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РОЛЬ ХРОНІЧНОГО ГАСТРИТУ СЕРЕД ПЕРЕДРАКОВИХ ЗАХВОРЮВАНЬ ШЛУНКА

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Метою дослідження є статистичний аналіз стану слизової шлунка, враженого Helicobacter pylori, у молодих людей, які займаються спортом. Проведені дослідження хронічного гастриту типу В у студентів-добровольців, які займаються спортом. У 92% хронічні гастрити були Helicobacter pylori-асоційовані. Між ступенем обсіменіння слизової оболонки Helicobacter pylori і ступенем лейкоцитарної інфільтрації слизової оболонки коефіцієнт кореляції Пірсона r_{xy} =0,935, тіснота зв'язку –дуже сильна, коефіцієнт детермінації $D=r_{xy}^2$–0.874, критичне значення коефіцієнта кореляції з вірогідністю 0,95–0,2732, критичне значення коефіцієнта кореляції з вірогідністю 0,99–0,3511, порівнюючи коефіцієнт кореляції r_{xy} з критичним значенням r_{cr} для значущості 0.95–r_{xy} >r_{cr}, порівнюючи коефіцієнт кореляции r_{xy} з критичним значенням r_{cr} для значущості 0,99 – r_{xy}>r_{cr}. до коефіцієнта коваріації становить 521,641, це дозволяє зробити висновок про статистично значущу залежність з імовірністю 0,99.

Таким чином, хронічний атрофічний гастрит, асоційований з Helicobacter pylori, є поширеним захворюванням молодих людей, що займаються спортом, і є центральним серед передракових захворювань шлунку.

Ключові слова: Helicobacter pylori, гастрит, лейкоцитарна інфільтрація, слизова оболонка шлунка.

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РОЛЬ ХРОНИЧЕСКОГО ГАСТРИТА СРЕДИ ПРЕДРАКОВЫХ ЗАБОЛЕВАНИЙ ЖЕЛУДКА

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Целью работы является статистический анализ состояния слизистой оболочки желудка пораженной Helicobacter pylori у молодых людей, которые занимаются спортом. Проведены исследования хронического гастрита типа В у студентов-добровольцев, которые занимаются спортом. В 92% хронические гастриты были Helicobacter pylori-асоциированные. Между степенью обсеменения слизистой оболонки Helicobacter pylori і степенью лейкоцитарной инфильтрации слизистой оболочки коэфициент корреляции Пирсона

r_{xy}=0,935, плотность связи–очень сильная, коэфициент детерминации $D=r_{xy}^2$–0.874, критическое значение коефициента корреляции с вероятностью 0,95–0,2732, критическое значение коефициента корреляции с вероятностью 0,99–0,3511, сравнивая коефициент кореляции r_{xy} с критическим значением r_{cr} для значимости 0.95–r_{xy} >r_{cr}, сравнивая коефициент кореляции r_{xy} с критическим значением r_{cr} для значимости 0,99 – r_{xy}>r_{cr}. к коефициенту ковариации составляет 521,641, это позволяет сделать вывод о существовании статистически значимой зависимости с вероятностью 0,99.

Таким образом хронический атрофический Helicobacter pylori–асоциированный гастрит является распространённым заболеванием людей в молодом возрасте, которые занимаются спортом, и занимает центральное место среди предопухолевых заболеваний желудка.

Ключевые слова: Helicobacter pylori, гастрит, лейкоцитарная инфильтрация, слизистая оболочка желудка.

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STUDY ON THE EFFECT OF THE VITAMIN AND MINERAL COMPLEX CONTAINING ZINC L-ASPARTATE ON THE PERIODONTAL CONDITION OF RATS IN THE PRESENCE OF PERIODONTITIS MODELING

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The purpose of the study was to study the effect of the vitamin and mineral complex containing zinc L-aspartate on the state of the periodontal tissues of rats under conditions of modeling periodontitis using exogenous collagenase. The vitamin and mineral complex containing zinc L-aspartate had a positive effect, to a greater extent, on the periodontal bone tissue. The complex has shown periodontal protection, anti-inflammatory, antioxidant properties.

Key words: zinc L-aspartate, periodontitis modeling, collagenase, collagen, glycosaminoglycans, gums, periodontal bone tissue, rats.

The study is a fragment of the research project "The effect of hypoxia on the processes of collagen formation and mineralization in models of dental pathology and correction of these disorders", state registration No. 0118U006963.

Extracellular matrix (ECM) of the connective tissue is defined by a complex system formed by multicellular structural macromolecules: proteoglycans, collagens, and elastins, which maintain its structural integrity. ECM consists of three essential components – a gelling medium, collagen and elastin fibers, and provides a rapid diffusion of substances and "construction" materials between blood and cells.

Changes in the state of the extracellular matrix (ECM) of periodontal tissues during periodontitis are carried out using matrix metalloproteinase (MMP_s) or collagenases, the main differences of which from other endopeptidases are associated with their ability to destroy the ECM structures of connective tissue, as well as with their dependence on metal ions. The balance between degradation and synthesis of ECM determines the state of periodontal soft tissues and bone tissues during periodontitis.

Zinc is essential in the body for cell growth, protein production, and wound healing. It helps to stabilize blood sugar levels, has a positive effect on the body's immune system, and exhibits antioxidant properties. Zinc is a part of more than 80 enzymes: carbonic anhydrase, RNA and DNA polymerases, carboxypeptidases.

Zinc is one of the most important trace elements that is part of many enzyme systems. It regulates the basic metabolic processes, participates in the metabolism of carbohydrates. Zinc is necessary for the functioning of more than 200 metalloenzymes (carbonic anhydrase, carboxypeptidase A, alkaline phosphatase, RNA polymerase, etc.), as well as for the normal structure of nucleic acids, proteins and cell membranes [2]. Zinc promotes cell growth and development, the normal functioning of the immune system and the provision of an immune response [11]. Zinc deficiency causes difficulties in concentration and memory, decreased cellular and humoral immunity, poor wound healing [4].

Zinc is a potent inhibitor of MMP_s and helps to improve the absorption of B vitamins. This element is important for the normal development of bone tissue. The accumulation of metals in connective tissue can affect the formation and absorption of extracellular components of ECM. It was found that Zn^{2+} ions have an inhibitory effect on MMP_s-2 and MMP_s-9. Studies of the effect of different ions (zinc, tin, copper, mercury) on gum gelatinase (MMP_s-2 and MMP_s-9) have shown that $ZnSO_4$ is its most potent inhibitor, while $CuSO_4$, $HgSO_4$, and others are less effective [10].

All of the above predetermined the study of the correction of damage to the extracellular matrix of the periodontium as a result of the periodontitis modeling with a complex of substances necessary for the normal functioning of the connective tissue.

The purpose of the study was to study the effect of the vitamin and mineral complex containing zinc L-aspartate on the state of the periodontal tissues of rats under conditions of modeling periodontitis using exogenous collagenase.

Materials and methods. The experiment was carried out on 21 white female breeding rats of the Wistar line. Animals of the 1st group (6 animal units) were intact. In Groups 2 and 3, periodontitis was modeled by injecting exogenous collagenase solution under the gums (from Clostridium histolyticum lyophilisat 2000 E/mg, Merk, Darmstadt (Germany) at a dose of 1 mg/ml in four areas of the jaws three times during the experiment. In the 3rd group, 7 rats were administered per os with the vitamin and mineral complex "Active Zinc" for 1 tablet/0.2 ml 5 times a week in the morning hours for 55 days, against the background of periodontitis modeling.

In addition to zinc L-aspartate, complex "Active Zinc" (TOV "Elit-PHARM", Dnipro, Ukraine) contains components that contribute to the better absorption of the zinc ion. Zinc is absorbed with the help of phosphorus, calcium, manganese, vitamins A, C, D_3.

1 tablet (0.250 g) of the complex contains zinc L-aspartate (active zinc – 12.6 mg), manganese aspartate (Mn^{2+} – 0.05 mg), calcium hydrogen phosphate (phosphorus – 30 mg), vitamin A – 1666 IU, vit. C – 10 mg, vit. D_3 – 2.5 mcg. Excipients: lactose, sorbitol, starch, calcium stearate.

At the end of the experiment, the rats were sacrificed by total exsanguination from the vessels of the heart under anesthesia with thiopental (40 mg/kg). All experiments were carried out in accordance with the European Convention for the Protection of Vertebrate Animals used for Experimental or other Scientific Purposes (Strasbourg, 1986). After separating the gums, the jaws were dissected out and the resorption of the alveolar bone was assessed morphometrically. The objects of biochemical studies were the gums and the alveolar bone of rats.

The ECM state of the connective tissue was determined by the levels of hydroxyproline (state of collagen) and glycosaminoglycans (GAGs). The lipid peroxidation level (LPL) was assessed by the content of malondialdehyde (MDA). The activity of antioxidant enzymes catalase [1], and glutathione peroxidase (GPx) was determined. To assess the state of rats' tissues, biochemical parameters were determined by unified methods using commercial reagent kits: alkaline phosphatase (ALP) activity, calcium (Ca^{2+}), phosphorus, Mg^{2+}, zinc, sialic acids. All kits were manufactured by DAC-SpectroMed, Moldova.

The experimental results were processed by conventional statistical methods with the determination of t-criteria for the reliability of differences according to Student's t-test.

Results of the study and their discussion. The study of the complex containing zinc L-aspartate effect on the periodontal tissues of rats was carried out under conditions of periodontitis modeling by subgingival administration of exogenous collagenase at a dose of 1 mg/ml. Oral administration of the

complex against the background of reproduced experimental periodontitis had a positive effect on the state of collagen in rats' periodontal tissues. Thus, under the influence of the complex, the free hydroxyproline level increased by 2.7 times (p = 0.004), the total level increased by 58% (p = 0.003) compared to the group "model of periodontitis", while the bound hydroxyproline level did not change significantly (table 1).

Table 1

Effect of the complex containing zinc L-aspartate on the state of the extracellular matrix (ECM) of rats' periodontium during the periodontitis modeling (M±m; p)

Studied indices	Groups of animals	
	periodontitis model (M)	M+"Active Zn"
	Gum	
Content: GAGs (mg/g)	3.87±0.59	3.55±0.32
hydroxyproline (μmol/g) free	2.12±0.41	5.65±0.87 p=0.004
bound	4.59±0.44	4.94±2.05
total	6.71±0.84	10.6±0.50 p=0.003
	alveolar bone	
Content: GAGs (mg/g)	2.12±0.074	2.56±0.055 p=0.001
hydroxyproline (μmol/g) free	1.41±0.21	2.06±0.00 p=0.011
bound	1.06±0.21	2.47±0.00 p<0.001
total	2.47±0.00	4.77±0.61 p=0.005

Note: In tables 1-3, the significance index p was calculated in comparison with the intact group.

Hydroxyproline content in the periodontal bone tissue also increased significantly: free – by 1.5 times (p = 0.011); bound – by 2.3 times (p<0.001); total – by 93% (p = 0.005). GAGs level in the alveolar bone increased by 21% (p = 0.001; table 1).

Under the influence of a complex containing zinc L-aspartate, the concentration of sialic acids in the blood serum decreased by 22% (p = 0.004; table 2), which indicates both its anti-inflammatory properties in the organism, and the partial restoration of glycoproteins, which indicates an improvement in the ECM state of the connective tissue.

Under the conditions of modeling periodontitis by subgingival administration of collagenases, the alveolar bone resorption in rats increased: in the mandible by 16% (p <0.001) – 41.2±0.88% compared with the intact group: 35.5±0.90%. In the maxilla, the resorption increase was 20% (p=0.016): 31.3±1.43% versus 26.1±1.20% in the intact group (100%).

Table 2

Effect of a complex containing zinc L-aspartate on the content of sialic acids and metals in the periodontium connective tissue of rats in the modeling of periodontitis (M±m; p)

Studied indices	Groups of animals	
	periodontitis model (M)	M+"Active Zn"
	blood serum	
Content: sialic acids (mmol/ml)	2.40±0.059	1.88±0.096 p=0.004
	gum	
Content: Zn^{2+} (μmol/g)	5.96±0.16	7.56±0.074 p<0.001
	alveolar bone	
Content: Mg^{2+} (μmol/g)	0.098±0.0095	0.16±0.00 p<0.001
Zn^{2+} (μmol/g)	2.09±0.28	2.44±0.39

Vitamin and mineral complex "Active Zinc" against the background of periodontitis model significantly reduced the resorption of periodontal bone tissue: by 14% in the mandible (100% in the control group; p = 0.01) and by 6% in the maxilla (p> 0.05; Table 3) compared with data from control groups. Thus, the periodontal protective properties of the complex containing zinc L-aspartate were revealed.

The content of Zn^{2+} increased in the soft tissues of the periodontium by 21% (p <0.001); in the alveolar bone by 17% (p> 0.05; table 2). At the same time, the content of Mg^{2+} ions in the periodontal bone tissue significantly increased (by 63%; p <0.001), which indicates an improvement in the state of the ECM in the alveolar bone. It is known that a lack of magnesium leads to a slowdown in protein synthesis. In addition, there was deterioration of the mechanical properties of the gel, which forms the ECM basic substance as a result of the hyaluronidase activation [3].

The results are quite justified in connection with the improvement of the mineral metabolism in this study object – an increase in alkaline phosphatase activity by 56% (p = 0.02) as a result of osteoblast activation, since alkaline phosphatase is their marker enzyme (table 3). The levels of calcium and phosphorus in these conditions increased insignificantly (p> 0.05; table 3).

Table 3

Effect of the complex containing zinc L-aspartate on the state of bone mineral metabolism in rats' periodontium during the periodontitis modeling (M±m; p)

Studied indices	Groups of animals	
	periodontitis model (M)	M+"Active Zn"
	alveolar bone	
Activity: ALP (nmol/s.g.)	0.27±0.035	0.42±0.035 p=0.002
Content: Ca^{2+} (mmol/g)	0.024±0.0017	0.026±0.0046
Phosphorus (mmol/g)	0.023±0.0035	0.027±0.0052
Periodontal bone resorption indices (%) mandible	41.2±0.88	35.3±1.66 p=0.01
maxilla	31.3±1.43	29.5±1.31

Under the influence of the complex, the level of LPL processes decreased – the MDA content in the gums decreased by 14% (p = 0.02; table 4). At the same time, the activity of the antioxidant enzyme catalase in this study object increased by 28% (p = 0.05). Insufficient functioning of the antioxidant enzyme glutathione peroxidase in the gums was evidenced by a 1.9-fold decrease in its activity (p = 0.003) compared with the "Periodontitis model" group (table 4). In the periodontal bone tissue, the complex, more significantly than in the gums, reduced the level of peroxide products: the MDA content decreased by 2.2 times (p = 0.001), which may indicate the antioxidant properties of the complex containing zinc L-aspartate.

Catalase activity in the periodontal bone tissue under the complex influence changed insignificantly, while the activity of glutathione peroxidase increased by 6% (p = 0.06) compared with the control group (table 4).

Table 4

Effect of a complex containing zinc L-aspartate on the MDA content and the activity of antioxidant enzymes in the periodontium connective tissue of rats in the modeling of periodontitis (M±m; p)

Studied indices	Groups of animals	
	periodontitis model (M)	M+"Active Zn"
	gum	
Content: MDA (nmol/g)	50.3±1.02	43.1±2.51 p=0.02
Activity: catalase (mkat/g)	13.9±1.90	17.8±0.84 p=0.05
GPx (μmol/s.g.)	148±6.38	77.5±7.30 p=0.003
	alveolar bone	
Content: MDA (nmol/g)	0.80±0.083	0.37±0.012 p<0.001
Activity: catalase (mkat/g)	10.4±0.84	10.7±1.61
GPx (μmol/s.g.)	68.9±0.97	73.0±1.68 p=0.06

Studies have shown that the vitamin and mineral complex containing zinc L-aspartate, administered orally to rats against the background of reproduced experimental periodontitis, had a positive effect on the periodontal tissues of rats. First of all, under its influence, the state of collagen, recorded by the level of hydroxyproline (total, free, bound) was improved, which levels significantly increased compared to the control group (periodontitis model). In addition, in hard periodontal tissues, the complex restored the state of the gel, which forms the ECM basis of the connective tissue.

The zinc-containing complex significantly, by 1.6 times, increased the activity of alkaline phosphatase, an enzyme localized in bone tissue on the outer surface of osteoblast membranes. In this regard, the complex against the background of the periodontitis model significantly reduced the resorptive processes in the alveolar bone process of rats.

According to the literature, zinc is one of the components of enzymatic systems, which activity affects the growth, development and physiological state of the organism. It is referred to as trace elements that promote the synthesis of collagen, glycosaminoglycans. It is directly involved in the synthesis of bone matrix [13]. According to a number of scientists [9, 10, 11, 14], zinc increases bone formation, accelerates its mineralization, reduces bone resorption and stimulates the activity of alkaline phosphatase in osteoblast cell culture. Alkaline phosphatase is one of the most representative proteins of osteoblast differentiation as bone markers. As a result of these studies, the authors demonstrated the anabolic role of zinc in the formation of bone tissue under the action of osteoblasts [12, 14].

Zinc stimulates osteoblast proliferation and differentiation, as well as protein synthesis in osteoblasts. The importance of the zinc effect on the bone tissue metabolism is evidenced by information about its deficiency, in which the amount of bone mass decreases [8, 9]. Zinc deficiency contributes to the stress of DNA synthesis and protein metabolism, which leads to disruption of the organic matrix metabolism [1, 11].

Severe metabolic disorders of trace elements, including zinc, occur already in the early stages of the pathological process. Thus, the zinc content in the blood plasma of rats was reduced during the gingivitis and periodontitis reproduction, and more pronounced changes were observed in the group of rats with gingivitis [5].

The molecular mechanisms of zinc deficiency are based on oxidative stress in cells and tissues, as well as an increase in the synthesis of proinflammatory cytokines [7]. Zinc is involved in redox processes. Zinc cations stabilize the permeability of cell membranes and effect as a protector of free radical reactions.

The studied complex, containing zinc L-aspartate showed anti-inflammatory effect at the body level. It reduced the level of sialic acids in the blood serum, since it is known that an increase in the sialic acid content indicates an increase in inflammation in the tissues. It is known that sialic acids, derivatives of neuraminic acid, are formed under the action of neuraminidase during the breakdown of ECM glycoproteins. In our studies, the complex containing zinc L-aspartate showed antioxidant properties. It reduced the level of peroxide processes in the gums and alveolar bone. Under the complex influence, the Zn^{2+} content in the gums of experimental animals increased by 21%, in the bone tissue of the periodontium – by 17%.

According to the data, the optimal amount of zinc ions in the organism is an important factor that ensures the regenerative function of the connective tissue. During wound healing, zinc provides a stabilizing effect on the cytoplasmic membranes, preventing the hydrolytic enzyme release, such as cathepsin D and collagenase, which control the rate of damaged tissue degradation [6].

Thus, considering the positive results of experimental studies, the zinc content in the complex "Active Zinc" was optimal. The complex can be recommended as a means that promote bone tissue formation, has a direct effect on reducing alveolar bone resorption, and also replenishes the deficiency of the osteotropic trace element zinc, which is involved in vital metabolic processes of the organism.

Conclusions

1. Vitamin and mineral complex containing zinc L-aspartate, administered orally against the background of reproduced experimental periodontitis, increased by 58% the level of total hydroxyproline in the gums; in the alveolar bone – by 93%; the glycosaminoglycans content in the periodontal bone tissue increased by 21%.

2. The complex significantly (by 14%) reduced the alveolar bone resorption, which indicates its periodontal protective properties. It significantly improved mineral metabolism – increased the alkaline phosphatase activity in the periodontal bone by 56%.

3. Under the complex influence, the level of LPL processes in the gums and more significantly in the bone tissue of the periodontium decreased significantly; on the body level, the complex containing zinc L-aspartate reduced the concentration of sialic acids in the blood serum, which indicates its anti-inflammatory properties.

4. Vitamin and mineral complexes containing zinc L-aspartate can be recommended for improving the metabolism of periodontal connective tissue and bone mineral metabolism in periodontitis.

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Реферати

ВИВЧЕННЯ ВПЛИВУ ВІТАМІННО-МІНЕРАЛЬНОГО КОМПЛЕКСУ, ЩО МІСТИТЬ ЦИНК L-АСПАРАГІНАТ, НА СТАН ПАРОДОНТА ЩУРІВ В УМОВАХ МОДЕЛЮВАННЯ ПАРОДОНТИТУ

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Метою дослідження було вивчення впливу вітамінно-мінерального комплексу, що містить цинк L-аспарагінат, на стан тканин пародонта щурів в умовах моделювання пародонтиту за допомогою екзогенної колагенази. Вітамінно-мінеральний комплекс, що містить цинк L-аспарагінат, мало позитивний вплив більшою мірою на кісткову тканину пародонту. Комплекс проявив пародонтопротекторні, протизапальні, антиоксидантні властивості.

Ключові слова: цинк L-аспарагінат, моделювання пародонтиту, колагеназа, колаген, глікозаміноглікани, ясна, кісткова тканина пародонту, щури.

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ИЗУЧЕНИЕ ВЛИЯНИЯ ВИТАМИННО-МИНЕРАЛЬНОГО КОМПЛЕКСА, СОДЕРЖАЩЕГО ЦИНК L-АСПАРАГИНАТ, НА СОСТОЯНИЕ ПАРОДОНТА КРЫС В УСЛОВИЯХ МОДЕЛИРОВАНИЯ ПАРОДОНТИТА

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Целью исследования явилось изучение влияния витаминно-минерального комплекса, содержащего цинк L-аспарагинат, на состояние тканей пародонта крыс в условиях моделирования пародонтита с помощью экзогенной коллагеназы. Витаминно-минеральный комплекс, содержащий цинк L-аспарагинат, оказал положительное влияние в большей степени на костную ткань пародонта. Комплекс проявил пародонтопротекторные, противовоспалительные, антиоксидантные свойства.

Ключевые слова: цинк L-аспарагинат, моделирование пародонтита, коллагеназа, коллаген, гликозаминогликаны, десна, костная ткань пародонта, крысы.

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EFFECT OF HORMONE-ACTIVE METABOLITES OF CHOLECALCIFEROL ON THE STATE OF THE ORAL CAVITY TISSUES IN RATS UNDER THE CONDITIONS OF ESTROGEN DEFICIENCY AND TRAUMATIC STRESS

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The purpose of the study was to comparatively establish the effect of active metabolites of vitamin D_3 on the periodontal condition of rats under conditions of experimental estrogen deficiency and traumatic stress. The experiment was carried out on 31 female breeding Wistar rats. 1st group was intact one (8 animals). The rats of the 2nd - 4th groups underwent ovariectomy and a f□m□r□l fr□□tur□. Gr□u□ 2 w□s □□ntr□l □n□ (8 r□ts); r□ts □f th□ 3rd □nd 4th gr□u□s r□□□v□d □r □s □r□□r□t□□ns □g□nst th□ background of the pathogenic effect: Group 3 (8 rats) – 1-α-hydroxycholecalciferol at a dose of 0.1 μg □r d□ □□r r□t; □n th□Gr□u□ 4 (7 rats) – 24,25-hydroxycholecalciferol at a dose of 1.25 μg per day/rat. At the time of sacrifice, the animals were 15 months old. Under the influence of risk factors for periodontitis, protective properties of hormone-active metabolites of vitamin D_3 were observed. They were expressed in inhibition of lipid peroxidation processes in the oral mucosa of rats, as well as periodontal protective effects when using 24,25-hydroxycholecalciferol.

Key words: vitamin D_3 metabolites, estrogen deficiency, traumatic stress, periodontal protection properties, antioxidant effect, rats.

The study is a fragment of the research project "The effect of hypoxia on the processes of collagen formation and mineralization in models of dental pathology and correction of these disorders", state registration No. 0118U006963.

Vitamin D_3 or cholecalciferol, which realizes its action in the organism through active metabolites – $25OHD_3$, $1,25(OH)_2D_3$ and $24,25(OH)_2D_3$, is directly involved in the bone tissue metabolism. $1,25(OH)_2D_3$ or calcitriol, is the most biologically active metabolite of vitamin D_3. With a deficiency of calcium and phosphorus, the metabolism of $25OHD_3$ follows the formation of 1,25 $(OH)_2D_3$, which is catalyzed by the enzyme 1-α-hydroxylase, which is present in the mitochondria of renal tubular epithelial cells. With an increased or normal concentration of calcium and phosphorus in the blood serum, an alternative metabolite, $25OHD_3$ – $24,25(OH)_2D_3$, is formed with 24-hydroxylase [10]. The fundamental