ENZYMATIC PROFILE OF FIBERS OF THE HUMAN TRICEPS SURAE MUSCLE DURING LOCAL ENDURANCE TRAINING (QUANTITATIVE HISTOCHEMICAL STUDY)

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Most studies of changes in the enzymatic profile of human muscle fibers during endurance training have been conducted on m. vastus lateralis. Significantly less research has been done on the effect of training on m. triceps surae, which is the muscle that carries the greatest load during both normal and increased physical activity.

The aim of this investigation was to study the character of adaptive changes in m. triceps surae during local endurance training.

METHODS

Nine physically active student volunteers aged 20-22 years, not previously trained in endurance, carried out plantar flexion of the ankle three to four times a week for 8 weeks with an effort of 30% of the maximal voluntary effort, with a duration of 40 min and a frequency of 60 movements/min. The exercise was carried out with one lower limb (the working limb) only. The other lower limb served as the control. Before and after 8 weeks of training, samples of muscle tissue were taken from m. triceps surae of both limbs by needle biopsy. The subjects were warned beforehand about the character of the procedure to be carried out and agreed to take part in it. The samples were frozen in liquid nitrogen and kept in it until the second sample was taken at the end of the experiment. Prolonged preservation of biopsy material in liquid nitrogen does not lead to any significant changes in the parameters studied [2]. Serial transverse sections 10 μ thick were then cut in a cryostat at —20°C. The sections were stained for myofibrillar ATPase with preincubation at pH 4.35 [7], for succinate dehydrogenase (SDH), NADH-tetrazolium reductase (NADH-TR), isocitrate dehydrogenase (IDH), malate dehydrogenase (MDH), glutamate dehydrogenase (GDH), mitochondrial alpha-glycerophosphate dehydrogenase (GPDH), lactate dehydrogenase (LDG), and myoglobin peroxidase (MP) [2], for the complex of enzymes for beta-oxidation of fatty acids (BO) [3], and for phosphofructokinase (PFK) [5] in our modification. The sections to be compared were stained simultaneously. Enzyme activity was assessed cytophotometrically with the aid of the MPV-2 microscope-photometer (Leitz, Germany), by the point two-wavelength method at 570 nm, and expressed in optical density units. Cytophotometry was carried out for all enzymes on the same fibers, identified by staining for myofibrillar ATPase of fibers of both I (slow) and II (fast) types. When the proportion of type I fibers was counted, no fewer than 200 fibers in the sample were taken into account. The results were subjected to statistical analysis by Student's t test.

RESULTS

Predominance of type I fibers was found in samples of m. triceps surae (Table 1). During training, no significant changes in the relative numbers of fibers of types I and II could be found during training in either the working or

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TABLE 1. Enzyme Activity in Fibers of Types I and II Before and After Local Endurance Training (in optical density units, M ± m)

Enzyme	Type of fiber	Working limb		Control limb	
		before training	after training	before training	after training
NADH-TR	I	0,70±0,05	0,67±0,04	0,78±0,05	0,80±0,05
	II	0,41±0,04	0,42±0,02	0,48±0,04	0,41±0,08
SDH	I	0,14±0,01	0,14±0,01	0,14±0,03	0,16±0,01
	II	0,13±0,01	0,12±0,06	0,13±0,02	0,14±0,01
IDH	I	0,10±0,01	0,12±0,01	0,10±0,01	0,13±0,01
	II	0,09±0,01	0,10±0,01	0,10±0,01	0,12±0,02
BO	I	0,14±0,01	0,14±0,02	0,13±0,01	0,15±0,01
	II	0,13±0,01	0,13±0,02	0,12±0,01	0,13±0,01
GDH	I	0,07±0,01	0,05±0,00	0,06±0,01	0,07±0,00
	II	0,08±0,01	0,05±0,01*	0,07±0,01	0,08±0,01
MDH	I	0,28±0,04	0,24±0,02	0,22±0,01	0,22±0,01
	II	0,24±0,02	0,20±0,01*	0,22±0,02	0,22±0,02
GPDH	I	0,06±0,01	0,05±0,01	0,05±0,01	0,05±0,01
	II	0,10±0,01	0,08±0,01	0,08±0,01	0,07±0,01
LDH	I	0,51±0,05	0,33±0,06*	0,47±0,07	0,43±0,06
	II	0,61±0,09	0,33±0,05*	0,51±0,09	0,47±0,05
PFK	I	0,17±0,02	0,13±0,02*	0,16±0,02	0,15±0,02
	II	0,27±0,03	0,20±0,03*	0,28±0,08	0,24±0,04
MP	I	0,06±0,01	0,06±0,01	0,05±0,00	0,06±0,01
	II	0,06±0,00	0,05±0,01	0,05±0,00	0,04±0,00

Note. *) Significant differences between enzyme activity before and after training ($p < 0.05$).

the control muscle. The absence of changes in the ratio of the two types of fibers is in good agreement with data obtained by most investigators [6, 9], who had repeatedly shown that this parameter is stable in man training under physiological conditions. Those authors who observed changes in the relative numbers of the two types of fibers as a result of training [12] as a rule used modified methods of staining for myofibrillar ATPase. These methods are not generally accepted at the present time and require special study of their adequacy.

Activity of glycolytic enzymes (LDH and PFK) was significantly reduced as a result of training about equally in the two types of fibers (LDH in type I fibers by 35.3%, in type II fibers by 45.9%; PFK in type I fibers by 23.5%, in type II fibers by 25.9%) (Table 1). Likewise, during training activity of MDH fell in type II fibers by 16.7%. In type II fibers GHD activity also fell by 37.5%. All the changes observed are significant at the $p < 0.05$ level. No such changes were found in the control muscle.

The decrease in glycolytic potential found in the present investigation as a result of endurance training may be linked with a gradual shift from the use of carbohydrate to the predominant use of fatty energy substrates [9]. The decrease in activity of MDH, one of the components of the mitochondrial malate-aspartate shuttle mechanism, may also be evidence of a decrease in the degree of carbohydrate utilization for energy production. It is noteworthy that a significant decrease in the activity of this enzyme was found in fibers of type II, in which carbohydrate constitutes the dominant type of energy substrate. A decrease in activity of GDH, an enzyme of protein catabolism, may indicate lowering of the level of oxidation of amino acids and a possible shift to the more economic energy substrate — fat. The "fatty shift" and lowering of the glycolytic potential as a result of endurance training also were observed previously, mainly in global investigations involving a large muscle mass [1, 4, 8].

No significant changes in activity of alpha-glycerophosphate dehydrogenase and myoglobin peroxidase in the muscles could be found as a result of training, in full agreement with data obtained by other workers, who noted the stability of these parameters during training [10, 11].

We found no significant changes in activity of enzymes of the oxidative series. Considering the predominance of type I fibers in this part of the muscle, which undoubtedly determines its high oxidative potential on the whole, it can be postulated that during training the functional demands of the muscle can be fully satisfied by its aerobic capacity, and that an additional increase in its oxidative potential was not required. Similar results were obtained by Scottish

workers during training under global working conditions with low physiological constraint [8]. Just as in the work cited, so also in the present investigation the character of response of the muscle to the training exercise is evidently determined by the relationship between its initial structural-metabolic state and the degree of specific activity of its motor units.

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