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Experimental Substantiation of the Course of Osteoreparation in the Intermediate Fragment in Case of Long Limb Polypocal Fractures Depending on the Method of Osteosynthesis

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Abstract **Purpose of the study:** to investigate reparative osteogenesis in the treatment of polyfocal injuries of long bones of the extremities in an experiment, using the exapmle of a tibial fracture. **Materials and methods:** an experimental study was conducted on an experimental model of a transverse segmental fracture in the lower third of the tibial diaphysis in rabbits. An analysis of the experimental study was carried out jointly with the National Medical Research Center for Traumatology and Orthopedics named after G.A. Ilizarov, Kurgan, Ministry of Health of the Russian Federation. In a group of animals, a double fracture of the diaphysis was simulated, and the fracture was fixed with an Ilizarov apparatus. Duration of the experiment: 28 days, 56 days of fixation and 35 days after dismantling the device. The total duration of the experiment was 91 days. **Results:** an analysis of the results of morphological changes in the fusion zones of a simulated double fracture on the tibia was carried out under the conditions of transosseous osteosynthesis according to Ilizarov and combined osteosynthesis in order to comparatively assess their effectiveness. The results of the experimental study showed that during microscopic examination using light microscopy, it was noted that with the orrect comparison of fragments, in the absence of their displacement, it is possible to achieve strong bone fusion at all levels by the end of the period of fixation in the apparatus after 56 days of the experiment. Combination of the transosseous osteosynthesis method according to G.A. Ilizarov and intramedullary reinforcement with a metal pin in the treatment of polyfocal fractures contributes to the activation of periosteal and endosteal osteogenesis, along with intramedullary osteogenesis, stimulation of angiogenesis in the intramedullary zone. This leads to the formation of complete bone fusion in the area of proximal and distal tibial fractures by 56 days of fixation. The created conditions help to reduce the porosity of the compact plate of bone fragments and the free median bone fragment, as well as the formation of more compact bone in the fusion area compared to the comparison series. In addition, stabilization of the distal fragment due to additional fixation with a reinforcing intarmedullary wire leads to more rapid restructuring of the vascular bed in the distal part of the tibia.

Keywords Polyfocal fractures, Transosseous osteosynthesis, Intermediate fragment, Reparative osteogenesis

1. Introduction

The treatment of polyfocal (double) fractures of long bones of the extremities remains one of the unresolved issues in traumatology. Double fractures account for 1.5-2.2% of all fractures of tubular bones. [1,2]

The need to classify double fractures as a separate group is determined by two factors. Firstly, these injuries are multiple in nature, and secondly, double fractures present a number of biological and biomechanical features that complicate the processes of reparative bone regeneration. [3,4]. These features are due to the presence of an intermediate bone fragment, which is excluded from the intraosseous blood flow, leading to significant hemodynamic disturbances. [5,6]. As a result, in 10% of cases, regardless of the treatment method, the fragment undergoes aseptic necrosis. Peripheral circulation disorders arise both from damage to the vascular bed (especially intraosseous nutrient arteries) and from damage to the endosteum, which plays a significant role in the reparative process of bone tissue. Double fractures of the diaphysis of long tubular bones are accompanied by more extensive damage to adjacent soft tissues (including blood vessels and nerves), which also negatively affects the process of fracture consolidation.

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The aim of this study was to substantiate the course of bone tissue regeneration in polyfocal fractures of long bones of the extremities under different methods of osteosynthesis.

2. Materials and Methods

The study was conducted on clinically healthy male rabbits of the Soviet Chinchilla breed, aged 14-16 months, weighing 4.4 (4.3-4.85) kg. The length of the tibia in the animals was 123.3 (121.35-121.65) mm. The animals were divided into the following experimental series and groups:

- Series 1 (Ilizarov method) (n=6) – osteosynthesis of the fracture using the Ilizarov method.
  Duration of the experiment: 56 days post-operation (56 days of fixation) (n=3) and 91 days post-operation (35 days after removal of the apparatus) (n=3).
- Series 2 (Combined osteosynthesis) (n=6) – osteosynthesis of the fracture using the Ilizarov method with additional intramedullary osteosynthesis with a stainless steel pin.
  Duration of the experiment: 56 days post-operation (56 days of fixation) (n=3) and 91 days post-operation (35 days after removal of the apparatus) (n=3).

In the operating room, a complete transverse polyfocal fracture was simulated in the lower third of the tibia. The length of the free (middle) fragment was 20.3%, and the length of the distal fragment was 16.26% of the length of the tibia. The fracture was complicated by circular detachment, compression, and overstretching of soft tissues over 1/3 of the segment, as well as complete damage (transection) of the bone marrow at two levels. Thus, the combination of these features indicates that a severe form of polyfocal fracture was reproduced.

Fixation of the short fragments (middle fragment and distal fragment) was performed at one level, in the middle of the longitudinal axis of the fragments. The frontal planes of the wire insertion were at a minimal distance from each other – 2 mm. Fixation of the short fragments at two levels or insertion of wires at a distance of 5 mm or more (closer to the ends of the fragments) was not possible due to the risk of longitudinal or marginal splitting of the fragments.

To evaluate the results, methods of in vivo observation, radiographic examination, and morphological analysis were used.

3. Results

In the early postoperative period, the condition of the animals was satisfactory, with an increase in body temperature to 39.8°C. Appetite was reduced. Lameness of the operated limb of the supporting type was observed. Serous-hemorrhagic exudate was discharged from the surgical wound through the drain in moderate amounts. Pain was severe. By days 6-12, the condition of the animals improved, and appetite was restored. The animals moved freely in the cage, but lameness of the operated limb of the supporting type persisted. Body temperature decreased to the lower limit and was 38.2-38.5°C. Pain in the intervention area was moderate. By days 14-16, the general condition of the animals was good, and food and water intake were normal. The surgical wound healed, and the swelling of the limb subsided. Lameness persisted. Sutures were removed. Pain was moderate. At the final stage of observation – 35 days after removal of the apparatus, the condition of the animals remained good, and food and water intake were normal. The rabbits moved on all limbs, and lameness and pain were absent.

In the postoperative period, the development of passive flexion and extension contractures of the knee and ankle joints of the operated limb was observed in both Series 1 (using transosseous osteosynthesis according to Ilizarov) and Series 2 (using combined osteosynthesis). [7,8,9]. The severity of joint contractures at 56 days of fixation was higher in Series 2. By the end of the non-apparatus period, the contracture of the knee joint tended to decrease, while the contracture of the ankle joint slightly increased. The development of contractures in the animals was associated with the fixation of the segment with an external fixation device, the severity of the surgical intervention, and the physiological characteristics of the rabbit – the habitual sitting posture with the limbs tucked under the body. The worsening of ankle joint contractures in the non-apparatus period was associated with the severity of the fracture and the absence of therapeutic exercises for limb development (physical therapy was not part of the study objectives). [10,11,12].

On radiographs, by day 14 post-operation, displacement of the fragments was observed in both Series 1 and Series 2. However, in Series 1, displacements occurred in 83.35% of cases, while in Series 2, they occurred in 33.34% of cases. In Series 1, displacement of the middle (free) and distal fragments was noted. In most cases, recurvatum displacement of the fragments at the proximal level (50.01%) was observed, with a magnitude of 172-175°, and antecurvatum angular displacement at the level of the distal fracture (50.01%) with a magnitude of 150-177°. Lateral displacement of the middle fragment was also noted in 33.34% of cases. In Series 2, angular displacement of the proximal end of the middle fragment forward (16.67%) was observed in one animal, leading to antecurvatum deformity at the level of the proximal fracture with a magnitude of 176° [13,14,15]. In the second rabbit, displacement of the proximal end of the distal fragment forward was noted, leading to antecurvatum deformity at the level of the distal fracture with a magnitude of 170°. Displacement of the middle fragment in width was not observed [16,17,18].

The frequency and variability of fragment displacement in Series 1 were due to the single-level fixation of the middle and distal fragments in their central part. The cause of fragment displacement in the sagittal (anteroposterior) plane was the traction effect of the extensor tendons of the ankle joint and compression of the fragments for repositioning after osteotomy, which provoked the occurrence of a torque on the middle and distal fragments. [19,20,21]. Lateral displacement of the middle fragment occurred due to the action of the lateral muscles of the lower leg. Displacement of the distal (articular) fragment occurred due to movement of the limb in the ankle joint. The occurrence of displacements was influenced by the absence of limb immobilization in the postoperative period – the animals used the limb without restriction.

In Series 2, displacements were noted in two animals and only in the sagittal plane. The reason for this was the intramedullary pin, which created an additional axis of stability for the fragments. It was noted that with further observation, no worsening of fragment displacement or angular deformities was observed.

By day 28 of fixation, periosteal layers appeared on the surface of the fragments, located on the lateral, anterior, and posterior surfaces of the bone. On the medial surface of the fragments, the shadows were weak or not visualized. A greater volume of shadows was determined in animals in Series 1. By day 42, signs of delayed osteogenesis were noted in two rabbits in Series 1, expressed in the rounding of the ends of the fragments, and the absence of a closing plate. Signs of fracture healing were noted – bone bridges connecting the fragments.

By day 56, fracture healing was noted at both levels. No cases of pseudarthrosis formation were identified. In the area of the distal fracture, due to fragment displacement, healing in the diastasis was partial, in the form of separate strands (bridges). An additional source of healing was the periosteal layers, which were massive, voluminous, and widely distributed mainly on the lateral, anterior, and posterior surfaces. This allowed avoiding refractures in the post-apparatus period.

In Series 1, the cortical plate of the middle fragment, both in the direct and lateral projections, had a zone of enlightenment in the middle part along the entire fragment. The outer and inner walls of the cortical plate had increased radiological density. As a result, the cortical plate of the fragment had a layered appearance. In Series 2, this phenomenon was noted only in two animals and was weakly expressed. The appearance of small foci of enlightenment, without clear contours, was noted in 50% of animals in the distal part of the middle fragment, with a decrease in radiological density in this zone.

By day 70 post-operation (14 days without the apparatus), organotypic restructuring of the bone in the fracture zones occurred. In the cortical plates of the free (middle) fragment in Series 1, signs of bone tissue destruction were preserved, and their contour became clearer. In Series 2, signs of layering appeared in the cortical plate of the middle fragment, and in one animal, single rounded areas of enlightenment (cysts) with a diameter of 0.1-0.3 mm were noted.

By day 91 post-operation (35 days without the apparatus), further anatomical restructuring of the tibial structures occurred in Series 1. In the area of the proximal and distal fractures, further restoration of the anatomical structures of the bone – cortical plates and the intramedullary canal – occurred.

In Series 1, the layering of the cortical plate of the middle fragment was preserved. Signs of destruction of the cortical plate were noted, expressed in the presence of chaotically located cysts with a width of 0.1 to 0.8 mm, of irregular shape, in the projection of the intramedullary canal. Such areas were noted in all animals. The condition of the fragment worsened with a decrease in bone mineral density.

In Series 2, the layering of the cortical plates was preserved but less pronounced. Periosteal layers on the surface of the middle fragment had the appearance of weakly expressed single smoothed exostoses. In the projection of the intermediate canal, the bone had a uniform structure. The radiological density in the distal and proximal parts of the fragment was lower than in the middle part. The areas of enlightenment identified at the previous stage (BA 14) became less pronounced and had an unclear contour.

When comparing the sizes of the fragments and radiological density indicators with the initial values, it was found that the linear dimensions had insignificant deviations from the operational values, and the radiological density indicators often exceeded the preoperative values. Radiological density below the preoperative values was noted in the medial cortical plate of the distal and middle fragments, the lateral cortical plate, and the intermediate canal of the proximal end and the middle part of the middle fragment. This indicates the absence of bone tissue atrophy, the presence of signs of sclerosis, and the loss of mineral density, "impoverishment" of the free fragment.

Healing of the proximal fracture occurred more favorably due to more stable fixation of the fragments. Healing of the distal fracture, due to micromobility of the distal fragment, occurred through paraosseous tissues.

In the cortical plate of the middle (free) fragment in Series 1, by the end of fixation (F56), signs of bone tissue destruction were identified, manifested in the "layering" of the cortical plate and the appearance of small cysts in it. In Series 2, such signs appeared later – by day 14 after removal of the apparatus.

By the end of the observation period (35 days BA), signs of the process of restoration of the cortical plate were noted in Series 2, while in Series 1, the condition of the bone worsened with a decrease in bone mineral density.

4. Discussion

The analysis of the obtained results of physiological studies showed that in all experimental series, under the created conditions, no disturbances in thermoregulation or heart rhythm were observed in the animals. This was evidenced by the absence of significant fluctuations in body temperature and heart rate.

When assessing the features of blood circulation in the area of surgical intervention, no significant changes in the studied parameters were predominantly recorded. There were no significant differences either in comparison with the physiological norm or between the series at different stages of the experiment.

In general, based on the results of the study, it can be concluded that the most favorable conditions for blood circulation in the tissues of the surgical intervention area were created in the second experimental series. Thus, no disturbances in the blood supply to the skeletal muscles of the lower leg were identified.

The conducted microscopic examination using light microscopy showed that with proper alignment of the fragments and in the absence of their displacement, the transosseous osteosynthesis method according to Ilizarov in the treatment of multilevel fractures allows achieving strong bone fusion at all levels by the end of the fixation period in the apparatus after 56 days of the experiment. [17,18,19].

In the proximal inter-fragmentary diastasis, the most mature and voluminous callus was formed. In this area, its formation involved cells of the osteon channels, osteogenic cells of the endosteum, and periosteum. Whereas in the distal regenerate, fusion was achieved through osteogenesis in the intermediate zone. Greater porosity was noted in the median bone fragment and in the area of the distal fragment, which tended to worsen with increasing duration of the experiment. The combination of the transosseous osteosynthesis method according to G.A. Ilizarov and intramedullary reinforcement in the treatment of polyfocal fractures contributes to the activation of periosteal and endosteal osteogenesis, along with intramedullary osteogenesis, and stimulates angiogenesis in the intramedullary zone. This leads to the formation of complete bone fusion in the area of proximal and distal tibial fractures by 56 days of fixation. The created conditions help reduce the porosity of the compact plate of bone fragments and the free median bone fragment, as well as the formation of more compact bone in the fusion area compared to the first series.

In addition, stabilization of the distal fragment due to additional fixation with a reinforcing intramedullary wire leads to more rapid restructuring of the vascular bed in the distal part of the tibia.

Morphometric examination of the numerical density of blood vessels showed that the use of combined osteosynthesis creates more favorable conditions for the formation of bone fusion of the distal tibial fracture, stimulates angiogenesis in the bone marrow canal, and contributes to a reduction in the porosity of the median bone fragment and the distal fragment. In the proximal regenerate, greater stability of bone fusion under conditions of combined osteosynthesis was ensured by the formation of an additional volume of endosteally and periosteally formed bone tissue, induced by intramedullary reinforcement. Greater stability of the distal bone fragment contributed to more rapid restructuring of the vascular component of the bone towards organotypicity.

Histomorphometric analysis of the volumetric fraction of bone tissue showed that the use of combined osteosynthesis creates more favorable conditions for the formation of bone fusion of fragments in the distal part of the segmental tibial fracture during the period of apparatus fixation. In the proximal zone of fragment fusion, with equal volumetric fractions of bone tissue, the use of combined osteosynthesis determines its higher volumetric density at the level of the bone marrow cavity.

In the non-apparatus period, intergroup differences in the volumetric fraction and density of bone tissue in the distal part of the fusion zone are leveled out as a result of adaptive restructuring of the newly formed bone areas. In the proximal part of the fusion zone, with equal values of the volumetric fraction, the density indicators of bone tissue change multidirectionally – under conditions of osteosynthesis with the apparatus, the density of bone tissue increases, while under conditions of combined osteosynthesis, it decreases. This observation indicates differences in mechano-biological conditions, presumably – less mechanical stability in the proximal zone of healing of the segmental fracture after removal of the external fixation device without additional intramedullary fixation.

The use of combined osteosynthesis ensures more accurate alignment of bone fragments during the period of apparatus fixation, as objectively evidenced by a reduction in the height of the fusion zones of the segmental fracture, statistically significant in its distal part. At the same time, in both experimental groups, the height of the fusion zones does not exceed 2 mm, which is within the optimal values of this evaluation parameter. An indirect sign of stable alignment of bone fragments under conditions of combined osteosynthesis is a significant reduction in the height of the callus throughout the operated area, as well as the thickness of the compact plate of the free bone fragment. The absence of objective differences in the values of other histomorphometric evaluation indicators indicates the fundamental similarity of the conditions for the course of the reparative process and reactive post-traumatic reorganization of bone tissue in both experimental groups.

5. Conclusions

Thus, the main factors influencing the course of bone tissue repair in polyfocal fractures are the severity of the traumatic impact and the stability of fragment fixation.

1. Severity of the traumatic impact on the segment. In our experiment, the traumatic impact was applied to the bone (fracture at two levels) and to the soft tissues – on the medial surface of the segment (skeletonization of the bone, tissue detachment, etc.). The result of these manipulations was poor blood supply to the structures of the periosteum, bone, and bone marrow, which was expressed in weak periosteal layers on the medial surface of the diaphysis of the bone, and destructive changes in the cortical plate of the middle and distal fragments.

The tissues of the lateral and anteroposterior surfaces of the segment were not subjected to traumatic impact, except for the insertion of wires for osteosynthesis. According to the results of physiological studies, they maintained their blood supply throughout the observation period. The preservation of the blood supply to the skeletal muscles served as a source of blood supply for the tissues of the lateral and anteroposterior surfaces of the bone, as evidenced by the active process of bone tissue regeneration in these areas of the bone.

2. Stability of fragment fixation. Due to the small size of the free and distal fragments, 20.3% and 16.26%, respectively, the fragments were fixed at one level in the central part, and the wires were inserted at a minimal distance from each other (2 mm). Fixation of the fragments at two levels or insertion of wires at a greater distance from each other in height was not possible due to the risk of bone splitting. The performance of reduction (compression) of the fragments and the traction forces of the tendons led to the occurrence of torque and displacement of the fragments. Displacement of the fragments was less common in animals with osteosynthesis using the Ilizarov method combined with intramedullary osteosynthesis, as the pin created stability of the fragments along the longitudinal axis of the bone.

In osteosynthesis using the Ilizarov method, after gross displacement of the fragments in the first 14 days post-operation, due to the fact that the animals used the limb without restriction, micromobility of the fragments persisted in the fracture zones until the periosteal layers compacted to a degree that allowed stabilization of the fragments (42 days of fixation). The response to micromobility of the fragments was the proliferation of the periosteum on the lateral, anterior, and posterior surfaces of the bone fragments. In addition, micromobility of the fragments led to local microtraumatization of tissues and the vascular bed in the diastases between the fragments, which slowed down the processes of bone regeneration and provoked the appearance of signs of pseudarthrosis formation.

Condition of the middle (free) fragment.

The intraosseous nutrient artery in rabbits enters the tibia at the level of its junction with the fibula – above the level of the proximal fracture. As a result of the performed osteotomy at two levels, the middle fragment was deprived of intraosseous blood supply, both from the proximal and distal ends. Destruction of the bone marrow structures and the nutrient artery, combined with micromobility, caused a deficiency in the nutrition of the middle fragment, which contributed to the development of destructive processes in the cortical plate by the end of fixation.

The condition of the bone tissue of the middle fragment was compromised in animals in both experimental series. In rabbits with combined osteosynthesis, by day 35 after removal of the apparatus, signs of restoration of the fragment structures were noted, while in the series using the Ilizarov method, no tendency towards restoration was observed. Most likely, the restoration of the bone structures of the free fragment will occur, but at a later stage.

Despite the above-mentioned signs complicating the process of reparative regeneration of bone tissue in polyfocal fractures, strong bone fusion of the fractures was achieved in both experimental series. No cases of refractures were identified in the non-apparatus period.

Thus, when choosing a method of osteosynthesis for polyfocal fractures, the size of the fragments should be taken into account. If the length of each fragment allows for the insertion of fixators at two levels, thereby ensuring reliable stabilization, then the Ilizarov method should be used, as it will eliminate additional trauma to the bone marrow. If the length of the fragments is 20% of the bone length or less, it is better to resort to a combined method of osteosynthesis.

To improve the results of treatment of polyfocal fractures, the following should be considered:

1. Exclude early loading of the limb during the first 14 days to prevent fragment displacement;
2. In the later periods of fixation, begin the development of adjacent joints of the limb to prevent the development of contractures;
3. Conduct drug therapy to restore intraosseous blood supply;
4. Conduct drug therapy to replenish the deficiency of vitamins and minerals in the body for the restoration of bone tissue.

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