on foxes increased in summer and winter.

Genetic and Developmental Study of Tattered^{hokkaido} (Td^{ho}) mouse

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Td^{ho} mouse is a newly found, X-linked dominant mutation which exhibits hyperkeratotic skin, reduced viability in affected females, dwarfism, growth retardation during weaning age, and prenatal lethality in affected males. To map this Td^{ho} locus, microsatellite markers of the proximal region on X chromosome were used. Linkage analysis suggested a possible gene order of cen- (Td^{ho}, *DXMit26, DXMit55, DXMit123*) *-DXMit161-DXMit54 -DXMit103 -DXMit52 -DXMit190 -DXMit138* on the X chromosome.

In the developmental study, it was found that the Td^{ho} male mutants died between E12.5 and E14.5. For examination of genes related to the cause of lethality in Td^{ho} male embryos, suppression subtractive hybridization and Northern blot analysis of globin mRNAs were performed at E12.5 embryos of Td^{ho} mutant and normal mice. Diminished expression levels of embryonic globin genes (ζ and ε globins) in the peripheral blood of E12.5 Td^{ho} male embryos were demonstrated. The increased apoptosis of yolk sac-derived erythrocytes was found at E12.5 Td^{ho} male embryos. Therefore, a defect of embryonic hematopoiesis was suggested to occur in Td^{ho} male embryos.

Ebp, which was identified as a responsible gene for *Td* mouse, was speculated as a candidate gene of Td^{ho}. Two point mutations were detected in the nucleotide sequence of *Ebp* of Td^{ho} mouse. These substitutions caused two amino acids exchanges in the third transmembrane domain of EBP. This mutation is considered to correlate with sterol content of plasma, cholest-8(9)en-3β-ol was accumulated in Td^{ho} female mutants. It was discussed how the toxicity of intermediate metabolites of cholesterol influenced em-