UDC 636.92.09:616-001.5/.073:591.83 **doi:** https://doi.org/10.15407/ubj98.01.098

REPARATIVE OSTEOGENESIS MARKERS DURING BONE DEFECTS SUBSTITUTION WITH GERMANIUM-DOPED CERAMICS UNDER EXPERIMENTAL OSTEOPOROSIS

T. TODOSIUK[1]✉, V. CHEMEROVSKIY[1], A. RUBLENKO[2], N. ULIANCHYCH[3], V. KOLOMIIETS[3], S. FIRSTOV[3]

[1]Bila Tserkva National Agrarian University, Bila Tserkva, Ukraine;
[2]Vinnytsia Mykhailo Kotsiubynskyi State Pedagogical University, Vinnytsia, Ukraine;
[3]Frantsevich Institute for Problems of Materials Science, National Academy of Sciences of Ukraine, Kyiv;
✉e-mail: tatyana.todosyuk@gmail.com

Received: 04 June 2025; **Revised:** 05 August 2025; **Accepted:** 30 January 2026

Osteoporosis, as a systemic skeletal disease, is characterized by the loss of bone mass, decreased mineral density, and microarchitecture changes. In cases of traumatic fractures and critical-size bone defects osteoporosis can lead to spontaneous fractures, impair regeneration and complication when using bone substituting materials. Ceramic implants, doped with germanium to impart osteoinductive properties, are among promising bone substituting materials. In this study we aimed to assess biochemical markers of reparative osteogenesis at bone defects substitution with germanium-doped ceramics in rabbits under osteoporosis. The study was conducted on California White rabbits with osteoporosis, induced by administration of 0.4% dexamethasone solution. The model defects were created in trabecular and cortical bones, following the exposure of the periosteum with drills of 3 and 4.2 mm diameters, respectively, in compliance with the anesthetic regimen and antiseptic rules. Calcium phosphate ceramic granules with a size of 700 μm, synthesized from hydroxyapatite and β-tricalcium phosphate and doped with 0.8 mass.% germanium (CPC-Ge) were used for healing. In the control group of animals (n = 9) bone defects were healed under a blood clot. In the experimental group (n = 9), the defects were replaced with CPC-Ge granules. Blood samples for biochemical studies were collected before modeling the bone defect and on the 7th, 14th, 30th, and 60th days of reparative osteogenesis. The activity of tartrate-resistant acid phosphatase, alkaline phosphatase and its bone isoenzyme, as well as circulating immune complexes, protein C and NO serum levels were determined. It was shown that substitution of both trabecular and cortical bones defects with CPC-Ge, as compared to healing under a blood clot, leads to reduced inflammatory and immune responses, prevented the depletion of protein C and promotes a more dynamic course of reparative osteogenesis in animals with glucocorticoid-induced osteoporosis.

K e y w o r d s: germanium, calcium phosphate ceramic granules, osteosubstitution, osteoporosis, reparative osteogenesis, phosphatases, protein C, immune complexes, nitric oxide.

Bone substitution and the restoration of bone defects caused by trauma, bone infections, neoplasms, or degenerative-dystrophic processes is a complex and multidisciplinary task involving various fields of human and veterinary medicine [1-4].

Among the risks of intensive bone resorption and spontaneous fractures, osteoporosis holds a prominent place [5-7]. It has reached epidemic proportions and involves genetic, hormonal, metabolic, and immunological mechanisms [8-10].

Osteoporosis, as a systemic skeletal disease, is characterized by the loss of bone mass, decreased bone mineral density, and changes in the microarchitecture and biomechanical properties of long tubular bones and vertebrae [11-13]. This leads to pathological fractures with a high mortality rate and impairs the process of osteointegration of implants during osteosynthesis and joint replacement surgeries [14-16].

Osteoporosis can lead to spontaneous low-energy fractures and impair regeneration in cases

of traumatic high-energy fractures and critical-size bone defects [17-19]. Disruption of bone metabolism results in an imbalance within the osteoprotegerin-cytokine system and regulatory mechanisms of the immune system, which play a key and direct role in reparative osteogenesis [20-21].

Numerous cytokines, hormones, and pharmaceuticals either stimulate or inhibit the effects of the receptor activator of nuclear factor ligand (RANKL) or the soluble receptor osteoprotegerin (OPG) [22, 23]. The latter inhibits osteoclast differentiation, thereby shifting the balance within the RANKL/OPG system [24, 25]. Excessive production of inflammatory mediators leads to bone tissue remodeling and impaired reparative osteogenesis due to the predominance of a pro-inflammatory macrophage phenotype in the affected area [26-28]. This results in a complicated course or even failure of osteointegrative coatings or bone-substituting implants [29-31].

In this context, local or systemic osteoporosis imposes specific requirements on biomaterials: osteoconductive and osteointegrative properties, controlled degradation, biocompatibility, avoidance of additional resorption of the surrounding host bone, induction of neoangiogenesis, the ability to shift the pro-inflammatory macrophage phenotype (M1) toward an anti-inflammatory (regenerative – M2) phenotype, and promotion of mesenchymal stem cell adhesion, proliferation, and differentiation into osteoblasts [32-34].

Among such promising alloplastic materials are ceramic implants, which account for approximately 60% of the synthetic graft biomaterials market [35, 36]. Given a number of biological properties, doping hydroxyapatite ceramics with silicon or germanium ions may impart osteoinductive properties to the material [37, 38].

To evaluate the effectiveness of bone-substituting materials under osteoporotic conditions, we aimed to assess biochemical markers related to cytokine system activation, inflammatory response, and hemostasis system activity. Specifically, we examined phosphatase activity, anticoagulant protein C levels, nitric oxide concentration, and circulating immune complex levels during bone replacement using germanium-doped calcium phosphate ceramics.

Materials and Methods

Calcium phosphate ceramics. For the studies, granules of non-doped biphasic ceramic, synthesized in the Laboratory of Biomaterials for Bone Tissue Engineering at the Frantsevich Institute for Problems of Materials Science, NAS of Ukraine were used. The granules, with a size of 700 μm (GTg-700), consisted of 65 mass.% hydroxyapatite (HA) phase and 35 mass.% β-tricalcium phosphate (β-TCP).

Doping the calcium-phosphate ceramic with germanium (GTlGer-700) was achieved by introducing 1.0 mass.% germanium, via metaphosphate germanium – $Ge(PO_3)_4$ in the form of a colloidal solution into the freshly precipitated hydroxyapatite gel. The synthesis of stoichiometric HA – $Ca_{10}(PO_4)_6(OH)_2$ was performed using the traditional chemical precipitation method, by mixing solutions of calcium nitrate tetrahydrate ($Ca(NO_3)_2 \cdot 4H_2O$) and ammonium hydrogen phosphate ($(NH_4)_2HPO_4$) with a calculated Ca/P ratio of 1.67. After sintering, the germanium concentration in the material was 0.8 mass.%. The adsorption activity was 120.3 mg/g [39].

The research was conducted within the framework of the project “Preclinical studies of products made from newly developed biomaterials” (Contract No. 48/1 dated August 27, 2019) as part of the national scientific and technical program titled “Development and clinical implementation of bone implants for various purposes made from advanced biomaterials for the restoration of bone tissue and function after combat-related injuries” (No. 0119U102083, NAS of Ukraine).

Animals and study design. The study was conducted on clinically healthy female California White rabbits, aged 3 months and weighing 2.5 kg, which were housed in the vivarium at Bila Tserkva National Agrarian University. The animals were kept in individual cages in a room with forced ventilation, combined lighting, and daily cleaning. They were fed a specialized, certified rabbit feed of the “Selevana” brand (Ukraine) at a rate of 200 g per rabbit per day, with free access to water.

To induce osteoporosis, a group of rabbits (n = 18) was formed, which were administered a 0.4% dexamethasone solution (4 mg/ml) (KRKA, Slovenia) intramuscularly at a dose of 1.2 mg/kg body weight once a day for 21 days [40-42]. Radiological confirmation of the osteoporotic process was carried out using the M. Singh [43] and H. K. Genant [44] indices, reflecting three-stage trabecular bone resorption in the proximal femur and a three-stage decrease in the height of the last lumbar vertebrae, leading to their concave bilateral deformation.

For modeling bone defects in rabbits with induced osteoporosis, the following anesthetic regimen was used: 2% acepromazine solution (Combistress, KELA, Belgium) intramuscularly (0.7 mg/kg), intravenous thiopental sodium solution (Tiopenat, Brovarpharma, Ukraine) at 6 mg/kg, and local infiltration anesthesia with a 0.5% lidocaine solution (3 mg/kg). During the postoperative period, for 72 hours, a nonsteroidal anti-inflammatory drug with analgesic activity, Melvet (meloxicam 0.2 mg per 1 kg of body weight), produced by Brovafarma (Ukraine), was administered.

The model defects were created in the cancellous bone on the lateral surface of the distal femoral metaphysis and in the cortical bone on the dorsolateral surface of the radius diaphysis, following the exposure of the periosteum with drills of 3 mm and 4.2 mm diameters, respectively, in compliance with aseptic and antiseptic rules. In the experimental group (n = 9), the defects were replaced with calcium phosphate ceramic granules, synthesized from hydroxyapatite and β-tricalcium phosphate, doped with germanium. In the control group (n = 9), bone defects were healed under a blood clot. Non-absorbable synthetic polypropylene monofilament sutures (OPUSMED, Ukraine) were used to close the soft-tissue wounds. The sutures were treated twice daily for 7 days with the antiseptic "Yoddicirin".

The study was approved by the Ethics Committee of Bila Tserkva National Agrarian University, protocol No. 1 dated January 23, 2019, and was conducted in accordance with the principles of the European Convention for the Protection of Vertebrate Animals used for Experimental and Other Scientific Purposes (Strasbourg, 1986), as well as the Ukrainian Law "On the Protection of Animals from Cruel Treatment" dated March 28, 2006, No. 27, Article 230, and the Ministry of Education and Science Order No. 416/20729 dated March 16, 2012, "On the Approval of the Procedure for Scientific Institutions to Conduct Animal Experiments".

Blood plasma sampling. Blood samples for biochemical analysis were collected from the external jugular vein before the bone defect modeling and on the 7th, 14th, 30th, and 60th days of reparative osteogenesis. Since reference values for the studied biochemical parameters are not available for rabbits, a group of clinically healthy rabbits (n = 27) was also formed.

Blood was collected from the rabbits in a volume of 2 ml for plasma and 2 ml for serum. To do this, the rabbits were fixed in a right lateral position, the blood collection site was shaved, and an antiseptic solution was applied. For plasma, blood was drawn from the external jugular vein into tubes with 3.8% sodium citrate and centrifuged for 30 min at 1300 g [45].

For serum, blood was collected into tubes without an anticoagulant. Before centrifugation, blood was allowed to clot for a specific period of time. Clotting time: 60 min at room temperature (20–25°C), away from sunlight and heating devices, after which it was centrifuged for 10 min at 350 g. Plasma and serum were stored at - 20°C.

The total activity of alkaline phosphatase (TALP) and its bone isoenzyme (BALP), as well as the activity level of tartrate-resistant acid phosphatase (TRAP) in blood serum, were determined using reagent kits (Granum Laboratory LLC, Kharkiv, Ukraine). The method principle involves the hydrolysis of alpha-naphthylphosphate to alpha-naphthol + phosphate, using tartaric acid as a specific agent for the bone isoenzyme of acid phosphatase.

The concentration of circulating immune complexes in blood serum was determined using reagents (Granum, Kharkiv, Ukraine) by the precipitation method using a polyethylene glycol-6000 solution at a concentration of 3.75% to detect large immune complexes (LIC) and 7% for small immune complexes.

The level of plasma protein C was determined using reagents (Granum, Kharkiv, Ukraine). Its value is expressed in the normalized ratio (NR), which reflects the ratio of the clotting time of the experimental plasma to the clotting time of normal plasma.

The content of nitric oxide was determined using reagents (Granum, Kharkiv, Ukraine) by the level of its metabolites in blood serum-nitrites. Metallic cadmium granules were used as a reducing agent, which were added to the serum samples after protein precipitation and incubated for 15 h. When nitrites in the serum interacted with the Griess reagent, a color complex was formed, which was then measured using a spectrophotometer (wavelength 540 nm).

Statistical analysis of the results was performed using Statistica 10 (StatSoft Inc., USA, 2011), with the data presented in the table as mean ± SD (x ± standard deviation). The significance of the difference between the numerical values of the control and experimental groups, as well as the values for clinically healthy rabbits before bone defect for-

mation, was determined using ANOVA. $P < 0.05$–0.001 was considered statistically significant, with the Bonferroni correction applied.

Results

In rabbits with induced osteoporosis, a moderate but statistically significant increase is observed in the blood serum activity of total alkaline phosphatase and its bone isoenzyme, as well as tartrate-resistant acid phosphatase (Table 1).

After bone defect substitution, the activity of total alkaline phosphatase (ALP) significantly increased from day 7, reaching a peak on day 14 – 79.82± 0.29 U/l. However, in the group without bone substitution, the ALP activity reached its peak only on day 30 – 73.68 ± 0.21 U/l, and was significantly lower than that observed in the experimental animals. Thus, the dynamics of ALP activity under conditions of bone substitution in osteoporosis are similar to the pattern of its changes during regeneration under a blood clot, but with a significantly earlier peak – by two weeks.

The difference in the dynamics of activity between the experimental and control groups was more pronounced when analyzing the widely recognized biochemical marker of osteogenesis – the bone isoenzyme of alkaline phosphatase (BAP). Specifically, after bone substitution, the BAP activity reached its peak value – 58±0.37 U/L – as early as day 14 and remained at a relatively high level up to day 30. This was 1.3 to 1.5 times higher compared to clinically healthy rabbits. In contrast, in the control animals, PAB activity reached its peak only on day 30 of reparative osteogenesis, and this value was significantly lower than that observed in the experimental group. Thus, the level of BAP activity following bone substitution with HTlGeG-700 under osteoporotic conditions reflects an enhancement of osteogenesis processes, most likely due to the osteoinductive properties of the germanium-doped calcium phosphate ceramic.

The degree of bone tissue resorption is reflected by the activity of tartrate-resistant acid phosphatase (TRACP) in blood serum, which showed a significant increase after osteoporosis induction and remained at that elevated level throughout 30 days of reparative osteogenesis. Its maximal value – 39.58 ± 0.52 U/l – was reached only by day 60. In contrast, in animals after bone substitution, TRACP activity peaked earlier – 41.53 ± 0.24 U/l – on day 14, and thereafter, from day 30, it progressively declined to levels typical of clinically healthy animals. Thus, based on the dynamics of phosphatase activity, bone substitution with bioactive germanium-doped ceramic under osteoporotic conditions is accompanied by a transient inflammatory osteoresorption phase, followed by an early and intensive osteogenic response of osteoblasts – a process that, when bone defects heal under a blood clot, is prolonged over time.

Changes in the serum level of low-molecular-weight circulating immune complexes (CIC) (Table 2) reflect complement-independent persistence

Table 1. Dynamics of phosphatase activity in blood serum during reparative osteogenesis in rabbits with osteoporosis

Time of study, days		ALP, U/l	BAP, U/l	TRACP, U/l
Clinically healthy, *n* = 27		63.36 ± 0.32	39.94 ± 0.41	28.36 ± 0.26
0	Before surgery, *n* = 18	70.21 ± 0.42	43.12 ± 0.29	33.73 ± 0.35
7	CPC-Ge	74.58 ± 0.31***	50.76 ± 0.52***	34.91 ± 0.32
	under blood clot, *n* = 9	70.32 ± 0.23	44.56 ± 0.31	33.80 ± 0.47
14	CPC-Ge	79.82 ± 0.29***	58.47 ± 0.37***	41.53 ± 0.24***
	under blood clot, *n* = 9	70.56 ± 0.47	44.37 ± 0.25	34.00 ± 0.36
30	CPC-Ge	75.69 ± 0.25***	53.46 ± 0.64***	37.83 ± 0.29***
	under blood clot, *n* = 6	73.68 ± 0.21	46.71 ± 0.53	4.13 ± 0.35
60	CPC-Ge	67.37 ± 0.43***	46.00 ± 0.18	31.71 ± 0.41***
	under blood clot, *n* = 3	71.53 ± 0.38	45.62 ± 0.44	39.58 ± 0.52

Values *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$, compared to the control group. Alkaline phenyl phosphatase (ALP), Bone alkaline phosphatase (BAP) is the bone-specific isoform of alkaline phosphatase, Tartrate-resistant acid phosphatase (TRACP), CPC-Ge – Calcium phosphate ceramics doped with germanium

of immune complexes in tissues. Throughout the study, their levels in the control group were 1.2-1.6 times higher ($P < 0.05$ – $P < 0.001$) than in the experimental group, where the values returned to those observed in clinically healthy animals (5.71 ± 0.36 conventional units) by day 30. Meanwhile, the level of complement-dependent high-molecular-weight CICs did not change after bone substitution, whereas in the control group it peaked between days 14 and 30, increasing 1.8-fold ($P < 0.001$).

Then, during reparative osteogenesis under osteoporotic conditions, both complement-dependent and complement-independent immune complex formation is enhanced, which likely contributes to a more intense production of various classes of mediators involved in the inflammatory-resorptive phase of reparative osteogenesis. In contrast, this process is considerably limited in the presence of bone substitution.

The anticoagulant potential of blood plasma, as indicated by protein C levels, did not change under induced osteoporosis. However, during reparative osteogenesis, it initially decreased and then increased. On day 7, protein C activity in the experimental group decreased only 1.2-fold ($P < 0.05$), whereas in the control group it decreased 2.6-fold ($P < 0.001$). In the bone substitution group, protein C activity showed no significant difference from the levels in clinically healthy rabbits by day 14, whereas in the control group, this normalization occurred only by day 30 of reparative osteogenesis.

It was found that under conditions of corticosteroid-induced osteoporosis, the serum level of nitric oxide (NO) moderately decreased. Subsequently, during reparative osteogenesis under a blood clot, the NO content gradually and moderately increased, peaking on day 60. In contrast, with bone substitution using CPC-Ge, NO levels peaked earlier – on days 14 and 30 – followed by normalization by day 60. Thus, bone substitution with germanium-doped ceramic under osteoporotic conditions promotes enhanced endothelial function and earlier, more intense neoangiogenesis.

Discussion

The key phases of the osteoreparative process include inflammation-driven resorption of damaged tissue, cell differentiation and proliferation, and the formation of new bone (identical to the undamaged area), followed by its remodeling under the influence of biomechanical factors. This process also involves the formation of an organic extracellular matrix and its subsequent mineralization [46, 47]. However, in cases of interfragmentary gaps or post-traumatic bone defects of critical size, full restoration of bone structure and function requires the recovery of its reparative potential through bone substitution with various implants. This is especially relevant for re-

Table 2. Dynamics of circulating immune complexes, protein C and NO levels in blood serum during reparative osteogenesis in rabbits with osteoporosis

Time of study, days		CIC, U		Protein C, NR	NO, μmol/l
		high-molecular-weight	low-molecular-weight		
Clinically healthy, *n* = 27		3.17 ± 0.14	5.73 ± 0.48	2.07 ± 0.16	28.93 ± 0.32
0	Before surgery, *n* = 18	3.18 ± 0.31	5.67 ± 0.39	2.08 ± 0.03	26.55 ± 0.42
7	CPC-Ge	3.34 ± 0.31*	6.28 ± 0.21*	1.67 ± 0.17***	26.58 ± 0.37
	under blood clot, *n* = 9	4.31 ± 0.23	7.45 ± 0.36	0.79 ± 0.03	26.56 ± 0.29
14	CPC-Ge	3.59 ± 0.24***	7.95 ± 0.26**	1.89 ± 0.14***	38.74 ± 0.62***
	under blood clot, *n* = 9	5.62 ± 0.18	11.83 ± 0.47	0.80 ± 0.12	29.41 ± 0.48
30	CPC-Ge	3.39 ± 0.16***	5.98 ± 0.19***	2.17 ± 0.20	38.00 ± 0.36***
	under blood clot, *n* = 6	5.60 ± 0.37	9.46 ± 0.41	1.69 ± 0.18	30.78 ± 0.56
60	CPC-Ge	3.19 ± 0.13**	5.71 ± 0.36**	2.15 ± 0.06	28.75 ± 0.41***
	under blood clot, *n* = 3	4.46 ± 0.38	7.13 ± 0.29	2.07 ± 0.03	33.32 ± 0.25

Values *$P < 0.05$; **$P < 0.01$; ***$P < 0.001$, compared to the control group; CIC – circulating immune complexes, NO – nitric oxide, CPC-Ge – Calcium phosphate ceramics doped with germanium, NR – normalized relation

ducing the incidence of complications during the consolidation of fractures with bone defects, such as high-energy fractures, pathological fractures on the background of osteoporosis, or bone neoplasms [48-50]. One such material is calcium phosphate ceramic, which consists of two biocompatible phases: hydroxyapatite and β-tricalcium phosphate [51, 52].

Germanium, as a modifier of hydroxyapatite ceramics for the purpose of osteoinduction, possesses a broad spectrum of biological activity. Its organic and complex compounds exhibit antitumor, analgesic, anti-inflammatory, antioxidant, immunomodulatory, fungicidal, antiviral, and antimicrobial properties [61, 64]. Germanium may also hold promise for osteoinductive applications due to its stimulatory effect on osteoblast proliferation and its inhibitory effect on osteoclast activity – a balance that is disrupted in secondary osteoporosis and fractures associated with it.

The present study established that the use of germanium-doped ceramic promotes dynamic course of reparative osteogenesis in animals with glucocorticoid-induced osteoporosis.

This is particularly evident in the changes observed in bone metabolism markers. Specifically, it was found that bone substitution with bioactive germanium-doped ceramic is accompanied by a transient phase of inflammatory osteoresorption, followed by an early and intense osteogenic response of osteoblasts.

Corresponding with the rapid increase in nitric oxide levels, substitution with doped ceramic enhances endothelial function and promotes earlier and more robust neoangiogenesis under osteoporotic conditions.

The immune response under bone substitution was primarily mediated through the formation of low-molecular-weight circulating immune complexes (CIC). It was revealed that reparative osteogenesis in osteoporosis is characterized predominantly by complement-independent immune complex formation, which contributes to an intensified production of various classes of mediators involved in the inflammatory-resorptive phase. In contrast, this phase is significantly reduced in the presence of germanium-doped ceramic.

Although bone trauma is known [32, 67] to induce a hypercoagulable state that may become persistent – compromising neoangiogenesis and oxygenation of the regenerating bone – substitution with germanium-doped ceramic effectively prevented the depletion of the natural anticoagulant protein C.

Conclusions. Bone substitution with germanium-doped calcium phosphate ceramic in bone defects of both trabecular and cortical bones, compared to healing under a blood clot, not only improves tissue regeneration but also leads to faster rehabilitation, reduced inflammatory and immune responses, and prevents the development of thrombophilia in rabbits with osteoporosis. Doping the ceramic with germanium for bone substitution is a promising approach, and its application could significantly improve the outcomes of such surgeries.

Conflct of interest. Authors have completed the Unified Conflicts of Interest form at http://ukrbiochemjournal.org/wp-content/uploads/2018/12/coi_disclosure.pdf and declare no conflict of interest.

Funding. This study was carried out within the framework of the project state theme “Preclinical studies of products from developed biomaterials”, funded by the National Academy of Sciences of Ukraine (state registration number 0119U102083).

МАРКЕРИ РЕПАРАТИВНОГО ОСТЕОГЕНЕЗУ ПРИ ЗАМІЩЕННІ КІСТКОВИХ ДЕФЕКТІВ КЕРАМІКОЮ, ЛЕГОВАНОЮ ГЕРМАНІЄМ, ЗА ЕКСПЕРИМЕНТАЛЬНОГО ОСТЕОПОРОЗУ

Т. Тодосюк[1]✉, В. Чемеровський[1], А. Рубленко[2], Н. Ульянчич[3], В. Коломієць[3], С. Фірстов[3]

[1]Білоцерківський національний аграрний університет, Біла Церква, Україна;
[2]Вінницький державний педагогічний університет імені Михайла Коцюбинського, Вінниця, Україна;
[3]Інститут проблем матеріалознавства ім. І. Францевича НАН України, Київ;
✉e-mail: tatyana.todosyuk@gmail.com

Остеопороз, як системне захворювання скелета, характеризується втратою кісткової маси, зниженням мінеральної щільності кісток та змінами мікроархітектури. У разі травматичних переломів та дефектів критичного розміру остеопороз може як призвести до спонтанних переломів, так і погіршити регенерацію, що потребує використання кісткозамінних матеріалів. Перспективними кісткозамінними матеріалами вважаються керамічні імплантати, леговані германієм з метою надання їм остеоіндуктивних

властивостей. У цьому дослідженні ми мали на меті оцінити біохімічні маркери репаративного остеогенезу при заміщенні кісткових дефектів керамікою, легованою германієм, у кролів з експериментальним остеопорозом. Дослідження проводилося на кролях породи каліфорнійський білий з остеопорозом, індукованим введенням 0,4% розчину дексаметазону. Модельні дефекти створено в трабекулярній та кортикальній кістковій тканині після оголення окістя з використанням свердел діаметром 3 та 4,2 мм відповідно, з дотриманням вимог анестезіологічного забезпечення та асептики і антисептики. Для заміщення дефектів використовували кальцій-фосфатні керамічні гранули (CPC-Ge) розміром 700 мкм, синтезовані з гідроксиапатиту та β-трикальційфосфату та леговані 0,8 мас.% германію. У контрольній групі тварин (n = 9) дефекти кісток загоювалися під кров'яним згустком. В експериментальній групі (n = 9) дефекти заміщували гранулами CPC-Ge. Зразки крові для біохімічних досліджень відбирали перед моделюванням дефекту кістки та на 7-й, 14-й, 30-й та 60-й дні репаративного остеогенезу. Визначали активність тартрат-резистентної кислої фосфатази, лужної фосфатази та її кісткової ізоформи, а також циркулюючих імунних комплексів, протеїну С та NO у сироватці крові. Встановлено, що заміщення дефектів як трабекулярної, так і кортикальної кісток за допомогою CPC-Ge, порівняно з загоєнням під кров'яним згустком, супроводжується зменшенням запальної та імунної відповіді, запобігає виснаженню протеїну С та сприяє більш динамічному перебігу репаративного остеогенезу у тварин з остеопорозом, індукованим глюкокортикоїдами.

Ключові слова: германій, кальцій-фосфатні керамічні гранули, остеозаміщення, остеопороз, репаративний остеогенез, оксид азоту, фосфатази, протеїн С.

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