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RESEARCH ARTICLE

Morphological Picture of Bone Tissue from Shooting Fractures in Peresous Compression - Distraction Osteosynthesis According to G.A. Ilizarov in Low Mountains

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ABSTRACT

In an experiment in low-mountain conditions, the features of changes in the structure of bone tissue in the zone of a gunshot fracture of the shank diaphysis were studied. It was revealed that when using the compression-distraction method according to G.A. Ilizarov, a pronounced angiogenic effect is observed, an improvement in reparative processes. The results obtained confirm the need to use the method of extrafocal compression-distraction osteosynthesis to accelerate the healing of gunshot fractures of tubular bones.

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Introduction

Despite the tremendous advances in modern medicine, the treatment of gunshot fractures of the long bones of the skeleton continues to be a complex and unresolved problem [1.3]. The course of the wound process of a gunshot wound is determined by the nature and volume of damaged tissues, vascular reaction, the development of hypoxia in tissues, especially muscle, which occurs against the background of microcirculation disorders.

The impact of injuring projectiles on the human body has a colossal effect, the study of which has been devoted to a huge number of works [1,4,5]. Studying the literature, we managed to find very few scientific works devoted to the study of gunshot wounds in conditions of high and low mountains. Since the previous series of our work presented extensive experimental material on the features of bone tissue regeneration during stable osteosynthesis in high altitude conditions [1]. This series of experiments was performed in order to obtain basic digital data on the state

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of the bone and soft tissues of the limb for a comparative study of their response in high altitude conditions.

Experimental Methods

Experimental studies were carried out on adult outbred 27 dogs, which, on the 1st day of stay in the vivarium in low-mountain conditions (Bishkek, 780 meters above sea level), reproduced a model of a gunshot tibia fracture with the imposition of the Ilizarov apparatus. Gunshot fractures were inflicted in dogs in the diaphysis of the shin bones with the application of the Ilizarov apparatus under neuroleptanalgesia. The point of infliction of a fire wound was in the area of the middle - lower third of the leg from the lateral side from a distance of 20 meters from the PM, with a conical bullet (9 mm caliber), blunt-pointed shape with a steel core, 102 mm long, weighing 6.4 g. [1].

Research Methods

Morphological examination was performed every 7 days for 2 months using conventional histological methods. In the bone tissue, the number of vessels, osteons, bone cells (on an area of 1 mm^2), the area of osteon canals (in μm^2). All stages of preparation processing were carried out in accordance with the principles of quantitative methods for the study of histological objects [2,4,5]. At all stages of the experiment, the preparations were processed in accordance with the generally accepted principles of quantitative methods for the study of histological objects. The data obtained were processed in the statistical program for processing biomedical data SPSS 16.0, as well as in the Microsoft Office Excel 2008 application. When comparing the data, the Student's t test was used for unrelated samples.

Research Results and their Discussion

By the end of the first week of the experiment, in the zone of the gunshot fracture in the periosteum, hypertrophy and loss of a clear structure of the periosteum are observed, which is more pronounced somewhat deviating from the fracture line, in this regard, the periosteal area of proliferating cellular elements has a characteristic appearance of a drop-shaped "influx", which is directed thickened part to the line of damage. In the same place, among the cellular elements that form heavy-like structures of irregular shape, narrow fields of oxyphilic-staining osteoid tissue are visible. In the central part of the defect there are bone trabeculae separated by strands of delicate fibrous connective tissue. Here you can find massive fields of proliferating connective tissue elements without clear boundaries. Elements of fibroblastic type cells - fibroblasts prevail in the interfragmental zone of the regenerate (35,42±2,46%) and fibrocytes (9,45±1,24%). A high content of relative proportions of osteoblasts (24,53±1,12%), and the presence of osteocytes (3,15±0,41%), osteoclasts (0,73±0,01%) indicate the intensity of bone formation processes, and a high content vascular endothelial cells (21,56±2,52%), evidence of intense vasculogenesis.A zone of necrotic structureless tissue of varying thickness is adjacent to the surface of the compact bone substance along the line of damage. On the side of the bone margins of the defect, short trabeculae of reticulofibrous bone tissue are formed, originating from lamellar bone tissue with preserved osteogenic cells, overgrowing areas with neglected lacunae.

By the 14th day, an intense periosteal osteoplastic reaction is observed in the fracture zone. Periosteal strata, located somewhat away from bone defects, occupy significant territories, merging with the endosteal callus. In all cases, there is an intensive formation of reticulofibrous bone tissue, the cellular sources of which are cambial bone elements. Active osteoblasts produce bone matrix components from both the periosteum and the endosteum. Intermediate osteogenesis is especially pronounced, which leads to the filling of the defect in this area with a mesh of young bone structures. At this time of the experiment, among the cells of the interfragmental part of the regenerate, a high content of elements of the fibroblastic series remains - fibroblasts make up 20,10±2,12%, and fibrocytes 17,10±1,21% of the total number of cells. Along with this, there is a significant, 1,9 and 2,5 times, respectively, increase in the proportion of osteocytes and osteoclasts with a practically unchanged percentage of osteoblasts, a slight decrease in the proportion of endotheliocytes (77,5% of the value of the previous period).Despite an increase in the number of vessels by 24,6% compared with the previous observation period, the lumens of the vessels demonstrate a constrictor reaction (narrowing by 12,3%), which leads to a decrease in the cross-sectional area of the bloodstream by 4,5%. Calculation of the relative areas of the tissues of the regenerate showed that with an almost unchanged relative area of blood vessels (91,5% of the previous period), the proportion of connective tissue decreases by 14,7%, with an increase in the relative area of bone tissue by 2,2 times.In the bone edge of the fracture zone, signs of resorptive changes appear in the form of some erosion of the bone edge. Compact bone tissue of the diaphysis in a state of active remodeling. There is a 19,6% decrease in the number of osteons. Dilated canals of osteons are revealed (by 14,7% compared to the previous period), filled with fibrous tissue with an increased content of cells and containing thin-walled blood vessels, the number of which is 77,0% of the previous period. In these canals, activated osteoblastic cells of the endosteum are visible, which are large in size, rounded basophilic nuclei. The number of osteoblasts increases by 36,7%. There are osteoclasts, the number of which also increases - by 36,4%. Along with this, the avascular and acitic areas of the bone are determined. This leads to a decrease in the number density of osteocytes by 24,1% compared with the previous observation period. A decrease in vascularization leads to bone remodeling and the development of porosity phenomena, as evidenced by the expansion of the osteon canals.

On the 21-28th day, the regenerate in the zone of the gunshot fracture is represented by dense fibrous tissue, in which the newly formed and rearranging bone tracts of the forming periosteal callus are visualized. Bone tracts of unequal size and different orientation Morphometrically in

the bone regenerate by the 28th day, a decrease in the relative content of fibroblasts and osteoblasts is determined, amounting to 85,6%, and 64,1%, respectively, from the indicators of the previous period. The content of mature fibrocytes increases by 13,8%, and osteocytes and osteoclasts – 1,9 and 2,9 times. The numerical density of the vessels, the size of their lumens and the cross-sectional area of the vascular bed have some tendency to decrease in comparison with the previous period. Calculation of the relative areas of the regenerate components showed an increase of 1,9 times in the relative area of bone tissue with a decrease in the connective tissue and vascular components by 25,8% and 8,5%, respectively.In this period, there is an increase in signs of resorption of the bone edge of the defect. The compact bone substance adjacent to the cavity of the defect is avascular, its edges are acitic and serrated. The compact bone substance in the endosteal zone preserves the lamellar structure of the bone substance throughout the fragments. Along the periosteal and endosteal surfaces, the fragments are closely soldered with a small-looped network of newly formed bone beams, and in some places the connection of the haversian canals of the compact substance with the interbeam spaces of the newly formed osteoid and bone tissues is visible. The central channels of compact substance osteons, mainly of a resorptively rearranging type, their number increases by 24,1%, contain a large number of cellular elements and dilated blood-filled vessels. The numerical density of the latter is 35,0% higher than the indicator of the previous period. At the same time, further expansion of the central canals of osteons is noted, the area of which is 974,10±23,40 μm2, which are characterized by an irregular shape. The number density of osteocytes and osteoclasts during this period of observation increases by 10,6% and 10,0%, respectively, with a tendency towards a decrease in the content of osteoblasts.

By the 35-42 day of observation in the interfragmental zone, spongy bone tissue from different thicknesses of bone beams is found, which has a lamellar and in places coarse-beam structure. Massive bone trabeculae are visualized in the periosteum zone, which merge to form a compact substance with extensive interbeam spaces and wide haversian canals of primary osteons.Spreading into the interfragmental zone, small areas of the newly formed cortical layer connect the ends of the fragments. Microscopically, the periosteum without a clear delimitation of the layers, and the surface of the cortical layer is uneven, with numerous resorption lacunae and narrow areas of deposition of the coarse-beam and lamellar bone. On the surface of the bone beams, osteoblasts, in places osteoclasts, can be mostly seen. Between the bone trabeculae - a loose fibroreticular tissue with a moderate amount of cellular elements is visualized.In the medullary canal, the spongy bone tissue has a wide-lobed character, the bone trabeculae are thinned, in the interbeam spaces - the cellular-fatty bone marrow. In some places, osteoblasts form irregular clusters on the surface of the bone beams. The cellular composition of the bone regenerate at this time of observation demonstrates a decrease in the proportion of fibroblasts (14,56±2,40%) with a slight increase in the number of fibrocytes. In the composition of the regenerate, the number of osteoblastic cells - osteoblasts - increases by 5,4%, and osteocytes – 1,8 times compared to the previous period.These changes occur against the background of the proliferation of osteoclasts, the relative content of which increases by 1,5 times and indicates active processes of restructuring of the bone tissue of the regenerate. The number of endothelial cells decreases 1,9 times. At the same time, the numerical density of the vessels and the cross-sectional area of the bloodstream decrease by 17,9% and 13,2%, respectively, compared with the previous period. The relative area of the bone tissue of the regenerate is 62,1± 2,5% on the 42nd day of observation and is 27,8% higher than the indicator of the previous period, the proportion of connective tissue decreases by 28,8%, and the relative proportion of vascular formations is 9,2±0,4 and slightly decreases compared to the previous period. The bone wall of the defect itself acquires an even character, which is associated with the active neoplasm of the osteoid in the form of homogeneous eosinophilic masses filling the areas of resorption of the maternal bone, from which the osteoid is separated by mild adhesion lines. The processes of neoplasm of bone matter are also observed in the bone structures adjacent to the wall of the defect, both from the side of the walls of the bone marrow spaces and the canals of osteons. As a result, we can talk about bone compaction. This is also evidenced by a 9,5% increase in the number density of osteons, a 21,5% decrease in the area of their central canals. The normalization of the structure of bone tissue is evidenced by an increase of 22,6% compared with the previous period in the number of osteocytes, a decrease by 19,5% and 18,0%, respectively, in the number of osteoblasts and osteoclasts. The process of replacing areas of bone loss with newly formed bone substance at this time is active and creates a general picture of structural compensation of bone tissue. Productive processes with the differentiation of newly formed structures, their maturation and a gradual return of the characteristics of the tissue components of bone tissue to normal become prevailing. Nevertheless, a significant number of dilated osteon canals remain in the bone tissue, giving it in some areas the appearance of spongy bone tissue.

At the final stage of observation, already formed trabeculae in the central parts of the defect are microscopically determined, which merge with each other and the edges of the defect, an increase in mineralization processes is noted. It should be noted the formation of a typical lamellar bone tissue with intensive restructuring of bone tissue, as well as restoration of the processes of organotypic histoarchitectonics with the formation of a cancellous bone and an increase in the proportion of bone tissue in the regenerate.The presence of young bone structures, which have different sizes and different orientations, are also determined. The bone canals that pierce them have an irregular shape and are enlarged. It contains connective tissue rich in cellular elements, in places infiltrated by leukocytes. The outer surface of the bone beams is covered with layers of osteoid. The periosteum is already clearly differentiated with the outer fibrocellular

and inner cell layers. External general plates are formed along the outer rough surface of the cortical layer. The cellular composition of the regenerate at this time is represented mainly by osteocytes, the relative content of which is 32,3±1,9%, the relative content of fibrocytes remains at a fairly high level, the relative content of which is 20,5± 2,2% of the total number of cellular elements. The relative proportion of osteoclasts remains at a high level (6,53±0,21%), which indicates their joint participation with osteoblasts in regenerative endosseous osteogenesis and remodeling of bone regenerate. The preservation of the number density of vessels at the level of the previous period and the tendency to narrowing of their lumens lead to a decrease in the cross-sectional area of the bloodstream by 14,4%, which is apparently a physiologically determined process, since it does not affect the intensity of the processes of restructuring and calcification of bone tissue. This is confirmed by the calculation of the relative areas of the tissue of the regenerate, mainly represented by the bone component – 75,9±3,2%, with a decrease in the relative proportion of fibrous tissue by 2 times.

Conclusion

Thus, in our experiment, we obtained a full consolidation of the fracture, an important condition for this was the correct reposition of the fragments and their stable fixation, which excluded additional traumatization of the vessels in the fracture zone, while the hemodynamics of the regenerate was minimally disturbed. Which leads to increased tissue trophism, accelerated restructuring and mineralization of the regenerate The method of stable out-of-focal compression-distraction osteosynthesis provides optimal conditions for regeneration and restoration of the injured limb.

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