

THE REGENERATION OF SKELETAL MUSCLE IN MAMMALS UNDER NOVOCAIN BLOCK

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The method of novocain block, developed by A. V. Vishnevskii, has been widely used in medical practice in the treatment of a variety of diseases. It has been shown that by its use the intensity of the inflammatory reaction is considerably reduced, and that inflammation in the stage of a serous effusion may even undergo regression [1, 2].

A differing action of novocain on the inflammatory reaction, depending on the place of its injection, was observed by K. A. Meshcherskaya, N. Melent'eva and G. Obogrelova [3]. These workers also found a weakening of the inflammatory reaction in the spleen after subcutaneous injection of novocain, and conversely, intensification of the inflammation after intraperitoneal injection.

In view of the statements of certain authors that the failure of regeneration of striped muscle after mechanical trauma is dependent on a well-developed inflammatory reaction and subsequent connective tissue reaction, we carried out histological investigations in order to study the effect of novocain block on the process of repair of skeletal muscle.

EXPERIMENTAL METHOD

Experiments were carried out on sexually mature white mice of both sexes, weighing 20-22 g. The calf muscles were used in the investigation. By means of ophthalmic scissors a linear incision was made in the muscles in their middle third, through half their thickness. Aseptic precautions were observed during the operations. Altogether 134 white mice were subjected to operation.

The experiments consisted of 3 series:

Series I - a study of the course of regeneration in control animals.

Series II - observations on regeneration during paranephric block (this series was performed on males only): immediately after operation an injection of 0.2 cm^3 of a 0.25% solution of novocain was given into the paranephric cellular tissue, alternately on the right and left sides (the injection was subsequently repeated after every second day).

In setting up this series we took into consideration the findings of several workers, especially L. N. Vlasova [4], that paranephric block reduces the intensity of an inflammatory process.

Series III - examination of the special features of regeneration after the injection into the lumbar division of the sympathetic trunk of 0.2 cm^3 of a 0.25% solution of novocain or 0.2 cm^3 of physiological saline. Injections were given immediately after operation, and then either every day (4-5 injections in all) or every second day (4 injections).

The role of the sympathetic system in the processes of regeneration was studied by A. Yu. Sozon-Yaroshevich [5]. This author showed that extirpation or division of the sympathetic trunk supplying the limbs had a favorable effect on the processes of healing; stimulation of various divisions of the sympathetic system, and in particular the ganglia, had the opposite effect.

The muscles were fixed in Zenker-formol. Paraffin wax sections, 6-8 μ in thickness, were stained with azure II-eosin, Heidenhain's iron-hematoxylin, Mallory's mixture, and survey films by Carazzi's hematoxylin.

The description of the course of regeneration in the control animals will be utilized later only for purposes of comparison, in view of the large number of references to it in the work of other investigators.

EXPERIMENTAL RESULTS

The difference between the controls (series I) and series II was clearly visible on only the second day after operation, and concerned the phenomena of immigration of blood cells into the focus of injury, and of phagocytic activity.

Whereas in the control mice on the 2nd day a large number of immigrating cells could be detected, both between and within the destroyed muscle fibers, and many phagocytes were present in necrotic masses of debris, in the series II animals at the same time small numbers of cells were seen between the damaged fibers, but the necrotic masses of debris contained hardly any phagocytes.

In this way a delayed resorption of the necrotic masses of debris took place, and hence on the 4th day and sometimes at later periods a large number of unabsorbed remnants of damaged muscle fibers remained.

Proliferation of connective tissue cells lagged behind the control animals: the collagen fibers that formed were thin; the development of collagen bundles was delayed, the delay being greater the more injections were given. The connective tissue intermuscular septa remained poor in cells throughout the whole period of regeneration.

Our findings were in agreement with the observations of A. S. Ershtein [6], who studied the effect of novocain block on the processes of sensitization and found weakening of the Schwartzman phenomenon, and in particular a reduction in the infiltration by cells (histiocytes).

Another difference between series II and the control animals was the greater regularity of growth of the muscle fibers and also the small diameter of the regenerating muscle formations, giving the impression of dystrophy. This may possibly be the consequence of failure of development of the cells of the interstitial connective tissue which is known to carry out a trophic function.

In the series II animals, compared with the controls, there was some degree of retardation of differentiation of muscle cells. However, by the 25th day, all these differences had become to a large extent obliterated, apart from the fact that the regenerating muscle fibers were arranged in a more parallel fashion.

In relation to the mechanism of action of a paranephric novocain block, there are findings, in particular those of T. S. Miryushchenko [7], that the block brings about phasic changes in the nervous processes in the central nervous system in which inhibition predominates.

Before turning to the description of the series III experiments, we must emphasize that the differences between the experiments in which injections of novocain were given and those in which physiological saline was injected were not found, and the variations from the controls were the same in both cases. This is perfectly in order, since according to A. V. Vishnevskii [1] the essence of the novocain block is weak stimulation of corresponding areas of the nervous system, a result of which can be achieved by the use of other substances, as was pointed out by the originator of the novocain block method himself. The findings of A. M. Vikhert [8], who studied the course of the Arthus phenomenon during local anesthesia, are also in accordance with this view.

In a series of experiments, A. M. Vikhert used physiological saline instead of novocain and also observed some degree of acceleration of the development of the Arthus phenomenon. Further, despite the fact that the injections were given on the left, there were corresponding changes, but expressed to a lesser degree, in the right calf muscle, which must be explained by the presence of connections between the left and right sympathetic trunks, which were demonstrated in particular by G. A. Orlov and N. I. Batygina [9].

Differences between the control and series III animals were apparent on the first day after operation: on the 2nd day, in the mice of series III a larger number of phagocytes than in the controls could be detected among the damaged muscle fibers. It must be pointed out that besides necrotic masses of debris thickly strewn with phagocytes, there were also masses in which no leucocytes at all were present.

The connective tissue activity in individual cases in series III was higher than in the controls on the 2nd day, as shown by the formation of bands of fibroblasts -- the result of the increased mitotic activity of these cells. The fact that the immigration of cells into the damaged muscle fibers continued intensively on the 3rd day also was remarkable, because on the 4th day sarcolemma tubes, densely packed with cells, were seen, with necrotic masses of debris included within them, whereas in the control animals the process of resorption was almost completed. It might appear strange that in spite of the increased phagocytic activity observed on the second day in series III, the process of resorption lagged behind the controls. This was due to the increased fibroblastic activity which exerted an inhibitory effect on phagocytosis and autolysis, and the preservation of necrotic debris for a long time in the region of the wound itself prolongs the process of immigration of blood cells into the focus. Also of importance was the accumulation in the region of the injury of the enzyme macrocytase, possessing a proteolytic activity, as a result of the large number of macrophages, a feature first pointed out by I. I. Mechnikov and later confirmed in special research by V. Z. Gorkin and M. N. Kondrashova [10]. The appearance of new areas of necrosis was due to the action of this enzyme.

The subsequent course of restoration of the muscle depended on the character of the injections. In cases where the injections into the sympathetic trunk were given daily, growth of the corresponding muscle fibers took place more energetically than in the experiments in which injections were given at intervals of 2 days. The reactions on the part of the connective tissue in the latter case were more severe, which led subsequently to the presence of a large number of connective tissue cells in the intermuscular spaces.

M. P. Galashnikova [11], who studied the vascular and temperature reactions in response to stimulation of the sympathetic nerve, pointed out that besides acting on the vessel wall, the sympathetic nerve also has a direct action on the biochemical processes in the tissues. This effect is usually inhibitory in character, but in response to repeated stimulation this author observed stimulation of tissue metabolism. This stimulation of tissue metabolism also led to the more energetic growth of the muscle formations, when injections were given daily into the sympathetic trunk.

An essential difference from the control series was the retention of necrotic masses of debris in the regenerating muscle throughout the entire period of regeneration, mainly when the injections were given at intervals of 2 days. Part of the necrotic masses of debris were remains of unresorbed areas of damaged muscle fibers, other parts were newly formed areas of necrosis.

In some cases the necrotic masses of debris filled the greater part of the regenerating tissue, and they were found up to the 25th day. In such cases, there was a particularly intensive reaction on the part of the connective tissue, as shown by an increase in the saturation of the connective tissue interspaces and the marginal areas of the necrotic masses with cells.

Experiments carried out on females revealed differences in the structure of the regenerating muscle between the males and females, both experimental and control. In the control females on the 6th, 8th and 15th days after operation, many necrotic masses of debris were detected. The corresponding event in the males took place after the fourth injection (at intervals of 2 days) of novocain and of physiological saline into the sympathetic trunk.

After injection of novocain into the sympathetic trunk of females, as a rule necrotic masses of debris were not present in the regenerating muscle, and less reactivity was observed on the part of the connective tissue.

DISCUSSION

Hence the paranephric novocain block brought about a fall in the intensity of the inflammatory reaction, and later, a diminution of the connective tissue reaction. In consequence of this, the growing muscle fibers took on a more parallel course although the fibers themselves were somewhat dystrophic in character.

During novocain block of the sympathetic trunk, on the other hand, a considerable increase was observed in the intensity of the inflammatory reaction. The reparative processes depended on the character of the

injections: in response to daily injections of novocain, signs of regeneration of the muscle formations themselves were predominant, but when the injections were given at intervals of 2 days, there was considerable development of connective tissue, rich in cells.

SUMMARY

The regenerative process of the skeletal muscles (m. gastrocnemius) was studied in experiments on white mice in conditions of novocain block. It was shown by the histological examination that the course of repair depends on the site of novocain injection. Paranephral novocain block caused a decreased intensity of inflammation and reduction of the reactivity of connective tissue. A parallelism of muscular fibers and their slight dystrophy was noted in these experiments.

A significant increase in the intensity of the inflammatory reaction was noted in novocain block of the sympathetic trunk (administration of 0.2 cc of 0.25% novocain solution). The regenerative process varied depending on the character of the novocain injection.

Thus, in daily administration of novocain, the prevalence of regeneration of the muscle formations proper was observed, while in injections with 2-day intervals a pre-elective development of connective tissue rich in cells was noted. The structure of the regenerate was different in the males and in the females.

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