

THE LIPID CONTENT OF RABBIT CALF MUSCLES FOLLOWING SECTION OF TIBIAL NERVE

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Histological and chemical changes are known to occur in skeletal muscles following various lesions of the nervous system.

G. N. Orlova's [6] experiments on white rats demonstrated that section of the sciatic nerve is followed by clearly seen microscopic changes in muscle tissue within one day after the operation.

Considerable areas of connective tissue acquired the character of adipose tissue. M. L. Borovsky [1, 2] observed in experiments on dogs that after section of the sciatic nerve the denervated calf muscle showed atrophy of muscle fibers and appearance in them of fat globules 1 1/2 months after operation.

Numerous communications are devoted to the question of chemical changes in skeletal muscles following various lesions of the nervous system, but they are mainly concerned with protein and carbohydrate-phosphorus metabolism [3, 7, and others].

Our observations [4, 5] show that 2-3 weeks after section of the tibial nerve in rabbits there is considerable loss of weight in the denervated muscle. Definite changes of labile phosphorus compounds and creatin also appear at this time. Thus there is definite correlation between the weight (degree of atrophy) of the denervated muscle and the extent of chemical changes. Chemical changes in the muscle are the more pronounced the greater the loss in weight. However, the weight criterion of the degree of atrophy is obviously insufficient, since atrophy is a complex process requiring special study.

It is well known that denervated muscles undergo fatty degeneration. But the histological method of determination of fat in skeletal muscle in neuromuscular dystrophies does not give a sufficiently precise indication of the quantitative lipid changes in denervated muscle.

The present communication presents results of studies concerned with the quantitative changes in lipids, as well as the distribution of phosphorus between proteins and lipids, in denervated muscles in rabbits during various stages of the atrophic process evoked by section of the motor nerve.

EXPERIMENTAL

The left tibial nerve in an adult rabbit weighing about 2.5 kg was sectioned and after different intervals postoperatively the amount of protein and lipid was determined in the denervated gastrocnemius muscle; phosphorus was also determined in the protein and in lipids. The gastrocnemius muscle of the other limb served

* Deceased.

as control. The muscles were dissected under general magnesia anesthesia and then immersed in liquid nitrogen. The muscles were then weighed and ground in mortars with liquid nitrogen. 5 g of this pulverized muscle was then treated with 5 ml 6% trichloroacetic acid solution and ground in a cooled mortar. The proteins and lipids remaining after filtration were ground and dried to constant weight at 100°. The dried mass so obtained was ground to a powder and placed in a Soxhlet apparatus. The lipids were extracted successively with ether, acetone and a mixture of methyl alcohol and chloroform (1:1); extraction with each solvent was continued for 12 hours. On completion of the extraction the remaining proteins were dried to constant weight and the amount of lipids was determined by the difference between the weight of the proteins and lipids and the weight of the proteins. The lipid content was determined as percentage in relation to the dry weight of proteins and lipids. The amount of phosphorus was estimated colorimetrically by the Fiske-Subbarow method.

RESULTS

Experiments on normal rabbits showed insignificant differences between the lipid content of the right and left gastrocnemius muscles (Table 1).

TABLE 1

Lipid Content of the Gastrocnemius Muscles in Normal Rabbits

No. of experiment	Amount of proteins and lipids (as % of dry weight of combined proteins and lipids)			
	In right gastrocnemius		In left gastrocnemius	
	Proteins	Lipids	Proteins	Lipids
1	91.83	8.17	91.23	8.77
2	88.33	11.63	88.98	11.02
3	90.15	9.85	89.77	10.23
4	91.07	8.93	90.25	9.75
5	89.52	10.48	89.79	10.21

Comparison of the weight of denervated muscle, the amount of trichloroacetic acid filtrate, and changes in the composition of some phosphorus compounds led to the conclusion that appreciable changes in these occur 2-3 weeks after the operation. It could have been expected that other chemical changes would also be clearly apparent after the same time. Actually, quantitative changes in lipid content became definitely apparent 3 weeks after the operation. (Table 2).

Table 2 shows that, 1-2 weeks after operation, the lipid content of the denervated muscle is higher than that of the intact muscle of the other limb. Beginning with the 3rd postoperative week the amount of lipids in the denervated muscle shows a steep rise.

As has already been mentioned the phosphorus compound content of muscles undergoes considerable changes after denervation. The increased lipid content of denervated muscle noted in these investigations led to the question whether this process affected the distribution of phosphorus between the proteins and the lipids.

Considerable accumulation of phosphorus in the muscle lipids was observed after denervation (Table 3).

This accumulation gradually increased as more time elapsed after the operation. While in the normal no consistent differences in the phosphorus content of the lipids from the right and left limbs were observed, and the variations were within the limits of experimental error (Table 4), on the operated side (in this case the left) 2 weeks after denervation the phosphorus content was, on an average 34% higher than on the intact, after 3 weeks it was 52% higher, and after a month, 93% higher. No appreciable differences in the protein phosphorus content were observed.

TABLE 2

Protein and Lipid Content of Rabbit Gastrocnemius Muscles After Section of Tibial Nerve

Time after operation	Amount of proteins and lipid (% of dry weight)			
	Control muscle		Experimental muscle	
	Proteins	Lipids	Proteins	Lipids
One week	93.1	6.90	90.78	9.28
	92.38	7.62	90.51	9.49
	93.31	6.69	90.69	9.31
Two weeks	93.59	6.41	89.28	10.72
	93.29	6.71	91.79	8.21
	90.66	9.34	88.78	11.22
Three weeks	94.65	5.35	80.52	19.48
	93.39	6.61	84.41	15.59
	93.31	6.69	88.24	11.76
One month	94.43	5.57	82.51	71.49
	92.85	7.15	83.58	16.42
	91.17	8.83	84.31	15.69
One and one-half months	92.82	7.18	80.30	19.70
	94.07	5.93	85.94	14.06
	90.75	9.25	80.92	19.08
Three months	92.02	7.98	76.44	23.56
	91.57	8.43	69.04	30.96

TABLE 3

Phosphorus Content of the Gastrocnemius Muscle Proteins and Lipids in Rabbits After Section of the Tibial Nerve

Time after operation	Amount of phosphorus (in % of dry weight of total proteins and lipids)			
	In control muscle		In experimental muscle	
	Protein P	Lipid P	Protein P	Lipid P
Two weeks	0.49	0.090	0.42	0.119
	0.40	0.078	0.45	0.099
	0.40	0.093	0.48	0.133
	0.46	0.115	0.53	—
Three weeks	0.43	0.105	0.54	0.166
	0.46	0.098	0.47	0.183
	0.36	0.076	0.43	0.097
	0.41	0.105	0.41	0.135
One month	0.37	0.08	0.45	0.177
	0.55	0.096	0.59	0.134
	0.46	0.113	0.48	0.157
	—	0.085	0.61	0.196
	0.42	0.103	0.46	—
One and one-half months	0.39	0.092	0.46	0.135
	0.39	0.079	0.39	0.117

TABLE 4

Phosphorus Content of Gastrocnemius Muscle Proteins and Lipids in Normal Rabbits

No. of experiment	Amount of phosphorus (in % dry weight of total proteins and lipids)			
	In right muscle		In left muscle	
	Protein P	Lipid P	Protein P	Lipid P
1	0.45	0.092	0.45	0.082
2	0.42	0.090	0.49	0.092
3	0.42	0.097	0.43	0.097

The data obtained indicate that as the result of impaired motor innervation the denervated skeletal muscle undergoes profound trophic changes, expressed in particular by an increased lipid content and a corresponding increase of phosphorus in the lipids.

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