



Title	Studies on the characteristics of energy metabolism in sheep and shiba goats
Author(s)	TABUCHI, Masaki
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crease on which recurrent  $\text{Ca}^{2+}$  spikes were superimposed. The second phase of  $[\text{Ca}^{2+}]_i$  rise was suppressed by 85% with application of 1  $\mu\text{M}$  nifedipine, a dihydropyridine-derivative blocker of voltage-dependent L-type  $\text{Ca}^{2+}$  channels.

3.  $\text{K}^+$  (50 mM) depolarization induced a rapid rise in  $[\text{Ca}^{2+}]_i$  consisting of a transient increase followed by a plateau phase. The  $[\text{Ca}^{2+}]_i$  rise on the plateau phase was suppressed by 30% with application of 1  $\mu\text{M}$  nifedipine and by 80% by the removal of extracellular  $\text{Ca}^{2+}$ .

4. The nifedipine-insensitive  $[\text{Ca}^{2+}]_i$  rise was suppressed by 85% by the removal of extracellular  $\text{Ca}^{2+}$ .

5. The nifedipine-insensitive  $[\text{Ca}^{2+}]_i$  rise was reduced by 20% with application of voltage-dependent N-type and P-type  $\text{Ca}^{2+}$  channels blockers cocktail (1  $\mu\text{M}$   $\omega$ -conotoxin GVIA, 300

nM  $\omega$ -agatoxin IVA and 1  $\mu\text{M}$   $\omega$ -conotoxin MVIIC), by 40% with addition of 100  $\mu\text{M}$   $\text{Ni}^{2+}$ , a selective T-type  $\text{Ca}^{2+}$  channel blocker, and by 40% with 2 mM  $\text{Ni}^{2+}$ , a nonselective  $\text{Ca}^{2+}$  entry blocker, respectively.

6.  $\text{K}^+$  depolarization-induced  $[\text{Ca}^{2+}]_i$  rise was dependent on extracellular  $\text{K}^+$  concentrations, and the maximal  $[\text{Ca}^{2+}]_i$  rise was obtained at 40 mM  $\text{K}^+$ . The inhibitory effect of nifedipine was also related to extracellular  $\text{K}^+$  concentrations.

7. These results indicate that the  $\text{Ca}^{2+}$  entry through voltage-dependent L-type  $\text{Ca}^{2+}$  channels and dihydropyridine-insensitive mechanisms occurs within membrane potential ranges evoked by high concentration of glucose, and the dihydropyridine-insensitive mechanisms involves N-type and P-type  $\text{Ca}^{2+}$  channels. Additionally, it is suggested that unknown  $\text{Ca}^{2+}$  entry pathways exist in rat pancreatic  $\beta$  cells.

#### Studies on the characteristics of energy metabolism in sheep and shiba goats

Masaki Tabuchi

*Laboratory of Physiology, Department of Biomedical Sciences,  
Faculty of Veterinary Medicine, Hokkaido University  
Sapporo 060-0181, Japan*

In ruminants, blood glucose levels are consistently low due to their unique feature in digestion and absorption strategy. In central nervous system (CNS), glucose serves as the major energy source in non-ruminants. If this is a ubiquitous law, a question arises how the ruminants compensate for the shortage of glucose supply to their central nervous system. To address this question, an attempt was made by using sheep and shiba goats with implanted probes in their 3rd or lateral ventricles. Cerebrospinal fluids (CSF) were collected through the probe. Previous studies were performed under anesthesia and CSF collection was made at either cisterna magna or lumber spine. This can

underestimate possible changes of CSF components. Therefore, in the present experiments, the most suitable region for CSF collection was first determined by comparing several parameters (glucose, lactic acid, pyruvic acid, FFA, insulin, and electrolytes) between 3rd or lateral ventricle and cisterna magna or lumber spine. Then the effects of restricted feeding and fasting on daily plasma and CSF parameters were examined.

1. Xylazine-induced elevation of glucose concentration was larger in CSF collected from the lateral ventricle than that from cisterna magna or lumber spine. Thus, the 3rd or lateral ventricle was considered to be the most appropriate region

for CSF collection.

2. Restricted feeding (given 9:00 AM – 11:00 AM) caused postprandial changes in several parameters in plasma and CSF, indicating that energy source production via metabolism with the aid of microorganisms in the rumen seemed to be faster than expected in ruminants. It was confirmed that plasma and CSF glucose levels were markedly low in sheep and goats when compared with non-ruminants and the CSF/plasma ratio of glucose was maintained at a constant value ( $0.74 \pm 0.01$ ).

3. Lactic acid concentration in CSF of sheep and goats was about 5 folds of that in plasma. In non-ruminants, it has been reported that the concentration in plasma and CSF was nearly equal. This fact implies that lactic acid may play some role in energy metabolism in CNS of ruminants. No considerable changes were observed in electrolytes concentrations in plasma

and CSF.

4. By fasting, diurnal alterations of glucose and lactic acid observed in animals with restricted feeding were disappeared but plasma FFA concentration was increased and maintained at an elevated level throughout the fasting period. These results suggest that metabolic system has been changed by fasting and that digestion via fermentation can be terminated relatively quickly. Mobilization of FFA did not seem to be involved in brain metabolism since the FFA concentration decreased in CSF by fasting and its actual concentration in CSF is as low as 1/20 – 1/400 of that in plasma.

5. In ruminants, a possibility was suggested that, other than glucose, alternative energy sources such as lactic acid may be ordinarily utilized especially in CNS in order to compensate for the shortage of glucose supply.

Thermogenic uncoupling proteins : tissue distribution and mRNA expression  
by treatment with  $\beta$ 3-adrenergic agonist in obese mice.

Yoshiko NAKAMURA

*Department of Biomedical Sciences  
Laboratory of Biochemistry  
Graduate School of Veterinary Medicine  
Hokkaido University, Sapporo 060-0818 Japan*

Obesity is a serious problem in small animal practice. Current treatment of obesity relies on reducing caloric intake, or increasing energy consumption by exercise.  $\beta$ 3-Adrenergic agonist has been expected to become a new anti-obesity agent.

$\beta$ 3-Adrenergic receptor (AR) is localized specifically in the white and brown adipose tissue (WAT and BAT, respectively). Thus, administration of selective agonist for  $\beta$ 3-AR is expected to induce lipolysis in WAT and thermogenesis in BAT, without side effects. Chronic

treatment of genetically obese yellow KK mice with a selective  $\beta$ 3-AR agonist, CL316,243, caused a significant reduction of body weight, associated with a marked decrease of white fat pad weight.

The mitochondrial uncoupling protein 1 (UCP1) is usually expressed only in BAT and acts as a key molecule for metabolic thermogenesis under the control of sympathetic nerve system. Recently, two UCP homologues have been cloned and called UCP2 and UCP3. Both proteins showed ability as uncoupler in yeast. In