Available online **www.ijpras.com**

International Journal of Pharmaceutical Research & Allied Sciences, 2021, 10(3):8-14
https://doi.org/10.51847/fNtx5NuBtH

Original Article

ISSN : 2277-3657
CODEN(USA) : IJPRPM

Dietary Supplement for Better Metabolism in Fracture Healing: Study of Functional Properties and Efficacy

Boisjoni Tokhiriyon[1]*, Valeriy Poznyakovsky[1], Ekaterina Pastushkova[1], Chugunova Olga[1], Valentina Lapina[1]

[1]Institute of Commerce, Food Technology and Service, Ural State University of Economics, Ekaterinburg, Russia.

***Email:** tohiriyoni@gmail.com*

ABSTRACT

A new vitamin and mineral complex (a dietary supplement) has been developed. The active ingredients include magnesium oxide, vitamin C, zinc oxide, chromium picolinate, manganese sulfate, boron, hydroxyapatite calcium, silicon oxide, cholecalciferol, and chondroitin sulfate. The pharmaceutical formulation was carefully calculated taking into account biochemical properties of all the ingredients. The clinical trial was performed to evaluate the functional properties and efficacy of the supplement. The participants of the trial included patients with long bone fractures aged 3-7 (one capsule per day) and aged 8-14 (two capsules per day).
During the clinical trial various parameters were assessed. The levels of phosphorus, ionized calcium and alkaline phosphatase were tested and X-rays and laser Doppler flowmetry were employed. The dietary supplement helped to lessen the intensity of pain and swelling, therefore, the use of analgesics was limited and the treatment period was reduced. By the end of the treatment, biochemical parameters were within reference ranges. The study findings indicate the positive impact of mineral complex (the dietary supplement) and the new vitamin on bone tissue regeneration and the recovery processes as well as prevention of tropic changes in soft tissue and post-traumatic edema development. The recommendations on the safe use of the dietary supplement were developed. The dietary supplement is manufactured by the scientific research-to-production facilities of Art-Life scientific production association in the city of Tomsk. The manufacturing process is carried out in full compliance with ISO 9001, 22000, GMP to guarantee the product quality and competitiveness.

Key words: *Dietary supplement, Clinical trial, Functional properties, Fracture healing*

INTRODUCTION

Recent studies highlight the growing importance of nutrition in preventing and treating many non-communicable diseases. One of the common consequences of accidents is damage to the musculoskeletal system; it accounts for 65% of all injuries and often becomes the cause of disability [1-4]. To develop new therapeutic concepts, it is necessary to consider pathophysiologic alterations, changes in the microvasculature and the nature of bone regeneration [4-7].

Although paediatric bone healing is comparable to adult bone healing, more attention should be paid when choosing traditional and complementary treatment and considering the recovery process as there are concerns of the growing number of children that experience impaired bone healing. This is generally explained by the increased morbidity, the impact of the environmental factors, and the overall decrease in nonspecific resistance. Many physical, biological and chemical factors possess the osteoinductive activity and contribute to bone formation; however, 10–13% of all patients develop contractures, ankylosis and other complications.

It should also be noted that the traditional treatment that most patients receive in hospitals with the physical activity restricted is often the cause of respiratory inflammations, trophic changes in soft tissue and the slow remodelling process. Additionally, while lengthy restrictions of physical activity often lessen joint mobility, with the recovery process impeded, in order to manage severe pain in the acute period of injury, relevant prescription drugs are usually administered.

Considering all the shortcomings of the traditional treatment process, there is an obvious need to search for new approaches for the enhancement of the bone healing techniques. Recent studies emphasize the significant impact that dietary supplements can make on the bone regeneration process [7-16].

MATERIALS AND METHODS

To evaluate the dietary supplement efficacy, the clinical trial was performed in Children City Hospital No 4 of the city of Tomsk. A total of thirty-five children (21 boys and 14 girls) with long bone fractures participated in the clinical trial. The participants represented two age groups: aged 3-7 and 8-14.

For the trial, nineteen children were assigned into the control group and received traditional treatment, and sixteen children that comprised the experimental group were prescribed cryotherapy and Calcimax Recovery dietary supplement.

Diet therapy was aimed at treating rheological abnormalities, improving microcirculation, and activating bone regeneration. The regimen of one capsule per day taken with food was prescribed for children aged 3-7 and two capsules per day for children aged 8-14.

The clinical trial was supervised by G.V. Slizovskii, Doctor of Medical Sciences, professor of Paediatric Surgery of Siberian State Medical University.

During the trial, blood rheology and microcirculation parameters were assessed using both instrumental and laboratory methods. Blood tests were performed on the empty stomach on the day of the hospital admission, the following day, and then on the fourteenth and thirtieth days of the patients' hospital stay. Blood tests included the serum phosphate, ionized calcium, and alkaline phosphatase (ALP) tests.

X-rays were used to assess bone fractures and the healing process. The first X-rays were performed on the day of the hospital admission. The X-rays were taken at two different angles, the front and the side view (the anteroposterior and lateral projections). During the procedure, the children were positioned and asked to remain still, and then they were asked to hold their breath. The X-rays were also carried out after the closed reduction and the skeletal traction. Then, the X-rays were performed on the third, fourteenth and thirtieth days of the hospital stay depending on the severity of the fracture and the regeneration process.

To measure blood microcirculation, laser Doppler flowmetry was employed. This technique is non-invasive and the study was conducted using Lakk-20 laser Doppler flowmeter produced in Russia. The technique is based on laser probing and measures the change in frequency induced by moving red blood cells to the illuminating coherent light. Laser Doppler flowmetry that measures blood cell perfusion was applied to analyze the use of cryotherapy. Laser Doppler flowmetry was undertaken twenty minutes before and twenty minutes after the treatment of the damaged area.

RESULTS AND DISCUSSION

The findings of the blood tests were compared with the normal range of healthy children. The normal values of ionized calcium, inorganic phosphorus and alkaline phosphatase of healthy children aged 3-7 are 1.16 mmol/L, 1.18 mmol/L, and 105.7 U/L respectively, for children aged 8-14, the normal values are 1.09 mmol/L, 1.22 mmol/L, and 102 U/L, respectively. When the results of the blood tests taken by the children with long bone fractures were studied, the increased ALP activity was registered ($p_1<0.05$) **(Table 1)**.

Table 1. The Results of the Blood Tests of the Experimental and Control Groups (X ± m)

Parameter	Sampling period	The results of the blood tests of the children with long bone fractures			
		Group 1 (received Calcimax Recovery dietary supplement)		Group 2 (received traditional treatment)	
		Aged 3–7 $n = 11$	Aged 8–14 $n = 5$	Aged 3–7 $n = 14$	Aged 8–14 $n = 5$
ALP U/L	Day 1	241.42 ± 13.60 $p_1 < 0.05$	234.22 ± 10.33 $p_1 < 0.05$	217.79 ± 10.32 $p_1 < 0.05$	237.81 ± 11.25 $p_1 < 0.05$

	Day 14	220.85 ± 13.60 $p_1 < 0.05$	204.22 ± 11.10 $p_1 < 0.05$	208.85 ± 19.62 $p_1 < 0.05$	205.58 ± 10.74 $p_1 < 0.05$
	Day 30	131.14 ± 14.19 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$	140.70 ± 7.11 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$	129.80 ± 8.54 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$	130.70 ± 11.18 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$
Ca^{2+} mmol/L	Day 1	1.26 ± 0.07	1.20 ± 0.06	1.31 ± 0.12	1.12 ± 0.01
	Day 14	1.14 ± 0.03	1.12 ± 0.11	1.08 ± 0.05	1.20 ± 0.10
	Day 30	1.06 ± 0.02	1.08 ± 0.07	1.18 ± 0.01	1.06 ± 0.10
P mmol/L	Day 1	1.82 ± 0.07 $p_1 < 0.05$	1.56 ± 0.07 $p_1 < 0.05$	2.03 ± 0.08 $p_1 < 0.05$	1.56± 07 $p_1 < 0.05$ $p_3 < 0.05$
	Day 14	1.55 ± 0.03 $p_1 < 0.05$ $p_4 < 0.05$	1.83 ± 0.13 $p_1 < 0.05$	1.82 ± 0.04 $p_1 < 0.05$ $p_2 < 0.05$ $p_4 < 0.05$	1.60 ± 06 $p_1 < 0.05$
	Day 30	1.31 ± 0.03 $p_1 < 0.05$ $p_4 < 0.05$	1.46 ± 0.09 $p_1 < 0.05$ $p_4 < 0.05$	1.40 ± 0.02 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$	1.41 ± 0.07 $p_1 < 0.05$ $p_4 < 0.05$

Note: Hereinafter, p_1 refers to the statistically significant difference when compared to the normal values of healthy children of the same age; p_2 - children with fractures of the 1st and 2nd groups of the same age; p_3 - children aged 3–7 and 8–14; p_4 – when compared to the values of children of the same age with long bone fractures obtained before treatment; p_5 - the statistically significant difference when compared to the values of children of the same age with long bone fractures obtained on the 14th day after the start of the treatment.

According to the results of the blood tests taken on the first day of the trial, the phosphorous levels were 1.5 times as high as that for the children aged 3-7, and 1.3 times as high as that for the children aged 8-14 ($p_1 < 0.05$). At the same time, the phosphorous levels in the blood of the children aged 8-14 were lower than the phosphorous levels in the blood of the children aged 3-7 ($p_3 < 0.05$). The results of the blood tests taken on the fourteenth day of the trial demonstrated that the phosphorous levels were still higher than normal ($p_1 < 0.05$), although the phosphorous levels in the blood of the children aged 3-7 lowered compared with the results obtained on the first day of the trial ($p_4 < 0.05$). The results of the blood tests taken on the thirtieth day of the trial demonstrated that the phosphorous levels in the blood of the children aged 3-7 were 1.2 times as high as the phosphorous levels in the blood of the children aged 3-7 who received Calcimax Recovery ($p_2 < 0.05$). During the clinical trial, the ALP activity was decreasing, although it still remained higher than the normal values.

The average normal value of osteocalcin for children aged 3-7 is 83.95 ng/ml and it is 104.02 ng/ml for children aged 8-14. The results of the tests taken on the first day of the trial indicated that the children from the second group had a lower level of osteocalcin, although the children aged 8-14 demonstrated a higher level of osteocalcin than the children aged 3-7 ($p_3 < 0.05$) **(Table 2)**.

Table 2. The Impact of Calcimax Recovery on the Osteocalcin Level Registered during the Trial (X ± m)

Sampling period	The osteocalcin level (ng/ml)			
	Group 1 (received Calcimax Recovery dietary supplement)		Group 2 (received traditional treatment)	
	Aged 3–7 *n* = 11	Aged 8–14 *n* = 5	Aged 3–7 *n* = 14	Aged 8–14 *n* = 5
Day 1	83.89 ± 5.56 $p_1 < 0.05$	93.08 ± 12.4	68.12 ± 4.42 $p_1 < 0.05$ $p_4 < 0.05$	83.44 ± 4.04 $p_1 < 0.05$ $p_2 < 0.05$ $p_3 < 0.05$
Day 14	72.40 ± 2.46 $p_4 < 0.05$	91.91 ± 13.10 $p_3 < 0.05$	57.94 ± 4.04 $p_1 < 0.05$ $p_2 < 0.05$ $p_4 < 0.05$	72.89 ± 3.45 $p_1 < 0.05$ $p_2 < 0.05$ $p_3 < 0.05$
Day 30	65.32 ± 2.09 $p_1 < 0.05$ $p_4 < 0.05$	69.11 ± 4.23 $p_1 < 0.05$ $p_4 < 0.05$	59.65 ± 2.92 $p_1 < 0.05$ $p_4 < 0.05$	59.92 ± 2.31 $p_1 < 0.05$ $p_4 < 0.05$

When the results of the following blood tests taken by the children aged 3-7 from both groups were compared, the children aged 3-7 who received traditional treatment still had a lower level of osteocalcin ($p_2 < 0.05$).

The comparison of the osteocalcin levels in the blood serum revealed that the children aged 3-7 and 8-14 who were prescribed traditional treatment had the osteocalcin levels 1.4 times as low as the normal levels ($p_1 < 0.05$), while the children prescribed Calcimax Recovery had their osteocalcin levels within the normal limits.

To assess blood clotting function in children with long bones fractures, the following tests were taken: the Prothrombin Time (PT), the Activated Partial Thromboplastin Time (APTT), and the fibrinogen activity test. The changes in the osteocalcin level in the children with long bone fractures are shown in **Figure 1**.

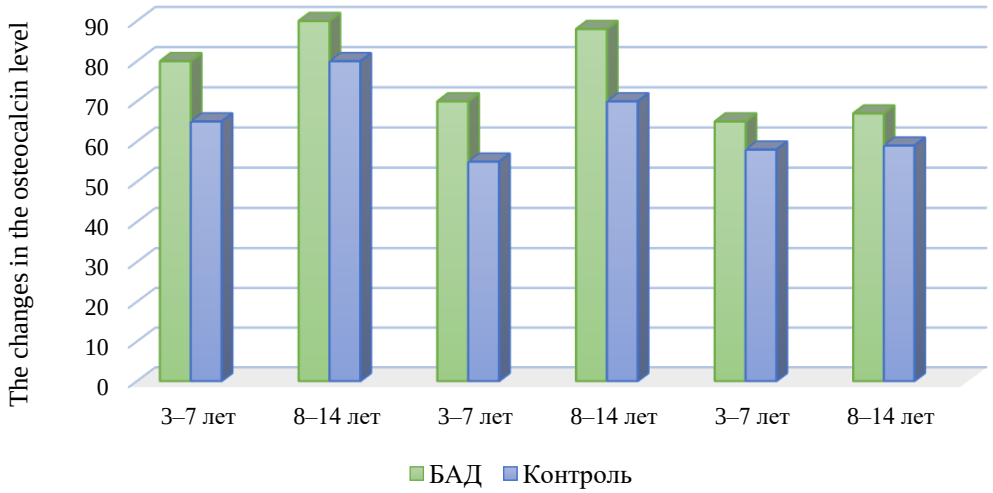

Figure 1. The Changes in the Osteocalcin Level in the Children with Long Bone Fractures

When comparing the data on the activated partial thromboplastin time in children with long bone fractures, it was noted that the children aged 3-7 and 8-14 with long bone fractures had a prolonged activated partial thromboplastin time ($p_1 < 0.05$). The activated partial thromboplastin time was 1.2 times as long as the normal value for both age groups of the children who took Calcimax Recovery dietary supplement, and in the children who were administered the traditional treatment the activated partial thromboplastin time was 1.1 times as long as the normal value for the 3-7 age group and 1.2 times for the 8-14 age group. The prolonged prothrombin time (PT) was detected in all children with long bone fractures ($p_1 < 0.05$), and was recorded as 1.4 times as long as the normal value **(Table 3)**.

Table 3. The Impact of Calcimax Recovery on the Blood Clotting Function (X ± m)

Parameter	Sampling period	The results of the blood clotting function tests			
		Group 1 (received Calcimax Recovery dietary supplement)		Group 2 (received traditional treatment)	
		Aged 3–7 $n = 11$	Aged 8–14 $n = 5$	Aged 3–7 $n = 11$	Aged 8–14 $n = 5$
The prothrombin time (PT), seconds	Day 1	21.60 ± 0.78 $p_1 < 0.05$	21.40 ± 0.83 $p_1 < 0.05$	21.90 ± 0.73 $p_1 < 0.05$	22.00 ± 0.83 $p_1 < 0.05$
	Day 14	16.00 ± 0.31 $p_4 < 0.05$	14.60 ± 0.60 $p_1 < 0.05$	18.90 ± 0.45 $p_1 < 0.05$ $p_2 < 0.05$ $p_4 < 0.05$	19.40 ± 0.56 $p_1 < 0.05$ $p_2 < 0.05$ $p_4 < 0.05$
	Day 30	14.00 ± 0.60 $p_4 < 0.05$ $p_5 < 0.05$	14.10 ± 0.53 $p_4 < 0.05$	15.00 ± 0.49 $p_4 < 0.05$ $p_5 < 0.05$	14.00 ± 0.61 $p_4 < 0.05$ $p_5 < 0.05$
The activated partial thromboplastin time (APTT), seconds	Day 1	41.40 ± 0.81 $p_1 < 0.05$	41.40 ± 0.8 $p_1 < 0.05$	39.20 ± 1.10 $p_1 < 0.05$	41.70 ± 1.06 $p_1 < 0.05$
	Day 14	33.10 ± 0.86 $p_4 < 0.05$	32.70 ± 0.80 $p_4 < 0.05$	35.00 ± 0.87 $p_4 < 0.05$	28.80 ± 1.32 $p_4 < 0.05$

	Day 30	30.80 ± 0.67 $p_4 < 0.05$ $p_5 < 0.05$	31.70 ± 0.72 $p_4 < 0.05$ $p_5 < 0.05$	26.60 ± 1.05 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$	24.40 ± 1.33 $p_1 < 0.05$ $p_4 < 0.05$ $p_5 < 0.05$
The fibrinogen activity, g/L	Day 1	2.92 ± 0.54	3.53 ± 0.56	3.10 ± 0.85	3.41 ± 0.78
	Day 14	2.26 ± 0.03	3.00 ± 0.64	2.76 ± 0.54	2.71 ± 0.52
	Day 30	2.44 ± 0.64	2.93 ± 0.52	2.57 ± 0.39	2.64 ± 0.69

The results of the fibrinogen activity tests obtained before and after the treatment in all the age groups were within the normal range ($p_1 > 0.05$).

The results of the prothrombin time tests taken on day 14 of the trial demonstrated that for the children from Group 2 (who received the traditional treatment) the prothrombin time was still prolonged, while the tests results of the children from Group 1 (who underwent cryotherapy) were close to the normal value ($p_1 > 0.05$).

It should be noted that the activated partial thromboplastin time was shortening when compared with the normal value ($p_1 < 0,05$) and the tests results for the children from Group 1 (who received Calcimax Recovery dietary supplement).

The data obtained from the tests done on day 30 demonstrated the significant reduction in both PT and APPT for all the children of Groups 1 and 2. The reduction is noticeable for the data received on day 1 ($p_4 < 0.05$) and day 14 of the trial ($p_5 < 0,05$). Those children with long bone fractures who received Calcimax Recovery dietary supplement reported the decreased pain intensity in the early days of the trial.

The data available from numerous research and our own studies enabled us to design the dietary supplement formula to treat the metabolic bone tissue disorders. The ingredients and their functional properties are shown in **Figure 2**.

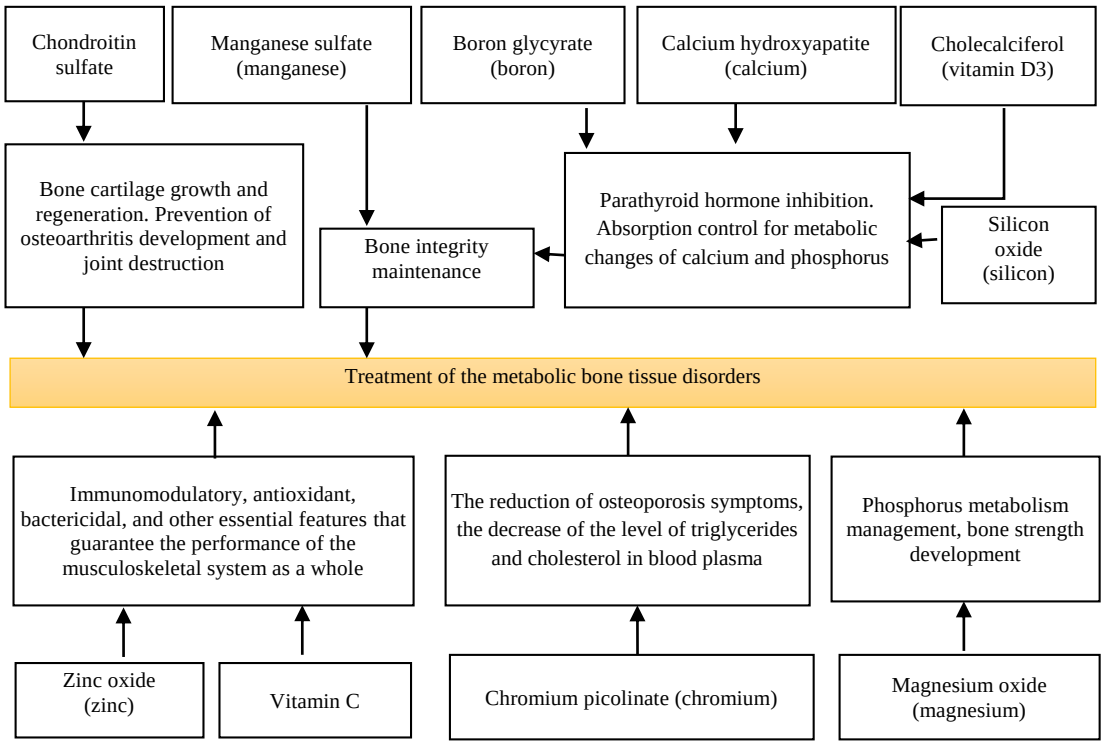

Figure 2. The Ingredients of Calcimax Recovery Dietary Supplement and their Impact on Managing Metabolic Disorders of Bone Tissue

To illustrate the efficacy of Calcimax Recovery dietary supplement, let us consider the treatment of a closed, right-sided, midshaft femur fracture that a five-year-old child sustained. Upon hospital admission, a closed reduction was performed to treat the displaced fracture. The manipulation was carried out under sedation for which local anaesthetic (novocaine, 50 ml) was used. The results from the X-ray indicated a fracture displacement. To treat the patient, analgesics, cryotherapy, the skeleton traction and the Balkan frame with a

weight of 3 kg were employed. The patient received Calcimax Recovery dietary supplement as a complementary treatment.

During the treatment, the following tests were performed: blood counts, coagulation tests, osteocalcin blood test, X-rays, and laser Doppler flowmetry to monitor blood microcirculation. When the results of the tests were compared with the results of tests of the patients who were not administered cryotherapy, a high osteocalcin activity was registered. The osteocalcin level reached 69 ng/ml during the second week of the hospital stay, while the patients who underwent traditional treatment demonstrated a 17% lower osteocalcin level (the average was 58 ng/ml). The ALP activity checked on day 10 of the treatment was 309 mmol/L. The results of the blood tests for ionized calcium and phosphorus stated 2.13 mmol/L of ionized calcium and 1.55 mmol/L of phosphorus. The laser Doppler flowmetry run on days 3, 8 and 16 confirmed the improved microcirculation. It should be noted that the blood circulation was 9% higher right after the cryotherapy and 5 % higher two hours after the procedure.

According to the results of X-ray, the first signs of callus formation were registered during the second week of the hospital stay, which was 4 days earlier than normal. The results of the coagulation test done during the first week of the treatment indicated a higher microcirculation activity than the normal, with the fibrinogen level of 5.3 g/L (the fibrinolytic activity of 7 minutes).

The patient stayed in hospital for 28 days, which is 3.5 days less when compared with the length of a usual hospital stay of the patients prescribed traditional treatment. The patient regained the full range of motion 3 weeks after the treatment.

The positive influence of Calcimax Recovery dietary supplement on the musculoskeletal system can be explained by the functional properties of its active ingredients and their synergistic effect on the metabolic processes.

CONCLUSION

The findings of the trial allow us to conclude the following: the use of dietary supplements in complex therapy makes a beneficial impact on the recovery processes and joints mobility in children with long bone fractures, improves patients' general well-being, prevents trophic changes of soft tissues, hinders the development of post-traumatic edema and allows for the reduction in analgesics use. Calcimax Recovery dietary supplement assists in the bone remodelling process and provides a favourable effect on the overall recovery period.

The findings of the present trial can help in the development of the complex therapy to stimulate bone metabolism and fracture healing in both children and adults.

ACKNOWLEDGMENTS : None

CONFLICT OF INTEREST : None

FINANCIAL SUPPORT : None

ETHICS STATEMENT : Participation in this research was entirely voluntary.

REFERENCES

1. Kuzhelivsky II, Urazova OI, Slizovskii GV, Maslikov VM. Dynamics of the content of osteocalcin and hemostasiological parameters of blood treated with cryotherapy in children with fractures of long bones. Sib Med J. 2008;(5):30-3.
2. Hayhoe RP, Lentjes MA, Luben RN, Khaw KT, Welch AA. Dietary magnesium and potassium intakes and circulating magnesium are associated with heel bone ultrasound attenuation and osteoporotic fracture risk in the EPICNorfolk cohort study. Am J Clin Nutr. 2015;102(2):376-84.
3. Evans E, Piccio L, Cross AH. Use of vitamins and dietary supplements by patients with multiple sclerosis: a review. JAMA Neurol. 2018;75(8):1013-21.
4. Kunakova RV, Zainullin RA, Khusnutdinova EK, Yalaev BI. Healthy nutrition of the XXI century: functional food products and nutrigenomics. Bull Acad Sci Repub Bashkortostan. 2016;21(3):5-14.
5. Karpouzos A, Diamantis E, Farmaki P, Savvanis S, Troupis T. Nutritional aspects of bone health and fracture healing. J Osteoporos. 2017;(8):1-10.

6. Cooper C, Dawson-Hughes B, Gordon CM, Rizzoli R. Healthy nutrition, healthy bones: how nutritional factors affect musculoskeletal health throughout life. Int Osteoporos Found, 2015.
7. Nilsson GE, Tenland T, Oberg PA. Evaluation of a laser Doppler flowmeter for measurement of tissue blood flow. IEEE Trans Biomed Eng. 1980;27(10):597-604.
8. Tokhiriyon B, Poznyakovsky VM, Beliaev NM. Biologically active complex for functional support of connective tissue: Scientific substantiation, clinical evidence. Int J Pharm Res Allied Sci. 2019;8(10):115-22.
9. Tokhiriyon B, Poznyakovsky VM, Andrievskikh S. Biologically active complex for multifactorial support of the central nervous system: new composition, efficacy. Carpath J Food Sci Technol. 2020;12(1):52-60.
10. Tokhiriyon B, Poznyakovsky VM. Full-scale testing of functional product in patients with vegetative-vascular dysfunction and chronic cerebrovascular disorder. Int J Pharm Res Allied Sci. 2019;8(3):91-7.
11. Kaishev VG, Seregin SN. Functional food products: the basis for disease prevention, health promotion and active longevity. Food Ind. 2017;(7):8-14.
12. Karagodin VP, Yurina OV. Assessment of safety and quality of biologically active food additives using two methods of biotesting. Technol Comm Sci Innov Food Prod. 2018;6(53):90-4.
13. Karamnova NS. Use of functional food products in the prevention of chronic non-infectious diseases. CardioSomatics. 2017;8(1):39.
14. Moiseenko MS, Mukatova MD. Food products of functional orientation and their purpose. Bull Astrakhan State Tech Univ. Ser: Fish. 2019;(1):145-52.
15. Biloklytska G, Viala S, Koval A. Marketing research on dietary supplements for periodontitis in patient diabetes. Ann Dent Spec. 2020;8(2):67-78.
16. Alabdulwahed RA, Khoja SO, Elshal MF, Bahlas S, AlGhamdi SA, Khojah SM. Modulation of CD28 and p38 Expression by Treatment with Vitamin D in Patients with Rheumatoid Arthritis. J Biochem Technol. 2019;10(3):76-82.