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## The Use of the Natural Amorphous Magnesite as a Source of Correction of the Magnesium Deficiency Condition.

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### ABSTRACT

Deficiency of magnesium in the body of an experimental animal may be corrected by means of oral administration of magnesium carbonate produced from the natural magnesium-containing mineral – magnesite. Keeping the rats of the test group on a magnesium-free diet resulted in the decrease in the magnesium content in the blood by 6.41% and by alimentary delivery of magnesium carbonate the macroelement level increased by 3.58% as compared to the period of the magnesium-free diet. The animals from the test group demonstrated after nutritional intervention of magnesium carbonate the activation of the reactive body state expressed in the left deviation which suggests the overstressing of the regulatory body systems. The diet on the basis of the corn starch and soya protein may be used by simulation of hypomagnesaemia in the small laboratory animals.

**Keywords:** magnesite, magnesium carbonate, magnesium deficiency, electron probe microanalysis, differential leukocyte count.

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## INTRODUCTION

The deficiency of magnesium is one of the predisposing factors of development of the cardiovascular diseases, primary hypertension, stroke, Alzheimer’s disease, increases the cancer risks [1-4]. As of today the magnesium deficiency can be bridged in two ways: the first one is the oral administration of the physiological doses of magnesium ions or intravenous administration of the pharmacological magnesium dose. It is understood that the first method of correction is the most favorable one since it is not followed by adverse effects and does not constitute the overdosing danger [5]. Taking into account the high incidence of diseases related to the magnesium deficiency condition the topical issue of the modern health care service is the search for the readily available, effective natural dietary supplements.

The objective of this study is the evaluation of efficiency of use of magnesium carbonate produced from the natural amorphous magnesite as a source of nutritional (alimentary) magnesium for hypomagnesaemia correction.

## MATERIALS AND METHODS

Within the frameworks of this study we used as a dietary supplement for correction of a magnesium-deficiency condition the magnesium carbonate obtained during the process of separation of the natural amorphous magnesite (Khalilovsky deposit, Orenburg Region, Russia). The magnesite concentrate derived contained the basic product – magnesium carbonate ( $MgCO_3$ ) in the amount of not less than 95%, calcium carbonate – not more than 3%, serpentine – not more than 2%, content of other materials made not more than 1%. The magnesite concentrate obtained was grinded in two stages. At the first stage the concentrate was grinded in the ball mill PULVERISETTE 5 during 1 hour, 300 rpm. At the second stage the powder was grinded in the brushing refiner at the pressure of 10 bars. The microstructure and elementary composition of the fine magnesite powder was investigated with the use of the scanning electron microscope Nova NanoSem450 and Quanta 200 3D equipped with the energy dispersive X-rays analyzer (Fig. 1).

The analysis of dispersability of the magnesite powder was conducted with the use of the laser particle-size analyzer “Analysette 22 NanoTec”. The average size of the powder made  $3.5 \mu m$  (Fig. 2). The specific surface area of the powder obtained made  $41226.09 \text{ cm}^2/\text{cm}^3$ . The number of the powder particles smaller than  $1 \mu m$  made 25%.

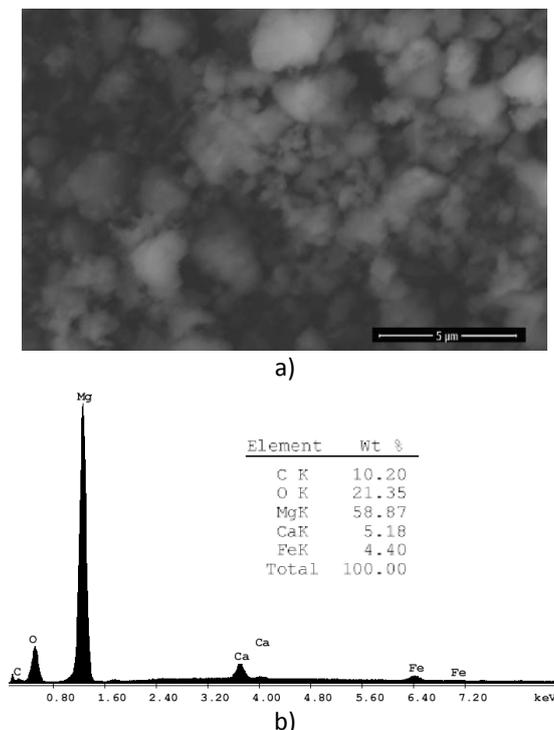
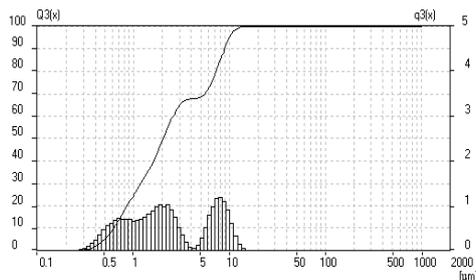


Figure 1: Microstructure (a) and elementary composition of the crushed magnesite powder (b) (SEM).



**Figure 2 Distribution of grain size of the crushed magnesium powder.**

Accumulation curve in the coordinates  $Q3(x)=f(\mu\text{m})$  (left scale) – each point at the curve shows which % of the sample has the particles lesser or greater than the specified value. Histogram within the coordinates  $q3(x)=f(\mu\text{m})$  (right scale) – the number of the samples with the specified particle size.

The analysis performed involved 20 male Wistar rats weighting 250 – 300 grams (“Stolbovaya” nursery, Moscow Region, Russia). For the experimental purposes the animals were divided into 2 groups in a random manner: the test and control one containing 12 species per each.

The animals of the control group were kept on a standard vivarium diet with free access to drinking water while the rats from the control group were kept on the magnesium-deficient diet and used distilled water during the first month of the experiment.

The magnesium-deficient diet was composed with regard to the existing diet produced by the company ICN Biomedicals Inc. (Aurora, Ohio, USA) [6, 7]. The main component of the diet was the corn starch (70%) whey protein (290%) and excipients (1%): vitamins: B1, B2, B3, B6, B12, E, H; vitamin C; pantothenic acid, folic acid.

Upon the expiry of the first month of experiment the experimental animals were subjected to the blood spot sampling (100  $\mu\text{l}$ ) from the tail vein for detection of magnesium and counting the leukocytes and the total number of the white blood cells. For the determination of the magnesium level a spot was put on a glass slide, air-dried and placed in the chamber of the scanning electron microscope Quanta 200 3D (FEI) for the electron probe microanalysis with use of the attachment TEAM EDS (EDAX).

The white blood cells were counted according to the standard formula where the leucogram is normally composed of the ratio between various white blood cells. A blood spot was applied to the degreased and clean slides. A slide with a blood spot was held with the left-hand fingers and the right-hand fingers held the ground glass that was put at the angle of  $45^\circ$  into the blood spot. Then after the blood spot ran over the edges of the ground slide this slide was moved forward along the slide so that the blood continuously towed along the edge of the ground slide. Then the blood film was air-dried, fixed with ethanol for 20 minutes and subjected to Romanowsky staining for 10 minutes, washed, air-dried and microscoped with the use of the optical microscope ECLIPSE E200 (Nikon, Japan).

The white blood cells were counted by means of the counter of white blood cells in the stained blood smears in the amount of 100 units divided into populations: neutrophils, eosinophils, basophiles, lymphocytes, monocytes. The total number of leukocytes in  $1 \text{ mm}^3$  of blood was calculated with the use of the Goryaev chamber. Using the data obtained: WBC differential and total number of leukocytes the leukocyte profile was specified consisting in the graphic recording of relationships between the main leukocyte groups and representation of the absolute content in  $1 \text{ mm}^3$  of blood for each of these groups.

After the decrease of the magnesium level in the blood was detected the rats of the test group began to receive through the feeding tube the magnesium-containing solution ( $\text{MgCO}_3+\text{H}_2\text{O}$ ) at 50 mg of elementary magnesium per 1 kg of the animal weight. At the end of the 2d month the animals of the test group were subjected to the blood spot sampling from the tail vein for detection of increase in the magnesium level in the blood after the deficiency thereof.

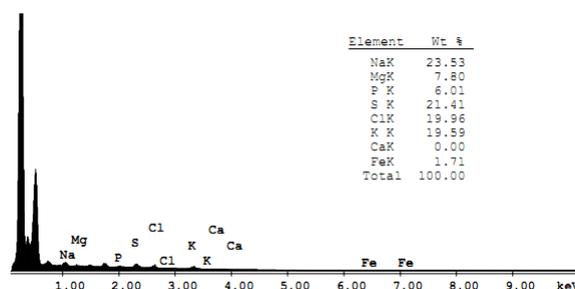
The conclusive results were obtained with the use of the computer variation statistics programs and the non-parametric Student's t-test.

### RESULTS

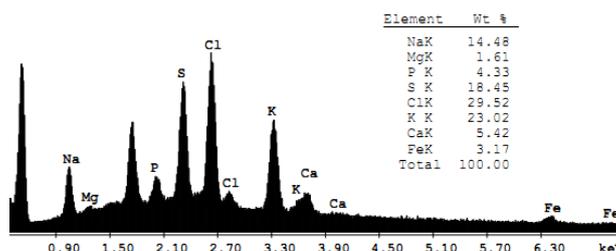
In the course of the study it was found out that the developed diet results in the magnesium deficiency in the peripheral blood and may be used by simulation of hypomagnesaemia in the small laboratory animals. Thus, in the animals kept on the magnesium-free diet the changes in the appearance were observed that were expressed in coat fading, hyperemia of the open skin areas (ears, tail, and legs). It was found that in the rats of the test group kept on the magnesium-free diet the concentration of magnesium in the peripheral blood was decreased by 6.41% as compared to the control group. Along with that by oral delivery of magnesium ( $MgCO_3+H_2O$ ) during the second month the level thereof in the peripheral blood of the test group rats increases by 3.58% as compared to the period of magnesium-free diet. However, the magnesium level attained remains lower than the values of the control group by 2.83% (Table 1, Fig. 3, 4, 5).

**Table 1: Weight content (wt %) of magnesium in the experimental groups (data obtained by means of the electron probe microanalysis)**

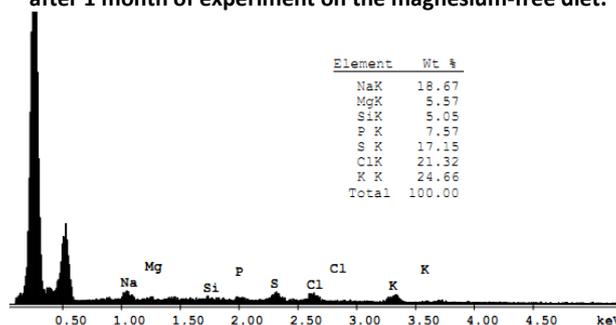
Control group (1st month)	Test group (1st month)	Test group (2d month)
8.31±3,90#*	1.90±0,44#	5.48±1,11*
<p><i>Remark: The differences are significant at <math>p \leq 0,01</math> # - control/test group (1<sup>st</sup> month), at <math>p \leq 0,05</math> * - control group/test group (2 months)</i></p>		



**Figure 3: The spectrum of the characteristic X-ray radiation obtained from a blood spot of the rat №1 of the control group after 1 month of the experiment.**



**Figure 4: The spectrum of the characteristic X-ray radiation obtained from a blood spot of the rat №1 of the test group after 1 month of experiment on the magnesium-free diet.**



**Figure 5: The spectrum of the characteristic X-ray radiation obtained from a blood spot of the rat №1 of the test group after 2 months of experiment by the correction of the magnesium deficiency by magnesium carbonate.**

The analysis of leucogram data, total number of leukocytes and leukocyte profile of the control and test group animals showed that in the animals of the control group after introduction of magnesium carbonate to the diet the activation of the reactive body state after correction of the magnesium deficient condition was observed. Thus, in the test group the left deviation is observed which suggests the oversteering of the regulatory body systems (Table 2).

**Table 2: White cell count in the rats of the control and test groups**

White blood cells	Control group	Test group after magnesium deficiency	Test group after correction of the magnesium-deficient diet
lymphocytes	42,0±11.74	43,37±12.32	46.75±10.47
monocytes	21.25±12.20	23.88±10.94	16.50±3.51
segmented neutrophils	30.88±7.04	29.75±7.15	24.63±8.62
band neutrophils	5.87±2.90#	3.00±1.51#*	12.13±7.20#*
basophiles	0	0	0
eosinophils	0	0	0
<p><i>Remark: The differences are significant at <math>p \leq 0,01</math> # - control/test group, * - test group after the magnesium deficiency/test group after correction of the magnesium deficiency</i></p>			

It was found out that in the control group the total number of leukocytes in 1 mm<sup>3</sup> of blood made 12000±2.5. On the basis of the white blood cell count the leukocyte profile was specified. Thus, the absolute count in the control group made: band neutrophils – 704.4; segmented neutrophils – 3705.6; lymphocytes – 5040.0; monocytes – 2550. In the test group after the magnesium deficiency the total number of leukocytes in 1 mm<sup>3</sup> of blood made 11000±1.5, and the absolute count in the test group: band neutrophils – 330.0; segmented neutrophils – 3272.5; lymphocytes – 4770.7; monocytes – 2626.8. It was also found that in the test group after correction of the magnesium-deficient diet the total number of leukocytes in 1 mm<sup>3</sup> of blood made 14000±2.3. The absolute count in the test group: band neutrophils – 1698.2; segmented neutrophils – 3448.2; lymphocytes – 6546.0; monocytes – 2310.0. On the basis of the normal range for absolute counts of each kind of leukocytes in 1mm<sup>3</sup> of peripheral blood it can be stated that the number of the band neutrophils in the test group after correction of the magnesium-deficient diet was increased as compared to the control group and values obtained during the period of the magnesium-deficient diet.

**SUMMARY**

The deficiency of magnesium in the body of a test animal may be corrected by oral administration of magnesium carbonate produced from the natural magnesium-containing mineral – magnesite. Keeping the rats of the test group on a magnesium-free diet resulted in the decrease in the magnesium content in the blood by 6.41% and by alimentary delivery of magnesium carbonate the macroelement level increased by 3.58% as compared to the period of the magnesium-free diet. The animals from the test group demonstrated after nutritional intervention of magnesium carbonate the activation of the reactive body state expressed in the left deviation which suggests the oversteering of the regulatory body systems. The diet on the basis of the corn starch and soya protein may be used by simulation of hypomagnesaemia in the small laboratory animals.

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powders by disposing of wastes of amorphous magnesium enrichment process for different branches of industry”.

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