Department of Molecular Physiology
Division of Physical Fitness

Shigeru Takemori, *Professor and Director* Hideki Yamauchi, *Assistant Professor*

General Summary

Research activities in our division have been focused on the plasticity of skeletal muscle and preventive medicine against metabolic syndrome in terms of exercise physiology.

Research Activities

Sarcopenia and mitochondrial function in WBN/Kob-Fatty rats
We have reported that voluntary exercise (Ex) combined with diet restriction (DR) dramatically suppressed the development of chronic pancreatitis, obesity and diabetes in WBN/Kob-Fatty rat. Ex is hypothesized to intensify DR effect synergistically through preserved mass and mitochondrial functions of skeletal muscle. From our experimental results, we tentatively concluded that Ex differentially affected fast and slow muscle as for mitochondria function, and that the preservation of skeletal muscle mass and mitochondrial functions may not be crucially involved in the preventive effect of Ex on chronic pancreatitis and diabetes in WBN/Kob-fatty rats. Increased sugar availability at the initial metabolic stage might have some significance for the synergistic effect of Ex on DR.

The effect of eccentric contraction on sarcomere structure and muscle anabolic signals
Using x-ray diffraction technique, we explored the upper stream sensors of eccentric contraction (ECC) leading to anabolic responses. Plantaris muscle of rat was stretched to the length for maximal isometric tension (L_O) from 0.9 L_O in 0.3 s. H10, M10 and L10 groups received 10 ECC elicited by 100, 75 and 50 Hz nerve stimulation, respectively. L30 group received 30 ECC by 50 Hz stimulation. ISO group received submaximal isometric contraction by 100 Hz stimulation, and CON group received no conditioning contraction. One hour after the series of conditioning tetanus, muscle contractility and x-ray diffraction indicated evident deterioration of sarcomere. Correspondingly, various anabolizing signals were activated and catabolizing signals were inactivated. X-ray diffraction patterns indicated that M10, L10 and L30 preserved the regular arrangement of sarcomere corresponding to sustained contractility. The intensity decrease in the troponin reflection on the patterns suggested regulatory system in sarcomere to be most sensitively affected by ECC, triggering some of the anabolic signaling pathways.

T2-relaxation change precedes denervation-induced muscle atrophy
To counteract the progress of neuromuscular atrophy, detection of early indication of possible muscle atrophy would be helpful. Since water in skeletal muscle has been shown to sensitively reflect muscle state through interaction with myoproteins, we explored early

signs preceding denervation-induced muscle atrophy with MRI. MR images were taken with BioSpec 94/20 USR (9.4T, Bruker) repetitively for 7 days. Compared with the sham-operated side, denervation atrophy was evident in cross-sectional area of the lower legs and muscles 7 days after the operation, while no sign was detected until 5 days. In T2-mapping of the whole lower leg, no specific effect of denervation was observed until 24 h. After 48 h, signal intensity became larger and T2 relaxation time shorter in the denervation side than in the sham-operated side. T2 relaxation time of soleus and plantaris muscle showed similar trends. Histological study with HE-staining of similarly treated muscles showed evident inflammation 12 h after the operation in both of the denervation and sham-operated sides. These inflammatory changes may have caused initial variation in MR images observed non-specifically to the denervation side. MRI could detect some early signs of denervation before the beginning of apparent atrophy.

Roles of polyamines in skeletal muscle hypertrophy

The polyamines are considered to be essential growth factors in virtually all cells. The proposed roles of polyamines are the functioning of ion channels, nucleic acid packaging, signal transduction, cell proliferation, and differentiation, as well as regulation of gene expression. In skeletal muscle, regulation of polyamine levels is reported to associate with muscle hypertrophy and atrophy, yet the underlying mechanisms of polyamine actions are not well defined. We studied how polyamines may affect the proliferation and/or differentiation of murine myoblast progenitor C2C12 cell line. Upon polyamine treatment of C2C12 cells during induction of myogenic differentiation, the number of myotubes significantly increased. Morphologically, polyamine-treated C2C12 cells exhibited elongated cell body and became multi-nucleated. On the other hand, the polyamine did not have influence on myoblasts proliferation. Compensatory muscle hypertrophy in the hind limb of C57BL6 mice with sciatic nerve transaction was enhanced by administration of polyamines. Our study demonstrates that polyamines may play an important role in regulating myogenic differentiation rather than myoblasts proliferation.

Does polyamine administration affect cardiac structure and function of athletes' heart?

Polyamines such as putrescine are poly-cation molecules indispensable for proliferation of eukaryotic cells. Polyamines are also known as modulators of ion channels regulating physiological excitability of cardiac cells. Therefore, polyamines may play a significant role in the hypertrophy and arrhythmia of athletes' hearts. To examine the effects of oral administration of polyamine, rats were bred for 9 weeks at four combined conditions of presence and absence of 1 mg/ml of putrescine in drinking water and freely accessible wheel for spontaneous running. At the end of the breeding period, putrescine concentration in the cardiac cells increased in the putrescine (+) / exercise (−) group, but not in the putrescine (+) / exercise (+) group. Electrocardiograph and structural parameters including heart weight, thickness of ventricle walls, and degree of fibrosis, showed no appreciable effect of putrescine administration with and without exercise. Polyamine was suggested to be strictly controlled to regulate exercise induced hypertrophy of the heart.

Publications

Kurosaka Y[1], Shiroya Y[1], Yamauchi H, Kitamura H[2], Minato K[1] ([1]Wayo Women's Univ, [2]Univ Marketing Distribution Sciences, Kobe, Hyogo). Characterization of fat metabolism in the fatty liver caused by a high-fat, low-carbohydrate diet: A study under equal energy conditions. *Biochem Biophys Res Com.* 2017; **487:** 41-6.

Reviews and Books

Kurosaka Y[1], Yamauchi H, Takemori S, Minato K[1] ([1]Wayo Women's Univ). Protective effects of dietary restriction and physical exercise on intrahepatic fat accumulation. *J Phys Fitness Sports Med.* 2018; **7:** 9-14.