BIOGERONTOLOGY

Reparative Process in Bone Tissue of Old Animals

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Plastic repair of the trephination hole in the parietal bone with syngeneic cartilage was carried out in old rabbits. The ossification process eventuated in the formation of a callus by day 28 postoperation, while in control animals bone tissue defect still persisted during this period.

Key Words: *reparation; osteogenesis; syngeneic cartilage*

Aging is associated with significant changes in the functions of the immune, endocrine, and hemopoietic systems leading to metabolic disorders in tissues of the aging organism [2,5,7]. Bone tissue (BT) reacts to these changes by a sharp decrease in bone remodeling [11]. It is known that aging is accompanied by the development of tissue resistance to glucocorticoids modulating activities of osteoclasts and osteoblasts [3,9,10]. Bone mineralization decreases during aging, which creates prerequisites for the development of osteoporosis [1,4, 6,8]. Age-related changes in the bone tissue, no doubt, have a negative impact on reparation of bone defects. This necessitates the search for new methods stimulating bone remodeling in aging organism.

We developed a method for optimizing the conditions favoring acceleration and normalization of the reparative process in old animals with BT injuries.

MATERIALS AND METHODS

Female Chinchilla rabbits (n=18; 5.0-5.5 kg) aging 5-6 years were divided into control and experimental groups. The animals were kept in a stationary vivarium on standard ration and free access to water.

Experimental animals were narcotized with ketamine, the cartilage was removed from the costosternal articulation and placed into a sterile Petri dish with saline. Directly after this, the skin on the head was cut, the epicranial aponeurosis was dissected, and a hole (2×2 cm) in the parietal bone was made with a trephine. Bleeding from emissary veins was arrested with a hemostatic sponge. Cartilaginous tissue, sliced transversely, was placed between two layers of a hemostatic sponge. The lower edge of the sponge prevented contact between the transplanted cartilage and brain structures, due to which the defect created in the bone could be completely closed without mechanical damage to the brain.

In controls, trephination was carried out similarly and the bone defect was closed with a skin flap.

Operation wounds were sutured layer-by-layer and the sutures were treated with iodine solution in ethanol.

The animals were sacrificed on days 7, 21, and 28. Fragments of the parietal bone for morphological study were fixed in neutral formalin; sections were prepared and stained with hematoxylin and eosin or with picrofuchsin after van Gieson.

RESULTS

On day 7, bone tissue defect occupying several visual fields and filled with dissociated cartilage

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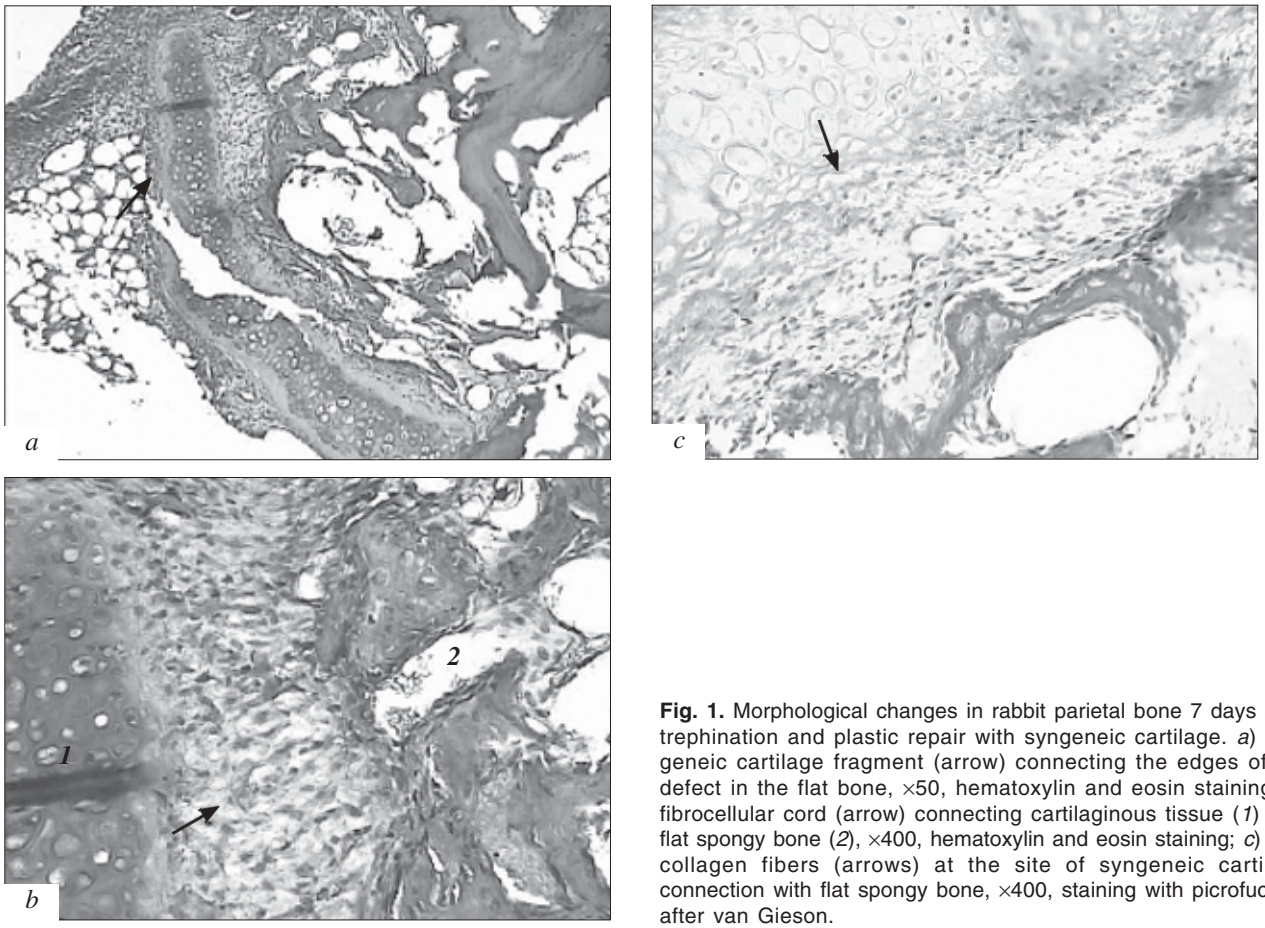

Fig. 1. Morphological changes in rabbit parietal bone 7 days after trephination and plastic repair with syngeneic cartilage. *a*) syngeneic cartilage fragment (arrow) connecting the edges of the defect in the flat bone, ×50, hematoxylin and eosin staining; *b*) fibrocellular cord (arrow) connecting cartilaginous tissue (*1*) with flat spongy bone (*2*), ×400, hematoxylin and eosin staining; *c*) new collagen fibers (arrows) at the site of syngeneic cartilage connection with flat spongy bone, ×400, staining with picrofuchsin after van Gieson.

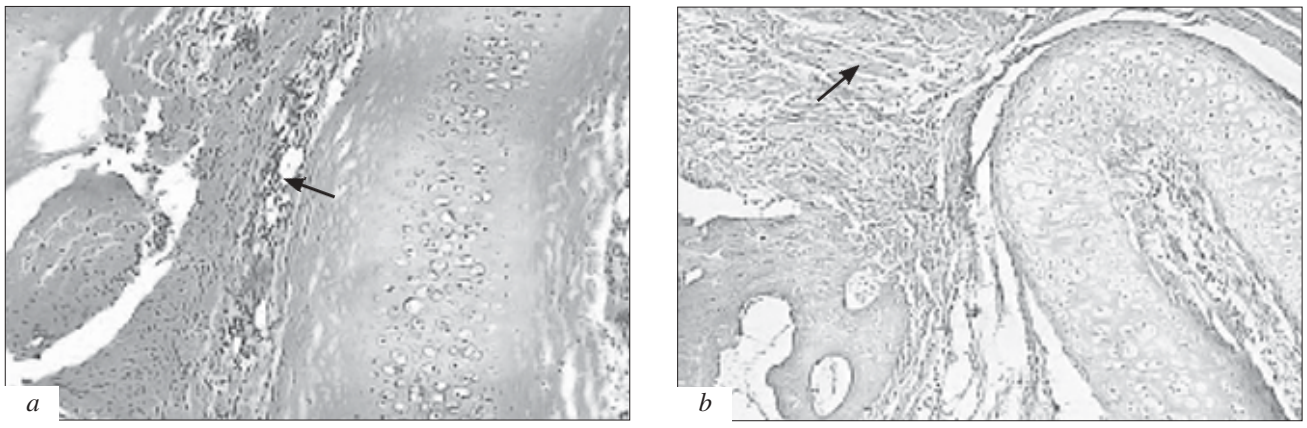

Fig. 2. Morphological changes in rabbit parietal bone 21 days after trephination and plastic repair with syngeneic cartilage. *a*) osteoclast accumulations (arrow) around syngeneic cartilage fragment, ×200, hematoxylin and eosin staining; *b*) compactly packed collagen fibers (arrows) in young granulation tissue, ×200, staining with picrofuchsin after van Gieson.

fragments was seen (Fig. 1, *a*). Cartilage fragments were connected with bone beams via fibrocellular cords containing primarily young fibroblasts (Fig. 1, *b*) and few fine collagen fibers (Fig. 1, *c*). Neutrophilic leukocytes with an admixture of lymphoid cells and few macrophages were also noted (Fig. 1, *b*).

On day 21 after the intervention, a callus was seen at the site of the defect on the X-ray picture. The histological picture of this reparation period was presented by small fragments of the cartilage tightly adhering to BT defect walls due to compact packing of collagen fibers (Fig. 2, *b*). Pronounced resorption was paralleled by intensive formation of new capillaries (Fig. 2, *a*).

On day 28 after the intervention, X-ray picture showed well-formed callus at the site of the defect.

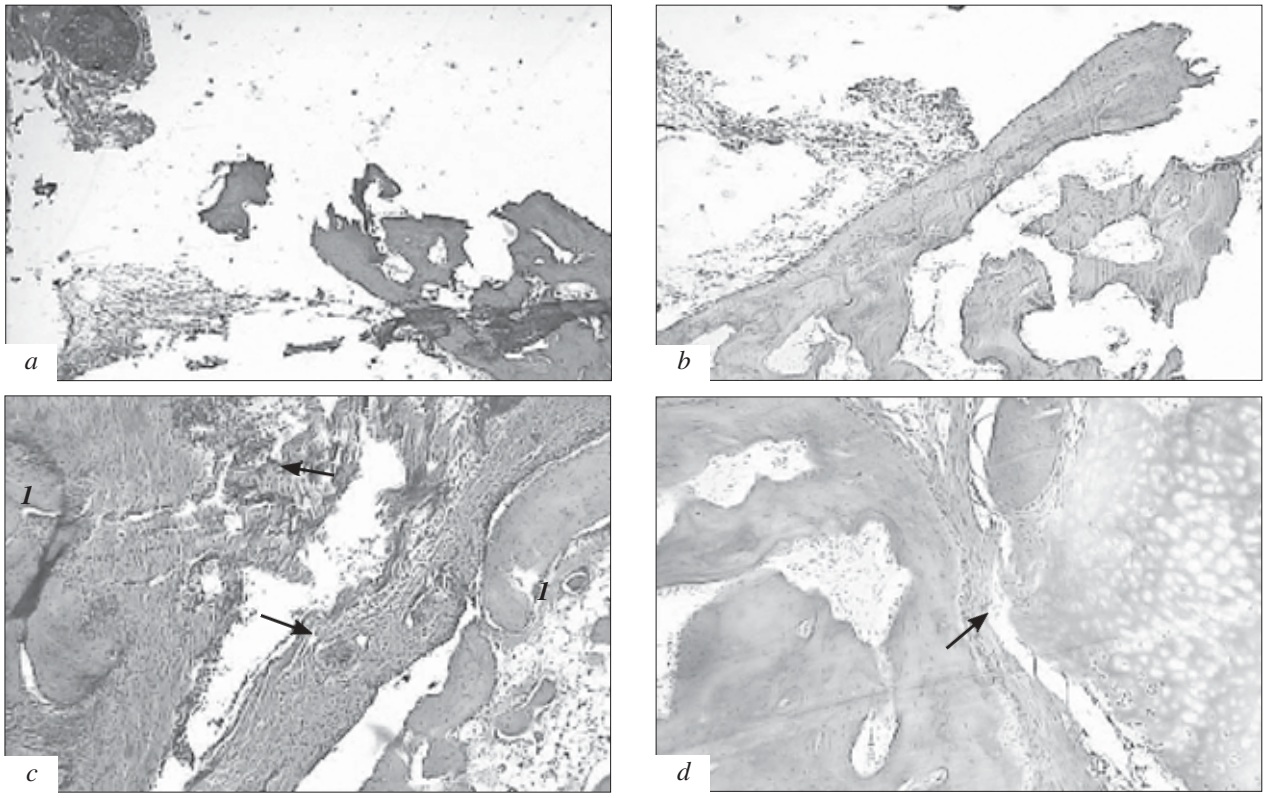

Fig. 3. Morphological changes in rabbit parietal bone 28 days after trephination (*a*, *b*) and plastic repair with syngeneic cartilage (*c*, *d*). *a*) edge of tissue defect in flat spongy bone, ×100; *b*) intense absorption of basic fuchsin along the edge of the bone defect, ×100; *c*) connective-tissue bridges (arrows) between the defect edges (*1*) in flat spongy bone, ×100; *d*) compactly packed collagen fibers (arrow) in mature granulation tissue, ×200.

The histological picture corresponded to the start of the ossification process. Fibroblasts in connective tissue cords acquired a spindle-like shape (Fig. 3, *c*). A clearly seen cord containing fibroblasts was seen along the entire surface of the defect between bone membranes (Fig. 3, *d*).

In contrast to this dynamics of the reparative osteogenesis, X-ray picture of the bone in controls still showed bone defect 28 days after trephination. Bone tissue of the defect walls was histologically homogeneous and intensely stained with acid fuchsin (Fig. 3, *b*). The periosteal reaction was seen at the defect edges and slight accumulation of fibroblasts along the entire perimeter was noted (Fig. 3, *a*).

Hence, the results indicate that BT reparation in old animals is significantly inhibited. Reproduction of conditions for endochondral osteogenesis by plastic repair of the BT defect with syngeneic cartilage in old animals stimulated reparative processes, which resulted in acceleration and normalization of BT recovery at the site of injury.

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