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Toxicity of monazite particulate and its attenuation with a complex of bio-protectors

B.A. Katsnelson, Olga S. Yeremenko, Larissa I. Privalova, O.H. Makeyev*, Tamara D. Degtyareva, Olga Yu. Beresneva*, Irene E. Valamina*, Ilzira A. Minihaliyeva, Marina P. Sutunkova, Ekaterina P .Kireyeva, V.A. Bukhantsev*, E.I. Dovzenko*, V.V. Minin*, E.S. Kulikov*, Svetlana V. Kostykova*, A.S. Nazukin

Medical Research Center for Prophylaxis and Health Protection in Industrial Workers, Ekaterinburg, Russia
* Urals State Medical Academy, Ekaterinburg, Russia

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Monazite; radiotoxicity; bioprotectors

Summary
Background: *Workers employed on mining, processing and storage of monazite are at risk of exposure to dust with expected adverse health effects.* **Objectives:** *To study the adverse health effects of monazite particles in experiments on rats and to test the possibility of attenuating these effects.* **Methods:** *Outbred white rats were injected intratracheally with a suspension of ground monazite concentrate (MC) in order to investigate the cellular response of the lower airways 24 hours later and the organism's status 6 months after the injection. The bio-protective complex (BPC) tested in these experiments consisted of glutamate, an iodine preparation, methionine, a polyvitamin-polymineral composition, and/or "Eicosavitol" (fish oil preparation rich in PUFA, predominantly of the ω3-group). Bio-protectors were administered together with the rat food and drink daily for one month before the MC injection in the short-term experiment, or over 6 months after such injection in the long-term experiment.* **Results:** *MC induced manifestations of its cytotoxicity, fibrogenicity and systemic toxicity as well as genotoxicity. The tested BPC attenuated virtually all these effects. Although a similar protective potential of "Eicosavitol" against almost all of them was lower compared with that of BPC, combining BPC with "Eicosavitol" provided, as a rule, the greatest protective effect.* **Conclusion:** *It may be assumed that the many-sided adverse effects of MC on the organism is due, at least partially, to the presence in its composition of not only rare earth elements but also of natural radioisotopes of the thorium and uranium families. The combination of the bio-protectors tested was highly effective and may be recommended for administering in periodic preventive programmes to exposed workers.*

Riassunto
«Tossicità del particolato di monazite e sua attenuazione con un complesso di bioprotettori» *I lavoratori addetti all'estrazione della monozite ed alle successive operazioni fino allo stoccaggio possono essere a rischio con possibili effetti sulla loro salute data l'esposizione a polvere. Per valutare i possibili effetti nocivi sulla salute delle particelle di monazite sono stati condotti studi sperimentali su ratti con lo scopo anche di valutare le possibilità di attenuare questi effetti con metodi farmacologici. Più in particolare, a gruppi di ratti è stata iniettata per via intratracheale una sospensione di particelle di monazite (MC) ed è stata valutata la risposta cellulare delle vie aeree profonde 24 ore dopo e le condizioni generali dell'animale a distanza di sei mesi. Il complesso bioprotettore(BPC) testato in questi*

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Corrispondenza: Professor Boris A. Katsnelson, MD, DSc, Dept. of Toxicology and Biological Prophylaxis, Medical Research Center for Prophylaxis & Health Protection, 30 Popov Str, Ekaterinburg, 620014 Russia -Tel. 7- 343- 3718-721
Fax 7- 343- 3718-740 - E-mail: bkaznelson@ymrc.ru; bkaznelson@etel.ru

esperimenti era composto da glutammato, un preparato a base di sodio, metionina, un composto polivitaminico e poliminerale e da Eicosavitol (un preparato di olio di pesce ricco in PUFA, prevalentemente del gruppo ω3). I bioprotettori sono stati somministrati ai ratti insieme alla normale dieta ed all'acqua da bere ogni giorno per un mese prima dell'esperimento a breve termine e per tutti i sei mesi dopo il trattamento intratracheale nell'esperimento a lungo termine. I risultati hanno dimostrato che il particolato di monazite (MC) è in grado di essere citotossico, fibrogenico e capace di indurre una tossicità sistemica oltre ad essere genotossico. I bioprotettori (BPC) usati hanno attenuato virtualmente tutti questi effetti e se anche il solo Eicosavitol ha dimostrato una certa capacità protettiva nei riguardi di quasi tutti questi effetti, seppure inferiore a quella del BPC, la combinazione di quest'ultimo composto con l'Eicosavitol ha esercitato, di norma, un più elevato effetto protettivo. Si può ritenere che gli effetti nocivi esercitati dalla MC sull'organismo animale sia dovuto, almeno in parte, alla presenza non solo di elementi di terre rare, ma anche di isotopi naturali della famiglia del thorio e dell'uranio. I bioprotettori utilizzati si sono dimostrati altamente efficaci e sono pertanto da raccomandare perché vengano somministrati ai lavoratori esposti nel corso di programmi di prevenzione.

Introduction

Monazite is a mineral from the class of rare earth phosphates, mainly of the cerium group, but also thorium and, in much smaller quantities, uranium. It also contains oxides of rare earth elements (scandium, lutecium, lanthanum, cerium, terbium, europium, holmium, etc.). While the toxic effects of soluble rare earth compounds consist of damage to the inner organs (the liver, in particular) and various adverse effects on metabolic processes, low soluble oxides and phosphates are highly cytotoxic to alveolar macrophages and seem to be capable of causing pneumoconiosis (10, 11). At the same time, systemic toxicity and irritating effects are also inherent in such low soluble oxides of rare earth elements as yttrium barium cuprite ($YBa_2Cu_3O_7$) and lanthanum strontium cobaltite ($La_{0.7}Sr_{0.3}CoO_3$). (30).

Concerning the organism's response to the effects of low-soluble compounds of thorium and uranium, of paramount importance is their radiotoxicity, mainly due to alpha radiation. In the vicinity of large monazite masses the radiation effect on the human body is caused also by the release of thoron and radon and their decay daughters. This explains the concern associated with the mining and processing of monazite sands or with residence near places where wastes are accumulated after the extraction of thorium from monazite (5, 29). However, the deposition of inhaled particles of monazite dust in the airways is of special importance to those who work with monazite sands or concentrates. At the same time, we are not aware of any experimental studies estimating the various harmful effects of mineral monazite as such, with the exception of the half-century-old experiments by South African authors who demonstrated the possibility of pneumoconiosis development after intratracheal administration of monazite (40).

In the Middle Urals, we are faced with this problem due to the fact that monazite concentrate (MC) has been stored in the area at a warehouse for several decades. About 10% of samples of this MC contain at least 25% thorium and 4% uranium, and about.90% of samples contain 5% thorium and 0.2% uranium. The specific radioactivity of the thorium series radioisotopes in average MC samples is equal to 218 - 230 Bq/g. The unsatisfactory technical condition of the warehouse has caused hazards to the health of not only workers but also of the population of the nearby villages and, hence, considerable public concern. The regional government has developed a range of preventive measures including not only building and other engineering interventions but also studies to test the possibility of increasing the resistance of the worker's organism to the harmful effects of MC with the help of a set of innocuous substances producing a beneficial influence on the toxicokinetics and toxicodynamics of monazite.

Over a number of years a team of researchers led by the first author of this paper have pursued the theoretical development, experimental modelling,

controlled testing and implementation of methods of the so-called biological prophylaxis (14-25). By the latter we mean a complex influence on the organism aimed at enhancing its resistance to the harmful actions of environmental pollutants, both within and without the workplaces. It was shown, in particular, that " bioprophylactic complexes" (BPC), i.e. combinations of variously directed bioprotectors selected rationally taking into account both their pharmacodynamic characteristics and the toxicodynamic and toxicokinetic features of the harmful substances against which the organism is to be protected are, as a rule, more effective than isolated bioprotectors. It is essential that a BPC in itself should not produce any harmful effect at the preventive effective doses.

We found in a number of cases that a BPC inhibiting various toxic effects of metals and of some organic substances can have a beneficial effect on micronucleus assay results (21, 22, 25). One might assume, however, that a decrease in the number of micronuclei is not a manifestation of the direct anti-mutagenic action of some BPC ingredients but, rather, is connected with a decrease in the effective internal dose of the mutagenic metal resulting from the bene proven ficial toxicokinetic effect of the BPC proven by us in the same experiments. A legitimate question arose as to whether or not it might be possible to enhance such inherently *secondary* anti-mutagenic (and thus, presumably, anti-carcinogenic) effects of bioprophylaxis by adding to the same or similar BPCs some *primarily* anti-mutagenic agents that would increase the resistance of nuclear DNA to the genotoxic action of harmful substances, the efficacy of repairing the DNA damaged by them and/or the apoptosis of cells with non-repaired damage.

In this respect, we directed our attention to the marked anti-mutagenic properties of polyunsaturated fatty acids (PUFA) and biologically active derivatives of such of these that belong to essential PUFA, namely eicosanoids. In addition to the many other well-known physiological roles of the eicosanoids, they interact with highly sensitive binding sites on a double-stranded DNA molecule and lead to a B-Z conformation transition of DNA, which is one of the DNA repair enhancement mechanisms. However, the administration of eicosanoids to the organism is inadvisable because their half-life in the body in the case of parenteral injection does not exceed five minutes. For practical purposes, therefore, we suggested using "Eicosavitol", a preparation of oil from salmon, surgeon and whitefish with a high concentration of PUFA (pertaining mainly to the omega-3 group – 13-14% of the oil) available for the formation of eicosanoids inside a cell. It also included natural antioxidants (α-tocopherol, coenzyme Q_{10}-ubiquinone, ubichromenol), and liposoluble vitamins A, E, F. Indeed, the initial results obtained in an experiment on rats exposed to a combination of toxic and mutagenic metals, fluoride and benzo(a)pyrene demonstrated that "Eicosavitol" enhanced the anti-genotoxic effect of a BPC consisting of glutamate, a pectin enterosorbent, a multivitamin-multimineral complex, and a calcium additive (24). Thus further investigation of its protective properties seemed justified.

When choosing other bioprotectors for inclusion in the BPC to be tested in this case, we directed our attention, first of all, to glutamate. The number of studies devoted to the biochemistry of glutamate is enormous, and their review is beyond the scope of this paper. Briefly, it is well known that glutamic acid and its amide (glutamine), being the main collectors of non-protein nitrogen, play a key role in supplying it for the synthesis of non-essential amino-acids and, through them, of several biologically active compounds. They also take part in ammonia transfer, synthesis of purine bases and nucleic acids, transamination, urea synthesis, and several other metabolic transformations. Finally, glutamic acid, as one of the participants of the tricarbonic acid (Krebs') cycle, is closely connected with the energy metabolism of the cell, in particular, with phosphorylation of ADP to ATP, and thus favourably influences the stabilization of cell membranes. This may account for the well-established extraordinarily high ability of glutamate to protect the alveolar macrophage from the cytotoxic action of engulfed particles and, in this connection, hinder both dust retention in the lungs and tracheo-bronchial lymph nodes and development of silicotic and asbestotic fibrogenesis (18, 20, 24, 25,

27, 28) [(the multifaceted key role of the cytotoxic damage to macrophages in the pathogenesis of pneumoconiosis, see (14, 18, 37)].

We decided to additionally include iodine in the composition of the BPC tested as another bioprotector that had been successfully established as an agent inhibiting fibrogenesis in experimental silicosis (2, 34). Presumably, this effect is connected with the normalization of bio-energetic processes (disturbed in macrophage damaged by cytotoxic particles) through the influence of iodine on the hormonal function of the thyroid.

In the mechanisms responsible for the damaging action of dust particles on macrophages, a certain role is played by the induction of lipid peroxidation and other free-radical processes (18). The role of free oxygen radicals is also well known in DNA damage and thus, probably, in the initiation of carcinogenesis. The inclusion of bio-antioxidants in moderate dosages into the composition of BPC might, therefore, be expected to potentiate not only its anti-cytotoxic and anti-fibrogenic action [as was observed in experimental silicosis (18, 20)] but also its anti-mutagenic (and thus, predictably, anti-carcinogenic) effect. The purpose of our experiments in the field of biological prophylaxis is usually to test a BPC composition that would be not only efficacious but also convenient and safe for subsequent practical use. We therefore prefer including preventive doses of such antioxidants as selenium and some vitamins into BPCs tested in a balanced combination given by a ready-made multivitamin and multimineral preparation rather than providing them separately. In this study, however, we decided to include an additional dose of methionine in the composition of BPC as this amino acid plays an active role in lipid metabolism, which is usually damaged in pneumoconioses, and is also interesting as one of the components of the organism's antioxidant system.

Materials and Methods

An average sample of MC from the above warehouse, used in all experiments, was ground in an agate mortar into powder with particle size distribution corresponding to ordinary mineral disintegration aerosols; there were: 65% particles with projection diameter up to 2.5 um, 25%: >2.5 um up to 3.5 um, 10%: >3.5 um up to 5 um, with only solitary particles of greater dimensions, if the count was made under x600 magnification; or: 71.4% particles with diameter up to 1 um, 19.7%: >1 up to 2 um, 7.9%: >2 um up to 4 um, 0.2%: >4 um up to 5 um, and 0.8%: >5 um, if the count was made under x1350 magnification (with immersion).

The BPC tested in this study included the following ingredients:

1. glutamic acid (by OAO "Tatkhimpharmpreparaty", Russia) neutralized with sodium bicarbonate and consumed by rats by drinking a 1.5% solution (the daily consumption of this drink was 10-12 mL per rat); this dosage proved in our previous studies to be an effective antagonist of silica (18, 20, 26, 27), asbestos (25, 38) and different toxic metals (20);

2. polyvitamin and polymineral pills "Selmevit" (by "OAO UfaVITA", Russia) containing (per pill) vitamins A (1650 i.u), E (7.5 i.u.), C (35.00 mg), B_1 (0.581 mg), B_2 (1.00 mg), B_6 (2.50 mg), B_{12} (3.0 ug), nicotinamide (4.00 mg), folic acid (0.05 mg), rutin (12.50 mg), calcium pantotenate (2.50 mg), lipoic acid (1.00 mg), methionine (100.00 mg), Ca (25.00 mg), Mg (40.00 mg), P (30.00 mg), Fe (2.50 mg), Cu (0.40 mg), Zn (2.00 mg), Mn (1.25 mg), Se (25 ug), and Co (50 ug) – the dosage of which was calculated based on the needs of laboratory rats for basic vitamins as published in the literature and amounted to 10 crushed pills added to the food for 60 rats;

3. iodine preparation "Iodomarine" (by "Berlin-Chemie AG") (2.5 ground pills in the food for 60 animals, i.e. an average of 4 μg of iodine per rat);

4. methionine (by "OAO City-med-sorb", Russia) – 7 pills (250 g each) for 60 animals in addition to that contained in the polyvitamin preparation so that the total dose amounted to 50 mg per rat (also as a food admixture); and/or

5. "Eicosavitol" (by "OOO Farmavit", Russia) added as oil to the rats' food on the basis of 1 ml per rat daily.

All experiments were carried out on outbred white female rats (not SPF) with an initial body

weight of 150-220 g. MC samples were preliminary sterilized at 105° C for 24 hrs and then suspended aseptically in sterile normal saline (pharmaceutical solution for injections). To study changes in the cell composition of the bronchoalveolar lavage fluid (BALF) obtained 24 hours after exposure, MC was injected intratracheally (i.t). under visual control as a suspension containing 10 mg MC in 1 ml. To study chronic effects, rats were injected once in the same way with 50 mg of MC, and were killed by rapid decapitation in 6 months. Both short- and long-term experiments were carried out on six groups (20 rats in each), the first of which received only MC; the second group, which had been administered the same MC injection, then received BPC 5 times a week; the third group received the same plus Eicosavitol; the fourth group received MC and Eicosavitol without BPC; the fifth group was not administered MC but received BPC with Eicosavitol; and the sixth was a control group. In the short-term experiment, control rats were injected i.t. with 1 ml of sterile normal saline; in the long-term experiment they were not. All rats were housed in conventional conditions, breathed unfiltered air and were fed with standard balanced food. The experiments were planned and implemented in accordance with the "International guiding principles for biomedical research involving animals" developed by the Council for International Organizations of Medical Sciences (1985).

To estimate the intensity of intoxication and of pneumoconiosis, a set of indices was used which may be considered usual and are not described in detail in this Section as they will be understandable from the presentation of the results and their discussion. We consider it necessary, however, to dwell on tests for DNA damage and repair that were carried out on blood samples taken from rats 6 month after the beginning of the long-term experiment. To this end, we used:

1. *The DNA-comet assay.* Upon lysis of cells contained in the agarose layer, fragments of damaged DNA migrate in an electrical field towards the anode, forming a structure resembling a comet with a "head" (or "nucleus"), presenting a ball of native (uninjured) DNA, and a "tail" of migrating damaged DNA fragments. By the extent of DNA migration one can judge the extent of damage because, as compared with fragmented ("tail") DNA, non-fragmented nuclear DNA has a very low extent of migration in the agarose gel. In whole blood samples combined from the above groups of rats, the leukocyte fractions were separated in a phycoll-verografin gradient. Using fluorescent microscopy we investigated 100 cells (monocytes or lymphocytes) from each group, visually estimating DNA distribution in the agarose gel in relative units and determining the ratio between the "head" and the "tail". Semi-quantitatively, cells were assigned to 5 classes: class C1=practically uninjured cells (up to 5% of the DNA in the "tail"); class C2=low damage (5-20%); class C3=medium damage (20-40%); class C4=high damage (40-95%); class C5 = fully damaged cells (>95%).

2. *The amplified fragment length polymorphism method (AFLP).* In contrast with the previous method, which is based on subjective visual estimation of the degree of damage to DNA of cells, this procedure allows quantitative determination of the degree of DNA fragmentation. The theoretical basis for the development of this technique is provided by many studies (1, 3, 4, 6, 9, 32, 33, 39, 43). In our experiment, the AFLP test was performed on lymphocytes separated from rats' blood as stated above. The separation of DNA was done by the standard agarose gel-electrophoresis procedure. The polymerase chain reaction (PCR) was carried out using specific primers complementary to the Alu sequences and nucleotides (dCTP, dATP, methyl-dTTP) labelled with tritium. The amplificate obtained was separated by horizontal agarose-gel electrophoresis in the TAE-buffer at 100V during 15 min. After the end of electrophoresis, the gel plates were divided into tracks, each of which was cut into 5 mm long sections. The resulting fragments of the gel were placed into bottles containing 3.0 ml of absolute isopropanol. The bottles were then heated up to 80°C over 2 hours. Following extraction of the labelled amplified fragments from the gel, ordinary toluene scintillator (6.0 ml) was added to the contents of the bottles. The results were recorded with a Beta-2 automatic liquid scintillation counter. The groups were compared by

the quantitative ratio between the compactly arranged head and tail parts, the so-called DNA fragmentation coefficient (Cfr).

Results

A comparative study of the cell composition of the bronchoalveolar lavage fluid (BALF) in the rats of all groups (table 1) showed that i.t. administration of the MC suspension caused a sharp increase in free cell count in the lower airways, basically due to sharply enhanced recruitment of neutrophile leucocytes (NL) against the background of a less expressed but statistically significant increase in the number of alveolar macrophages (AM), which manifests itself in an almost 9-fold increase in the NL/AM ratio. In the animals that had been prepared through a month-long course of BPC, this ratio was significantly lower due to a substantial limitation of the recruitment of neutrophiles, and because of the combined action of BPC and Eicosavitol the NL/AM ratio dropped most significantly although Eicosavitol itself provided a considerably less marked effect than the BPC alone. The BPC+Eicosavitol complex reduced the BALF cell numbers also in rats which had not been injected with MC.

The chronic experiment showed that 6 months after the i.t. injection the monazite dust caused certain pneumoconiotic changes. There was a statistically significant (P<0.05) increase in the lung mass in comparison with the control group (from 1.351±0.076 g to 1.921±0.136 g for wet lung; from 288±16 mg to 430±28 mg for dry lung), however, no significant changes in this index were caused by the administration of BPC and/or Eicosavitol. As can be seen from table 2, there was also an increase in the lipid content of the lung tissue under the effect of MC.

There was a statistically significant attenuation of this increase in the rats given BPC (or BPC + Eicosavitol) after the administration of MC, and in this case also Eicosavitol without BPC demonstrated a less marked attenuation. Good agreement with quantitative data on the effect of the bioprotectors on the lipid contents of the lungs is shown by semi-quantitative morphometric assessment of their effect on the amount of sudanophilic (phosopholipid) granules in the lung macrophages (table 2).

Exposure to monazite was found to increase the total hydroxyproline content of the lung tissue as well (4155±302 μg against 2444±100 μg in the controls, P <0.05); this increase was also reduced (though not statistically significantly enough) under the influence of BPC (3788±324 μg), particularly of BPC in combination with Eicosavitol (3594±380 μg), and to a lesser degree under the influence of Eicosavitol alone (3986±197 μg).

The effect of bioprotectors on the residual mass of the mineral dust in the lungs (determined as the remainder after incineration of dry pulmonary tissue and dissolution of the ashes in 0.5 N HCl) was

Table 1 - Main cytological characteristics of BALF in rats 24 hours after intratracheal injection of a monazite concentrate suspension ($x \pm s_x$)

Exposure to:	Number of cells $*10^6$			NL/AM ratio
	Total	Neutrophile leukocytes (NL)	Alveolar macrophages (AM)	
Normal saline	2.48±0.15	0.24±0.05	1.90±0.15	0.13±0.03
Monazite	49.90±6.63*	42.96±6.02*	4.57±0.80*	10.18±1.17*
Monazite+BPC	33.01±3.55*•	26.01±3.01*•	5.19±0.93*	5.15±0.67*•
Monazite+BPC+Eicosavitol	29.78±5.09*•	22.23±3.74*•	6.27±1.26*	3.83±0.24*•
Monazite+Eicosavitol	33.86±5.33*	28.29±4.68*	3.56±0.51*	7.73±0.44*
BPC+Eicosavitol	2.64±0.29•	0.13±0.03•	2.11±0.26•	0.06±0.01•

* means the statistical significance of the difference compared to the control group;
• compared to the "Monazite" group (P<0.05 by Student's t-test)

Table 2 - The lipid contents of rat lungs 6 months after intratracheal injection of monazite concentrate suspension ($x \pm S_x$)

Exposure to:	Total lipid content, mg	Same related to 100 g of body weight	Number of sudanophylic granules, score[1]
–	26.73±1.80	12.15±0.85	1.26±0.11
Monazite	45.86±2.22*	20.43±1.12*	2.11±0.17*
Monazite+BPC	36.81±3.41*•	16.47±1.48*•	1.53±0.06*•
Monazite+BPC+Eicosavitol	36.66±3.95*	15.93±1.52*•	1.45±0.05•
Monazite+Eicosavitol	39.48±2.51*	18.14±1.37*	No data
BPC+Eicosavitol	29.33±2.03	13.07±0.9	No data

[1] staining with black Sudan B; the number of sudanophylic (phospholipid) granules in a pulmonary macrophage is estimated in points from 0 to 3, and a weighted average score is calculated for each animal

* means the statistical significance of the difference compared to the control group;

• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

also beneficial, though weak and statistically not significant. In the group administered MC only, it was equal to 30.8±2.9 mg, while in the other groups it was: «MC+BPC»=25.6±2,5 mg, «MC+BPC+Eicosavitol»=27.1±3.3 mg, «MC+Eicosavitol»=29.3±3.6 mg. However, the increase in the hydroxyproline content of the lungs diminished under the influence of bioprotectors not only in the above mentioned absolute indices but (in all cases, except for the action of Eicosavitol alone) also per unit residual mass of dust in the lungs: in the same groups as above it was equal to 58.8±6.6, 49.2±13.1, 37.4±13.8 and 57.1±7.7 μg/mg, respectively; this effect of the BPC in combination with Eicosavitol was statistically significant at $P<0.05$. This index was calculated for each rat injected with the dust as a difference between the absolute hydroxyproline content of its lungs and the average hydroxyproline content of the lungs of the rats in the control group divided by the residual mass of dust in the lungs of the same rat.

Morphological examination of rat lungs following MC injection revealed diffuse moderate sclerosis of the inter-alveolar septa and the development of multiple small spherical or irregular-shaped cellular-dust nodules within them, and only solitary thin arigrophylic fibres penetrated into these nodules (figure 1). With the bioprotectors administered, such nodules were identifiable mostly in the alveolar lumen and much less often in the depth of the inter-alveolar septa. In these rats, such nodules were surrounded with fine collagen fibres, but no ingrowth of any fibres into them was observed. In the tracheo-bronchial lymph nodes, MC caused the coarsening of agrirophylic stroma and initial signs of sclerosis of sinuses, but these phenomena were less marked under the effect of BPC or BPC combined with Eicosavitol.

Morphological and semi-quantitative morphometric investigation of the thyroid and the thymus revealed a negative effect of MC on these organs, as well as attenuation of this effect by BPC and particularly by BPC combined with Eicosavitol. Due to the length of this paper we can illustrate this statement with two examples only. The mean estimation score for the feature «foamy colloid» in

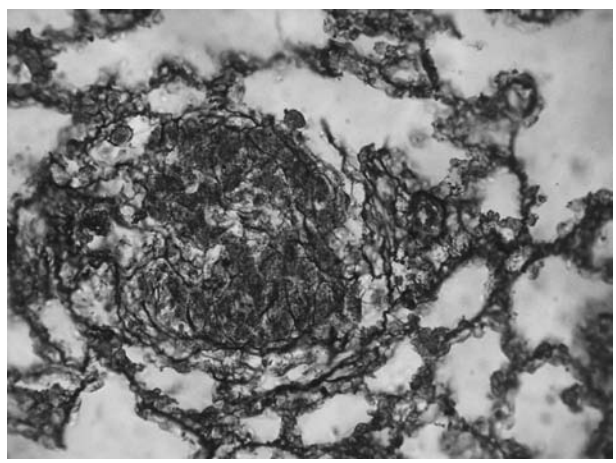

Figure 1 - One of many pneumoconiotic nodules in the lungs of a rat 6 months after intratracheal injection with monazite suspension. Gomori's silver impregnation stain. Magnification=x200

the thyroid follicles was equal to 0 in the rats of the control group, reaching 2.20±0.58 in those exposed to MC. Under the action of the same MC with the administration of BPC, it decreased to 0.17±0.17, and with the administration of BPC + Eicosavitol it was zero. Likewise, the disorganization of the thymus layers was characterized in the same groups by mean scores of 0 – 2.20±0.37 – 1.60±0.24 – 0.60±0.24, respectively. Similar ratios between the groups were found for many other signs describing the morphology of these organs.

Of the large number of extrapulmonary indices (biochemical, hematological, immunological) of the organism's general status that we used in the experiment, in table 3 we present, again due to length limitations of the paper, only those for which this effect was substantial and statistically significant. Concerning the other indices, it should be only stressed that in none of them did BPC and/or Eicosavitol cause any negative changes even where their beneficial effect was not apparent.

Table 4 shows group-average data on the percentage ratios between various cells which were counted (up to the total number of 500) in Pappenheim's stained smears of femur bone marrow (note that this investigation was not performed for the group of rats that received BPC and Eicosavitol without administration of MC).

The results of estimating MC's genotoxicity by the methods used for testing DNA damage and repair are given in tables 5, 6 and 7.

Discussion

The recruitment of phagocytizing cells in the lower airways manifesting as increased cell numbers in BALF is a typical reaction to the deposition of dust particles therein, and both total cell numbers and shifts in these towards polymorphonuclear (mainly neutrophile) leucocytes are the more marked, the higher the cytotoxicity of particles to alveolar macrophages (15, 17, 27, 28). This dependence on the number of destroyed AMs was experimentally modelled by intratracheal administration of products of preliminarily destroyed peritoneal macrophages obtained (without pre-incubation of these cells with any particles) by repeated freezing/thawing or ultrasounding, and particularly administration of lipids extracted from these products (15).

The neutrophilic response of the lower airways to the deposition of dust particles is quite often described as "inflammation", that is, as a pathological phenomenon; however, there are fairly strong reasons for considering it as a physiological mecha-

Table 3 - Some indices of the general status of rat organisms 6 months after intratracheal injection of monazite concentrate suspension ($X \pm S_x$)

Index	Exposure to:					
	-	Monazite	Monazite+ BPC	Monazite+ BPC+ Eicosavitol	Monazite+ Eicosavitol	BPC+ Eicosavitol
SDH, number of formasan granules in 50 lymphocytes of peripheral blood	901±36	736±22*	982±31•	876±49•	881±26•	910±17
Catalase in blood serum, μCat/L	18,76±1,92	13.47±1.44*	14.43±2.41	14.43±1.44	18.76±1.92	17.79±1.44
Malonyldialdehyde in blood serum, μmol/L	4.07±0.36	5.04±0.19*	4.43±0.23	4.52±0.37	4.88±0.18	4.68±0.13
Segmented neutrophils, %	34.50±3.24	26.20±2.28	31.71±2.84	28.8±0.67	27.00±2.42	28.07±1.83
Band neutrophils, %	2.20±0.39	0.80±0.20*	1.71±0.48	1.60±0.37	1.70±0.41	1.87±0.44

* means the statistical significance of the difference compared to the control group;
• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

Table 4 - Cellular composition of bone marrow 6 months after intratracheal injection of monazite concentrate suspension, %, ($x \pm S_x$)

Index	Exposure to			
	-	Monazite	Monazite+BPC	Monazite+ BPC+Eicosavitol
Erythroid lineage				
Erythroblasts	1.16±0.17	1.86±0.19*	1.67±0.32	1.67±0.32
Pronormoblasts	3.71±0.40	5.03±0.55	6.25±0.88*	5.60±0.64*
Normoblasts				
basophilic	10.78±0.92	18.27±1.67*	14.06±2.03	13.15±1.59•
polychromatic	7.79±0.47	6.46±0.83	8.42±0.65	8.63±1.01
oxyphilic	9.66±0.79	7.70±0.65	7.77±1.36	6.57±0.55*
Erythroid cell mitoses	0.62±0.15	0.76±0.15	1.36±0.34	1.05±0.15
Total cell number of erythroid lineage	33.71±1.35	40.09±1.88*	39.53±2.76	36.68±2.51
Erythroblast maturity index (haemoglobin-containing cells versus all erythroblastic cells)	0.52±0.03	0.36±0.02*	0.40±0.04*	0.43±0.03*
Lymphoid lineage				
Lymphoblasts	1.20±0.19	0.65±0.08*	0.74±0.17	0.78±0.17
Prolymphocytes	2.22±0.31	0.94±0.17*	1.04±0.16*	1.60±0.13•
Lymphocytes	4.31±0.45	2.36±0.27*	2.74±0.48*	3.41±0.47
Plasma cells	1.29±0.24	1.65±0.25	1.68±0.44	1.87±0.33
Total cell number of lymphoid lineage	9.03±0.94	5.60±0.59*	6.20±0.51*	7.66±0.65•
Myeloid lineage				
Myeloblasts	1.84±0.27	2.32±0.49	2.92±0.38	2.22±0.21
Promyelocytes	3.52±0.44	3.17±0.53	3.76±0.26	3.35±0.39
Neutrophilic granulocytes				
myelocytes	3.54±0.47	4.11± 0.51	3.54±0.44	3.71±0.53
meta myelocytes	2.43±0.36	2.93±0.51	3.61±0.51	3.27±0.57
band cells	6.78±0.48	5.29±0.53*	4.94±0.59*	5.11±0.60*
segmented cells	24.46±0.69	19.86±1.85*	19.80±1.83*	23.48±1.31
Eosinophilic granulocytes				
myelocytes	1.42±0.29	1.18±0.32	0.73±0.11*	0.89±0.23
meta myelocytes	2.56±0.27	2.29±0.32	2.39±0.43	3.13±0.37
band and segmented cells	8.83±0.48	10.45±1.82	8.54±1.56	8.28±1.33
Myeloid cell mitoses	0.72±0.07	0.41±0.08*	0.53±0.10	0.78±0.11•
Total cell number of granulocytic lineage	56.11±0.94	52.01±1.54*	50.77±2.99	54.20±2.31
Neutrophil maturity index (band cells+segmented cells versus all neutrophilic cells)	0.84±0.02	0.78±0.02*	0.78±0.03*	0.81±0.02
Monocytes	0.41±0.09	0.37±0.07	0.45±0.06	0.56±0.10
Megakaryocytes	0.80±0.13	1.08±0.19	1.08±0.27	0.95±0.20
Mast cells	0.37±0.09	0.27±0.11	0.22±0.04	0.40±0.17
Leukoblastic elements versus erythroblastic elements	1.96±0.14	1.47±0.12*	1.51±0.19	1.79±0.17

* means the statistical significance of the difference compared to the control group;
• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

Table 5 - Distribution of DNA damage by comet class (as %, $X \pm S_x$) in monocytes of rat blood 6 months after intratracheal injection of monazite concentrate suspension

Comet class[1]	Exposure to:					
	-	Monazite	Monazite+ BPC	Monazite+BPC+ +Eicosavitol	Monazite+ Eicosavitol	BPC+Eicosavitol
C1	50.30±5.34	0.36±1.35*	0.14±0.76*	14.14±2.69*•	8.33±2.61*•	54.38±6.35•
C2	30.40±4.13	8.64±4.13*	14.00±3.83*	27.86±2.69•	24.50±3.46•	30.54±4.37•
C3	9.50±4.83	27.45±4.68*	29.86±4.23*	29.14±4.07*	42.25±2.97*•	14.69±2.87•
C4	9.50±6.06	35.09±4.14*	28.57±3.02*•	28.57±2.79*•	16.50±4.13•	0.46±1.93*•
C5	0.30±1.35	28.45±4.85*	28.43±3.02*	0.29±0.98•	8.42±3.13*•	0•

[1] C1 - practically undamaged cells; C2 - cells with low DNA damage, C3 - medium level of DNA damage, C4 - high level of DNA damage, C5 - completely damaged cells

* means the statistical significance of the difference compared to the control group;

• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

Table 6 - Distribution of DNA damage by comet class (as %, $X \pm S_x$) in lymphocytes of rat blood 6 months after intratracheal injection of monazite concentrate suspension

Comet class[1]	Exposure to:					
	-	Monazite	Monazite+ BPC	Monazite+BPC+ +Eicosavitol	Monazite+ Eicosavitol	BPC+Eicosavitol
C1	50.20±3.10	9.09±1.89*	13.71±2.51*•	14.14±2.69*•	9.41±4.13*	61.69±4.11*•
C2	29.50±4.55	9.27±1.57*	13.71±2.23*•	44.14±4.23*•	33.33±5.21•	30.85±4.23•
C3	19.70±4.43	28.27±2.54*	28.86±3.15*	27.43±4.14*	32.75±3.63*	7.00±3.56*•
C4	0.20±0.84	35.73±3.80*	29.43±5.01*	13.86±2.69*•	17.08±6.95*•	0.31±1.26•
C5	0.40±1.03	17.64±2.87*	14.29±3.95*	0.42±1.07•	7.41±4.78*•	0.15±0.75•

[1] C1 - practically undamaged cells; C2 - cells with low DNA damage, C3 - medium level of DNA damage, C4 - high level of DNA damage, C5 - completely damaged cells

* means the statistical significance of the difference compared to the control group;

• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

Table 7 - Coefficient of DNA fragmentation[1] in lymphocytes of rat blood 6 months after intratracheal injection of monazite concentrate suspension ($X \pm S_x$)

-	Exposure to:				
	Monazite	Monazite + BPC	Monazite + BPC + Eicosavitol	Monazite + Eicosavitol	Control for BPC+ Eicosavitol
0.083308±0.005947	0.095828±0.030529	0.065873±0.000507*	0.034645±0.000774*•	0.054095±0.019407*•	0.058816±0.002708*•

1) Ratio of «tail» radioactivity to «nucleus» radioactivity with electrophoresis of amplified DNA tagged with tritium on agarose gel (AFLP analysis)

* means the statistical significance of the difference compared to the control group;

• compared to the "Monazite" group ($P<0.05$ by Student's t-test)

nism of partial compensation for the damage caused by particles cytotoxic to the alveolar macrophage, the main effector of pulmonary clearance. A mathematical multicompartmental model of pulmonary clearance which includes just this compensatory mechanism predicts very well the re-

tention of dust with varying degrees of cytotoxicity in the lungs at inhalation and the decrease in this retention under the effect of such potent protector of the macrophage against the cytotoxicity of particles as glutamate (16, 17).

In any case, when evaluating the data of table 1 it is important to note two circumstances. Firstly, given our sufficiently broad experience in investigating BALF in intratracheal or inhalation dust exposures, we have observed such a sharp increase in the total cell numbers and NL/AM ratio only for one, namely exposure to highly cytotoxic quartz DQ_{12} (14, 19). Secondly, with regard to MC action, both of these shifts were substantially limited against the background of preliminary month-long pre-treatment of rats with bioprotectors, particularly with combinations of BPC+Eicosavitol. In the light of the above data and reasoning, this attenuation of the MC effects studied allows us to presume a substantial increase in the resistance of the macrophage to the cytotoxicity of MC particles. And because such mechanisms are not qualitatively specific to any certain type of dust, it is not surprising that the bioprotectors tested produced some reduction in the NL/AM ratio in control rats as well, which inevitably inhaled some dust particles from the indoor air in conventional housing conditions.

An increase in the lipid content of the lung tissue is typical of experimental pneumoconioses and, as was shown a long time ago, correlates with the fibrogenicity of dust (13). There are experimental grounds for linking this effect to the enhancement of the lipopectic function of the lung macrophage activated either by dust particles or by products of decay of other lung macrophages under the effect of these particles (18, 36). The second mechanism of activation allows us to explain why the accumulation of lipids in the lung is usually reduced when the cytotoxic action is attenuated, for example, in experimental silicosis under the effect of glutamate (18, 21, 28). Similarly, the attenuation of this quasi-lipogenic effect of MC particles under the influence of the bioprotectors tested (table 2) may be confronted in our experiment with the attenuation of the above shifts in the BALF cell composition, providing indirect evidence of increased resistance of lung macrophages to the cytotoxic action of this dust.

Our experience of quantitative estimation of pneumoconiotic changes developing in rats with intratracheal administration of a broad range of mineral dusts allows us to regard the fibrogenic action of MC on the lung tissue (characterized by an increase of 1.7 in the hydrooxyproline content 6 months after intratracheal injection in comparison with the control index) as more than moderate considering the fact that we deal with a dust containing neither silica nor silicates. For comparison, one should consider the data of experiments in which rats were injected intratracheally with the same dose of ground quartzite rock containing 92% of quartz or of ground chrysotile asbestos (27). In the first case, the hydroxyproline content of the lungs was 2.3 times higher and in the second case only 1.6 times higher than in the rats of corresponding control groups. In a later experiment performed by the same laboratory, 50 mg of chrysotile asbestos caused almost a doubling of this index in 6 months (38). Thus, the experimentally estimated fibrogenicity of MC dust ranks among quantities inherent in two well studied mineral dusts which are associated with a doubtless risk of developing progressive forms of pneumoconiosis by humans.

We can only compare this with the above discussed high cytotoxicity of MC particles because it is just dust the damage to a proportion of the lung macrophages and the activation (directly, that is by dust particles or, in our opinion, mainly secondarily, i.e. by macrophage decay products) of other, still viable macrophages that leads via many mechanisms, both common to all types of dust and more specific ones (for example, for silica), to pneuomoconiotic fibrogenesis, let alone the fact that this damage contributes to the retention of cytotoxic dust in the lung tissue and tracheo-bronchial lymph nodes (14, 16-19, 35-37) It is exactly because of the complexity and diversity of these mechanisms that there is no one-to-one correspondence between the relative cytotoxicity and the relative fibrogenicity of mineral particles of different composition. However, it may be assumed, on the whole, that high cytotoxicity is a prerequisite for high fibrogenicity.

As for the high cytotoxicity of MC particles itself, we cannot rule out the possibility that this

may be inherent in some low soluble compounds of rare earth elements. For example, the oxides La_2O_3, Ce_2O_3 and Nd_2O_3 are toxic to rat alveolar macrophage *in vitro*, and Nd_2O_3 is even more toxic than the soluble salt $NdCl_3$ (31). The cytotoxicity of superconducting particles of $YBa_2Cu_3O_{6-7}$ to a culture of bovine alveolar macrophages is almost the same as the especially high cytotoxicity of quartz DQ_{12} (41). At the same time, it can be assumed that both the cytotoxicity of MC and its fibrogenicity are at least partly connected with the radiotoxicity of this substance.

The issue of dependence of the risk of pneumoconiosis development on the effect of ionizing radiations produced by the chemical composition of the dust itself or accompanying it has been rarely discussed in the research literature of the last few decades, whereas once it was the subject of numerous debates. This discussion was initiated at the end of the 1940's based on the hypothesis suggesting a major role of natural radioactivity of dust in the aetiology and pathogenesis of silicosis (8). Although that hypothesis was never confirmed, some experimental studies have shown that the fibrogenic action of dust, given an equal or similar quartz content, may be enhanced due to both the presence of radioactive elements in the dust-forming minerals (7, 12, 42) and the combined exposure to quartz and high concentrations of radon in the air (26). In the study carried out by the first author of this paper with his colleagues, use was made of a concentrate of pyrochlore, a niobium-containing mineral, with a low silicon dioxide content and with a small admixture of uranium and thorium oxides (12). The results of that study allowed the authors to assume that for a dust of mixed composition containing no more than 5% quartz, the alpha activity in the order of 10^{-13} Curie/mg does produce a certain enhancement of fibrogenicity. Moreover, they found that at this level a 3- to 5-fold increase in the radioactivity of dust is, apparently, the minimum for which a further marked increase in fibrogenicity should be expected while differences in the quartz content play a considerably more important role.

That study was carried out in the early 1960's when BALF cytological indices were not yet used in our laboratory as a criterion of the cytotoxic effects of mineral particles, but for similar purposes we often used an intraperitoneal test. This test showed that pyrochlore with the above alpha activity caused explicit signs of degeneration of peritoneal macrophages with engulfed dust particles in 11.2% of such cells 24 hours after intraperitoneal administration and in 19.4% after 48 hours, whereas in a similar test with a dust collected at the same production facility but with a radioactivity that was lower by one order of magnitude, only in 4.0% and 3.8%, respectively (12). At the same time, in macrophages with no visible dust particles there were very few degenerated cells. Thus, virtually quartzless dust particles, free of rare earths as well, were indeed highly cytotoxic to the macrophage at the total alpha activity of the same order of magnitude as that featured by the MC dust that we investigated here.

However, whatever the explanation of the cytotoxic, fibrogenic or quasi-lipogenic effect of MC particles on rat lungs, all these effects could be attenuated to a greater or lesser extent by the bioprotectors tested by us: BPC was more effective for the majority of indices than Eicosavitol, but a combination of BPC with Eicosavitol produced the most marked protective effect for some of the effects. As is pointed out in the Introduction, Eicosavitol was included in the bioprotectors owing, first of all, to its positive influence on protection against DNA damage and on DNA repair, which was confirmed experimentally (see below). It is possible that the protection of macrophage, as well as that of lymphoid immunocompetent cells participating in the pathogenesis of pneumoconiosis, against DNA damage somehow enhances the resistance of these cells to the effects of cytotoxic and fibrogenic dust load. In such a case, it is the combination of Eicosavitol with anticytotoxic and antifibrogenic bioprotectors operating on the basis of other mechanisms (briefly considered in the Introduction) that provides their synergy. This hypothesis seems to us to be more consistent than a simple explanation of the observed protective synergy as provided by the additional action of antioxidant vitamins contained in small amounts in Eicosavitol in addition to the antioxidants con-

tained in basic BPC. The influence of Eicosavitol's PUFA on the pneumoconiosis development mechanisms that are linked to lipid metabolism and lipoperoxidation should be a potentiating rather than mitigating influence. Indeed, earlier it was found that, in rats injected intratracheally with quartz DQ12, both lipoperoxidation and collagen synthesis were *enhanced* markedly by the background action of "Linaetholum", a preparation obtained from linseed oil consisting of a mixture of ethylic ethers of alpha-linolenic (about 57%), linoleic (about 15%) and oleic (about 15%) acids and only 7-11 % of saturated fatty acids. Thus, this preparation, too, contains PUFA of the omega-3 group, even in larger amounts than Eicosavitol. In this experiment, Linaetholum was administered weekly into the stomach of rats through a tube at a dose of 10 g per kg of body weight (18).

That the bioprotectors produced a beneficial effect just on fibrogenesis and "lipogenesis" pathogenetically connected with the development of monazite pneumoconiosis, is evidenced by the absence of any influence of the same combination of BPC with Eicosavitol on these processes in the lungs of rats which were not given MC.

The beneficial effect of the bioprotectors on pulmonary MC clearance appeared to be less marked than one might have expected against such a marked anticytotoxic effect. This apparent contradiction is easily explained by the fact that a massive momentary load of a large dose of any dust hinders access for phagocitizing cells to particles retained in the lung tissue, thus limiting the possible efficacy of even a protected phagocytic mechanism of pulmonary clearance. It is surprising that in these extreme conditions this efficacy was nevertheless somewhat increased by the bio-protection of this mechanism. The action of a BPC of a slightly different composition was found to provide a similar beneficial effect (and also statistically significant) in relation to a similar massive load of chrysotile asbestos dust (38). It is important to emphasize, however, that the ultimate attenuation of pneumoconiotic changes was apparently caused not only by a decrease in the MC mass retained in the lungs but also by the inhibition of the complex pathogenetic mechanisms that underlie the action of dust on the organism. This is convincingly evidenced by the reduction in the computed index of increment in the hydroxyproline content per unit residual mass of dust, with this index revealing the most marked (1.6 times) and statistically significant decrease in response to the action of the BPC+Eicosavitol combination. It should be noted that a substantial decrease in a similar index, along with a reduction in the effective mass of retained dust under the influence of a cytoprotective BPC, was also observed in the experiment with chrysotile asbestos (38).

The functional indices shown in table 3 provide evidence that the toxic action of MC is not exclusively local (on the lungs). Thus, the influence of MC was observed to be associated with a general reduction in the level of bioenergy metabolism, manifesting in a statistically significant suppression of the activity of succinate dehydrogenase (SDH) of blood lymphocytes estimated cytochemically. It should be noted that we had often observed a reduction in this index earlier under the influence of various toxic exposures of rats, and this reduction was attenuated or completely absent if these exposures were accompanied by the action of various bioprotectors; here again we can refer to the example of the recent experiment with chrysotile asbestos (38). Similarly, in an experiment with MC in rats receiving BPC, Eicosavitol or their combination, this effect was totally absent; on the contrary, SDH activity increased.

An indicator of the suppressing action of MC on the organism's antioxidant system may found in the reduction in the level of catalase in the blood serum, this reduction again being considerably less marked or unobservable against the background of bioprotector administration. The increase in the malonyldialdehyde (MDA) content in the group of rats that received only MC may indicate both the suppression of the antioxidant system and point to primary enhancement of the lipid peroxidation processes. The mechanisms of this enhancement are various, but one of them may be the activation of viable macrophages by products of the decay of a proportion of macrophages due to the action of cytotoxic particles as was shown earlier in experiments on cell cultures (18, 37). The action of bio-

protectors again resulted in this shift becoming smaller in relation to the control level and loss of statistical significance.

The same table 3 shows some reduction in the percentage of segmented neutrophiles in the blood under the influence of MC. For band neutrophiles this reduction was statistically significant but was not so apparent and not significant when MC was followed by bioprotector administration. This may point indirectly to the suppression of granulocytopoiesis under the influence of MC, presumably connected with internal alpha irradiation of bone marrow cells, and to the protection against it due to the action of bioprotectors.

Well consistent with this hypothesis is the analysis of the cell composition of rat bone marrow (table 4). Specifically, the ratio of leukoblastic elements to erythroblastic ones under the influence of MC decreased significantly with statistical signficance, while against the background of BPC or BPC+Eicosavitol it also decreased as compared to the control index but not so significantly and to a lesser extent than without bioprotectors. A statistically significant reduction was also observed in the percentage of segmented and band cells and in the neutrophyl maturity index. BPC alone practically did not influence this reduction but in response to the action of the BPC+Eicosavitol combination both indices normalized. We observed a substantial decrease in the percentage of granulocytic hemopoietic cells at various stages of mitosis, and this effect was significantly attenuated in response to effect of the BPC+Eicosavitol combination.

Intergroup differences were similar for bone marrow monocytes as well, though they were not significant statistically. They were similar also for lymphocytic cells, the total percentage of which was reduced with statistical significance in response to MC alone and even to MC against the background of BPC. However, in the case of MC against the background of the BPC+Eicosavitol combination, this reduction in comparison with the control index was less marked and not significant statistically. In this group, the total count of lymhocytic cells was higher with statistical significance than in the case of MC alone. The inhibiting action of MC was obvious at all stages of lymphocytopoiesis (lymphoblast-prolymphocyte-lymphocyte), with BPC and particularly BPC+Eicosavitol producing a more or less marked attenuation of this action at all stages.

Thus, on the whole, MC demonstrated sure toxicity with regard to both granulo- and agranulopoiesis.

An increase in the percentage of erythroid cells observed in all groups receiving MC was, basically, relative (that is, caused by a reduction in the number of leukopoetic cells), however, within this lineage there was a marked and statistically significant decrease in the proportion of haemoglobin-containing cells, pointing to slower maturing of erythroblasts: this adverse effect of MC was less pronounced in the groups receiving bioprotectors,.

As is well-known, dust particles retained in the lung tissue can penetrate into the systemic blood stream through the lymph and in this way be retained secondarily in any organ having cells of the reticuloendothelial system. It can be assumed that the changes in hematopoiesis observed by us are connected with a direct effect of ionizing radiations from MC particles retained in bone marrow tissue and possibly with the toxic effect of the rare earth elements slowly passing into the tissue from these particles. It can be expected that these influences lead not only to the above inhibition of the mitotic activity of hematopoietic cells but also to damage to nuclear DNA which should manifest as the appearance of leucocytes with signs of such damage in the blood.

As can be seen from tables 5, 6 and 7, the chronic effect of MC injected into the lungs did lead to enhanced fragmentation of the DNA not only in blood monocytes (which, being cells of the system of mononuclear phagocytes, could have been exposed while in the bone marrow to MC particles not only from the microenvironment but also as a result of their engulfment) but also in blood lymphocytes. This effect was attenuated by the bioprotectors tested, particularly by the combination of BPC and Eicosavitol. At the same time, BPC and Eicosavitol do not produce any genotoxic action of their own; on the contrary, when used in combination they provided some attenuation of the background level of DNA fragmentation.

Conclusion

Intratracheal administration of monazite particle suspension to rats induced in their organism changes manifesting cytotoxicity, fibrogenicity, systemic toxicity and genotoxicity of this substance, and these adverse properties are due most probably to the presence in its composition of not only rare earth elements but also of natural radioisotopes of the thorium and, to a lesser degree, uranium families. A bio-protective complex (BPC) comprised of glutamate, an iodine preparation, methionine, and a polyvitamin-polymineral composition attenuated all these adverse effects of monazite. Although a similar protective potential of "Eicosavitol" (a bioactive food supplement rich in polyunsaturated fatty acid, predominantly of the omega 3-group) against almost all effects of monazite is lower as compared with that of the above BPC, combining this BPC with "Eicosavitol" proved, as a rule, to give the greatest protective effect. We recommended this combination for periodic administration (once or twice a year) to workers dealing with monazite concentrates in the form of preventive programmes.

No potential conflict of interest relevant to this article was reported

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