

Administration of VA during pregnancy caused an increase in total EM, but the postimplantation EM rose only after administration of altax and santocure.

The vulcanization accelerators captax, santocure, santocure-mor, thiurams D and E and, in particular, altax cause sharp depression of reproductive function in albino rats.

LITERATURE CITED

1. Yu. V. Gul'kevich et al., Arkh. Patol., No. 2, 9 (1971).
2. T. B. Davydova, Gi. San., No. 4, 108 (1973).
3. A. P. Dyban, Ontogenez, No. 6, 582 (1977).
4. M. V. Korablev, Dithiocarbamic Acid Derivatives [in Russian], Minsk (1971).
5. I. V. Santoskii and V. N. Fomenko, Late Sequelae of the Effect of Chemical Compounds on the Organism [in Russian], Moscow (1979).
6. "Principles of testing therapeutic substances for teratogenicity," World Health Organization Technical Report Series No. 364 [in Russian], Moscow (1968).
7. The Effect of the Environment on Human Health [in Russian], Moscow (1974).
8. S. Epstein and H. Shafner, Nature, 219, 382 (1968).

REGENERATION OF SKELETAL MUSCLE AFTER MECHANICAL TRAUMA IN REPTILES

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The regenerative properties of muscle tissue have been studied most extensively in mammals [1, 4-10, 12, 14]. The ability of the skeletal muscles of reptiles to recover after injury has not been specially studied. There have been only incidental observations on the regeneration of muscle tissue as part of a study of regeneration after amputation of the tail in lizards [13, 15]. The present writers have shown that the muscles of reptiles can recover when the whole muscle is autografted [2, 11].

It was decided to investigate the regenerative properties of skeletal muscles of reptiles after mechanical trauma in order to compare differences in the regenerative activity of muscle tissue in representatives of different classes of vertebrates, and details of the study are given below.

EXPERIMENTAL METHOD

Turtles (*Testudo horsfieldi*) weighing 200-400 g, kept under animal house conditions with constant illumination and at a constant temperature of 25°C, were used. The gastrocnemius muscle was completely divided transversely. Between 2 and 6 months after the operation the contractile activity of the muscles was tested by electrical stimulation of the tibial nerve, after which the muscles were fixed and sections cut for microscopic and ultrastructural investigation.

EXPERIMENTAL RESULTS

After trauma the muscle stumps were separated by a defect measuring 3-5 mm. The defect 2 weeks after the operation was filled with loose connective tissue, but in the stumps, in the zones adjacent to the region of the defect, a well-marked process of destruction of injured muscle fibers was observed, not only in the immediate vicinity of the site of trauma, but also some distance from it. Destructive changes were more marked in the distal stump at

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Fig. 1

Fig. 1. Fragment of satellite cell in injured turtle muscle, 19,620 \times .



Fig. 2

Fig. 2. Turtle muscle bud 2 weeks after division of muscle. Hematoxylin, 200 \times .



Fig. 3. Muscle tube in region of defect 2 weeks after division of muscle. Hematoxylin, 200 \times .

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Besides destructive changes, two weeks after injury intensive regenerative processes developed in the damaged muscle. Electron-microscopic investigation revealed satellite cells, often arranged in chains at the periphery of the old muscle fibers, which preserved their structure (Fig. 1). As a result of phagocytosis of the destroyed parts of the muscle fibers, sarcolemmal tubes were formed, in which spindle-shaped cells (evidently myoblasts) could be observed under the light microscope. The region of the defect began to be filled not only with connective tissue and blood vessels, but also by myogenic cells. Myoblasts, myosymplasts, and single muscle tubes forming whole streams penetrated from both the proximal and the distal stumps into the region of the defect. They were arranged haphazardly. Among the free cells mitoses could be seen. In the proximal stump, formations resembling "muscle buds" of mammals could sometimes be seen on the ends of the fibers facing the defect: small pools of sarcoplasm, devoid of fibrillary structures and containing several muscle nuclei (Fig. 2). A picture of vacuolar degeneration was observed in some of the "buds," while others had no signs of degeneration. The appearance of "muscle buds" was observed later in the distal stump than in the proximal stump — 1-1.5 months after division of the muscle. By that time their number in the proximal part of the muscle was sharply reduced, and after two months the "buds" could no longer be seen in either part of the divided muscle. In the zone of the defect, besides myoblasts and myosymplasts, muscle cells in the subsequent stages of development were found: muscle tubes and young muscle fibers (Fig. 3). The process of regeneration continued for a long time, and not until three months after the operation were immature myogenic elements found in the zone of the defect. The defect was filled mainly with differentiated thin cross-striated muscle fibers, separated by wide bands of connective tissue.

Simultaneously with differentiation of the muscle cells, the innervation of the muscle was gradually restored. As early as one month after the operation the muscle contracted on account of the proximal stump in response to electrical stimulation of the tibial nerve. At this stage of the investigation single axons, some of them branching to form irregular plexuses, were growing into the region of the defect, and after two months developing axomuscular synapses could be seen on the newly formed muscle fibers in the region of injury. Restoration of the innervation of the distal stump took place slowly and was not complete until after 6 months.

As this description shows, processes of destruction and regeneration developed later in the distal stump and differed in some respect from the corresponding processes in the proximal stump. The reason is evidently that as a result of the operation the innervation and blood supply of the proximal stump are disturbed only in the zone of injury, but the distal stump is completely denervated and its blood supply is more severely upset. This causes edema of the distal stump in the immediate period after the operation and delay in the development of destructive and regenerative processes as well as atrophy of the muscle fibers and the more abundant development of connective tissue than in the proximal stump.

Previously [1, 3] the writers studied regeneration of the rat and frog gastrocnemius muscle after total transverse division. Comparison of the repair process in frogs, turtles, and rats showed that regeneration of the injured muscle takes place rather faster in reptiles than in amphibians, but this process is similar in character in turtles and frogs. In both animals myoblasts, myosymplasts, and muscle tubes are found simultaneously in the region of injury for a long time. Regeneration in mammals takes place rapidly and there is a clearer line of demarcation between the myoblast and myosymplast stages. The "muscle buds" of reptiles appear later than in mammals, they are fewer in number, and they last longer.

Consequently, reptiles occupy an intermediate position between amphibians and mammals as regards both the speed of the repair process in skeletal muscle and the method of regeneration of the muscle fibers.

LITERATURE CITED

1. R. P. Zhenevskaya, Neurotrophic Regulation and Plastic Activity of Muscle Tissue [in Russian], Moscow (1974).
2. R. P. Zhenevskaya, I. L. Novoselova, and M. M. Umnova, Byull. Éksp. Biol. Med., No. 10, 474 (1977).

3. R. P. Zhenevskaya, M. M. Umnova, I. L. Novoselova, V. M. Gorbunov, and A. A. Ovsepyan, in: Methods of Regeneration and Cell Division. Proceedings of a Symposium [in Russian], Moscow, p. 22.
4. A. A. Klishov, Histogenesis, Regeneration and Neoplastic Growth of Skeletal Muscle Tissue [in Russian], Leningrad (1971).
5. L. M. Kulagin, Histomorphology of Muscles under Extremal Conditions [in Russian], Kuibyshev (1977).
6. M. F. Popova, "Plastic protective and reparative state of regenerating tissue," Author's Abstract of Doctoral Dissertation, Moscow (1976).
7. A. N. Studitskii, Experimental Surgery of Muscles [in Russian], Moscow (1959).
8. A. N. Studitskii, Transplantation of Muscles in Animals [in Russian], Moscow (1977).
9. A. N. Studitskii and Z. P. Ignateva, Regeneration of Muscles in Higher Mammals [in Russian], Moscow (1961).
10. A. N. Studitskii and A. R. Striganova, Regenerative Processes in Skeletal Muscle [in Russian], Moscow (1951).
11. M. M. Umnova, R. P. Zhenevskaya, and I. L. Novoselova, Byull. Éksp. Biol. Med., No. 7, 94 (1977).
12. B. M. Carlson, The Regeneration of Minced Muscles, Basel (1972).
13. E. B. Kahn and S. B. Simpson, Dev. Biol., 37, 219 (1974).
14. M. Reznik, La Régénération du Muscle Strie Squelettique. Étude de Morphologie Expérimentale, Paris, Ed. Archives de Biologie (1971).
15. J. Zika and M. Singer, Anat. Rec., 152, 137 (1965).