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PC 12 cells, exposure to 50 mM KCl or 100 μ M ATP resulted in simultaneous release of norsalsolinol and dopamine. The time courses of release of dopamine and norsalsolinol evoked by 50 mM KCl or 100 μ M ATP coincided to each other. These releases were dependent on

the concentrations of secretagogues. These findings suggest that norsalsolinol is taken up with dopamine into secretory vesicles via the vesicular catecholamine transporter. These findings may also be related to postural abnormality in Parkinson's disease.

Mechanism of generation of reactive oxygen species with accumulation of copper in the liver of Long-Evans Cinnamon (LEC) rats and its relationship to the onset of acute hepatitis

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Long-Evans Cinnamon (LEC) rats accumulate excess copper (Cu) in the liver in a manner similar to patients with Wilson's disease (WD), and they spontaneously develop acute hepatitis with severe jaundice at 3 ~ 5 months after birth. Reactive oxygen species (ROS) generated as a consequence of the accumulation of excess Cu are thought to be responsible for acute hepatitis in LEC rats. However, the mechanisms by which the excess Cu generates the ROS and induces hepatitis have not yet been revealed in this animal model. In this study, the author investigated the mechanisms of ROS generation accompanying Cu accumulation in the liver of LEC rats and their relationship to the onset of acute hepatitis.

1) The author investigated the mechanism of increased lipid peroxidation (LPO) in the liver using an in vitro incubation system. The levels of LPO in the liver S-9 from hepatic LEC rats were increased by incubating the liver S-9 in the presence of NADPH-generating system (NADPH-gs). This in-

crease was inhibited by EDTA, butylated hydroxytoluene, and catalase (CAT), suggesting that the hydroxyl radicals (\cdot OH) generated by the Fenton-type reaction between H_2O_2 and free Cu, are involved in this increase in LPO. H_2O_2 and \cdot OH are known to be generated one after another during Cu-catalyzed GSH oxidation. Electron spin resonance study revealed a marked increase in \cdot OH generation in the liver cytosol from hepatic LEC rats in the presence of GSH and H_2O_2 . The cyclic regeneration of GSH from GSSG by NADPH-dependent glutathione reductase in the presence of NADPH-gs may cause sustained generation of \cdot OH in the presence of excess free Cu.

2) In vivo \cdot OH production in plasma and liver of hepatic LEC rats was quantified by trapping \cdot OH with salicylic acid (SA) as 2,3-dihydroxybenzoic acid (2,3-DHBA). The ratios of 2,3-DHBA/SA were significantly higher in plasma and liver of hepatic LEC rats and acute Cu-overload Wistar rats than in those of Wistar rats and LEC rats showing

no sign of hepatitis. Treatment with D-mannitol (an $\cdot\text{OH}$ scavenger) to LEC rats suppressed the increases in serum aspartate aminotransferase activity and total bilirubin concentration. D-mannitol also significantly reduced hepatic mitochondrial LPO, a process which is important in the pathogenesis of Cu-induced hepatotoxicity. The author showed the first in vivo evidence of accelerated $\cdot\text{OH}$ generation in the livers of hepatitic LEC rats, and this increase in $\cdot\text{OH}$ generation may play important roles in the development of acute hepatitis.

3) α -Lipoic acid has ROS quenching and metal chelating activities. DL- α -lipoic acid (LA) was administered to LEC rats by gavage at doses of 10, 30 and 100mg/kg five times per week, for 4 weeks. LA prevented severe jaundice in a dose-dependent manner. Anti-

oxidant system analyses in liver showed that LA treatment significantly suppressed the inactivations of CAT and glutathione peroxidase, as well as the induction of heme oxygenase-1. LA at the highest dose slightly suppressed the accumulation of Cu in crude mitochondrial fraction, but it had no effect on the accumulation of Cu in cytosolic fraction. While the increase in LPO in the microsomal fraction was completely suppressed by LA at the highest dose, the suppressive effect against LPO in crude mitochondrial fractions was slight. On the basis of these results, the author concluded that LA has antioxidant effects at the molecular level against the development of Cu-induced hepatitis in LEC rats. Moreover, mitochondrial oxidative damage might be important for the development of acute hepatitis in LEC rats.

Nutritional status of sika deer (*Cervus nippon yesoensis* Heude) in eastern Hokkaido, Japan

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Nutritional status of wild ungulate provides important information on population status, because it affects winter survival, reproductive rate, fetal growth, and body size. Sika deer (*Cervus nippon* Temminck) in Japan were shown to have a wide range of adaptability to their habitat. Few studies of nutritional strategy, however, have investigated sika deer. Hokkaido sika deer (*C. n. yesoensis* Heude) are widely distributed in eastern Hokkaido, which has the most severe winter condition in Japan. The basic information on nutritional ecology will help us to under-

stand the key factor of their life history, adaptation to the dynamism of their environment, and their management.

This study was designed to understand the characteristics of nutritional ecology of Hokkaido sika deer seasonally, and to set up the standard for nutritional assessment of the population. For the fundamental study, qualitative and quantitative analyses of food habits and nutritional value of sika deer diets were conducted. Second, to evaluate the nutritional status of the population, seasonal changes of fat indices were investigated and