J. Grad. Sch. Biosp. Sci.,
Hiroshima Univ. (2003), **42** : 19～20

Effect of endurance training and acute exercise on sarcoplasmic reticulum function

Shuichiro INASHIMA

Graduate School of Biosphere Science, Hiroshima University,
Higashihiroshima 739-8528, Japan

Repeated intense skeletal muscle contraction leads to a progressive loss of force generating capacity. This decline in function, referred to as fatigue, has a complex etiology which can involve various factors, e.g. reduced availability of energy, build-up of metabolites and dysfunction of excitation-contraction coupling. Although suggested already in 1976 by Eberstenin and Sandow, alterations in the function of the sarcolasmic reticulum (SR), which regulates the intracellular Ca^{2+} concentration, have only recently been considered, as a major contributor to fatigue. It is well established, that increased contractile activity, as occurs in endurance training, elicits pronounced increases in mitochondrial enzymes related to aerobic substrate oxidation and that enhanced aerobic capacity of muscle is a major contributor to improvement in muscular performance. Taking the role of SR in muscle contraction into account, the influence of endurance training on SR function is of interest. However, such studies are sparse and yield conflicting results. Little is also known about whether endurance training protects against deteriorating SR function with prolonged exercise. The only study of Bonner et al. showed that endurance-trained rats displayed a higher SR Ca^{2+}-ATPase activity at exhaustion than untrained rats. Surprisingly enough, inconsistent with previous observations in numerous studies, they found higher SR Ca^{2+}-ATPase activity after exercise than at rest. In the light of these findings, the present study was undertaken in order to assess unequivocally the effects of endurance training and/or a single bout of prolonged exercise on Ca^{2+} regulation by the SR in skeletal muscles.

Forty male Wistar rats were used. The rats were randomly divided to an untrained and a trained group (n = 20 animals per group), and introduced to treadmill running for 10 wk. Following 10 wk of training, animals of both groups were arbitrarily subdivided to a rest and an exercised group (n = 10 animals per group). Approximately 48 h after the last training run, the animals of exercised group were subjected to moderate-intensity running (speed, 27.5 m min^{-1} at 10% grade) to exhaustion on the treadmill. The plantaris muscles, composed mainly of fast-twitch fibers, and the soleus muscles, composed mainly of slow-twitch fibers, were removed from both legs of the rat within 3 min after the termination of running. SR Ca^{2+}-uptake, Ca^{2+}-release, and Ca^{2+}-stimulated ATPase activity were examined in homogenates of these muscles. SR Ca^{2+}-uptake and Ca^{2+}-release rates were measured using the Ca^{2+} fluorescent dye indo-1. SR Ca^{2+}-ATPase activity in the presence or absence of dithiothreitol (DTT), the disulfide reducing agent was measured spectrometrically.

The data in this study confirm that Ca^{2+}-sequesteration by the SR from fast-twitch muscle is depressed after training. Immediately after exhaustive running, decreases in SR function occurred in both muscles, but were more pronounced in the soleus. In the plantaris, reductions in SR Ca^{2+}-uptake rate and Ca^{2+}-ATPase activity were observed in untrained rats only, while in the soleus they were adversely affected irrespective of training status. We hypothesized that oxidation of SH

groups by reactive oxygen species (ROS) may be responsible for the exercise-induced decrement in SR Ca^{2+}-ATPase activity and then measured the activity after incubation with DTT. However, treatment with DTT failed to change the catalytic activity in all samples. Due to non-physiological conditions and concentration *in vitro*, the effects of exogenously generated ROS on the SR may not necessarily be the same as observed *in vivo*. Alternatively, oxidation of SH groups might be reversible *in vivo*. A number of reports have stated disturbances in Ca^{2+}-release after high-intensity, short-term exercise. On the contrary, it is equivocal whether a single bout of prolonged exercise has similar effects to those found in intense exercise. Our results showing that acute exercise evoked a reduction in SR Ca^{2+}-release rate in the soleus are in accordance with previous study on rat fast-twitch muscle and expand these findings to slow-twitch muscle. Currently several mechanisms are likely candidates to explain the reduction in SR Ca^{2+}-release following contractile activity, including alterations in metabolic homeostasis, e.g. Mg^{2+}, ATP, glycogen and lactate within the microcompartmentalized triadic space. Some of these factors could potentially influence *in vivo* Ca^{2+}-release during exercise. It is important to note that in the present study, SR Ca^{2+}-release was assessed *in vitro* under ideal conditions that resemble the environment in the resting muscle. Furthermore, 4-chloro-m-cresol used to stimulate Ca^{2+}-release has been shown to activate directly the Ca^{2+} release channel. This suggest that the decrement in SR Ca^{2+}-release rate shown here is attributable to structural alterations in the Ca^{2+} release channel, but not to the direct action of metabolites. Although the average run time to exhaustion markedly varied between untrained and trained animals (untrained-253.0 min; trained-559.4 min), no differences existed with regard to the magnitude of decreases in SR function in the soleus after exercise. The mean rate of a decline in SR Ca^{2+} handling capacity during acute exercise, as estimated from the run time and the extent of the decline, was more than twofold higher in untrained than in trained soleus, implying that training may be capable of delaying a progression of the deterioration in SR function. It is unlikely that this results from adaptive response of the SR per se to training, as SR function was unchanged by training. In view of the findings that the cellular environment primarily influences structure and function of the SR, one may assume that differences in the rate of a decline would be explained by a slower change in factor(s) that adversely affect(s) SR function. For example, it has been previously shown in rat soleus that endurance training elicits the increases in antioxidant enzyme activity, which result in the protection of the SR against oxidative stress-induced damage. In summary, this study advances our understanding of endurance training and/or acute exercise effects on SR function in skeletal muscles. Endurance training evoked a decrease in SR Ca^{2+}-sequestering ability in resting fast-twitch muscle, but not in slow-twitch muscle, whereas Ca^{2+}-release was unchanged in both muscles. A single bout of prolonged exercise decreased to the same extent SR Ca^{2+}-handling capacity in trained and untrained slow-twitch muscles. These findings indicate that training did not prevent the depressive effects of fatiguing exercise on SR function. From the present study, it is unclear whether there exists a casual relationship between muscular fatigue and SR function because the run time to exhaustion was not significantly correlated with any of parameters indicative of SR Ca^{2+} handling capacity, but suggested that endurance training may be capable of delaying a progression of the deterioration in SR function that occurs during exercise.