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MATERIALS OF THE FINAL REPORTS ON THE JOINT SOVIET-AMERICAN EXPERIMENT ON THE KOSMOS-936 BIOSATELLITE, MOSCOW, 1978

by

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Biological Research

The Kosmos-936 biological satellite functioned in Earth orbit for 18.5 days from August 3 through 22, 1977. The main purpose of the research done during the flight of the biosatellite was a further deep study of the effect of factors of space flight, mainly weightlessness, on living organisms.

In this space experiment, as in the preceding, foreign specialists including those from the USA participated with the Soviet researchers. During the flight, general biological, physiological, radiobiological and radiation-physical studies were carried out.

In the article, results are presented only of the biology experiments done on insects, higher and lower vegetation using genetics, cytologic, biochemical, analytical, morphological and ultranorphological methods.

Studies on fruit flies (Drosophila melanogaster) were carried out with American scientists on the Oregon-R strain according to the program agreed on. The fruit fly specimens of this strain were obtained from the Ames Scientific Research Center NASA, USA, on December 18, 1976. From this time until the beginning of the flight experiment (August 1977) the strain was kept in a laboratory insectarium at 25°C. Specialists from the USA neaded by J. Miguel expended their efforts in studying the somatic structures using different anatomical, morphological, biochemical and behavioral methods.

The general purpose of their research included studying the effect of weightlessness on the processes of aging or, in other words, in determining the rate of the processes of life activity in conditions of weightlessness.

The Oregon-R strain was selected for the experiment in space upon request of the American specialists inasmuch as

*Numbers in the margin indicate pagination in the foreign text.

all of the background laboratory data for many years had been obtained on species of this strain. This suited us because this strain is characterized by high genetic stability, low frequency of the basic types of spontaneous mutations, and good previous studies. In particular, using this strain, V. N. Timofeyev-Resovskiy pointed out the role of differences in frequency and direction of mutation of separate chromosome loci in different populations for divergent evolution.

The goal of the Soviet section of the experiment consisted of studying the effect of weightlessness on hereditary structures; and for this they had to determine the frequency of chromosome and gene mutations occurring.

On board the biosatellite cultures were placed with the fruit flies themselves at age 24 days (average age), 3-7 days (young), cultures with virgin females and cultures with eggs placed in a nutritive medium 3 days before launch. The cultures were contained in glass test tubes with diameter 20 mm and height 110 mm with a standard nutritive medium with a somewhat increased content of agar agar (8% instead of the usual 6%) in order to avoid spilling the nutritive medium. Moreover, in order to prevent the appearance of mold, 0.8 ml of [word illegible] acid in 100 ml of water was introduced into the nutritive medium and subjected to ultraviolet for 30 minutes. External examination of the flies in the landing area indicated that the material was in good condition; no mold was noted in the culture, only individual flies were dead; in these no anomalies in the development of the specimens were observed. In those cultures which were intended for obtaining progeny, the density of the population was maximum, that is, 80-100 specimens. The average weight of the males was equal to 6.8 ± 0.1 mg, the weight of the females, 7.8 ± 0.1 mg. The dimensions and weight of the flies grown in conditions of weightlessness did not differ from the weight of the flies in the laboratory control. A sample was taken in the landing area and crossbreeding of the fruit flies was carried out in accordance with the program (crossbreeding of males and females from specimens of the test strain of the opposite sex).

/5

For calculation of deletion: crossbreeding was set up with females having special X-chromosomes with genes labeled yellow, white, and forked. No "exceptional" females with "darkened" genes were obtained. 2575 [numbers difficult to read in the original text] test and 1305 control flies were analyzed.

For calculating the frequency of microdeletions and visible mutations in [word illegible] loci of male test and control groups crossbred with virgin females of the test strain

which have the following gene markers in the female chromosome, yellow, white, cut, forked, Bar (y, w, ct, f, B). The Bar gene was used for control of the "virginity" of the females. The results of studies using [word illegible] gene markers are presented in table 64; they attest to the fact that frequency of microdeletion or mutation in the specific loci do not increase under weightlessness. The frequency of mutation of all loci both in the test and in the control exceeds 1%. This indicates that the X chromosome in specimens of the Oregon-[word illegible] strain in a metastable state.

Table 1

Number of specimens considered **y** $(\% \pm m)$ $(\% \pm m)$ $ct (\% \pm m)$ $f(\% \pm m)$ Test 2613 35 I,33±0,22 37 I,4I±0,23 40 I,53±0,24 44 I,69±0,25 Control 503 6 I,I9±0,48 6 I,I9±0,48 7 I,39±0,52 7 I,39±0,52 [Commas in the tabulated material are equivalent to decimal points.]

The frequency of recessive fatal mutations in the female chromosome was calculated according to the Meller-5 method; when analyzing 672 chromosomes in females from the flight material, 523 chromosomes of the synchronous experiment and 1037 chromosomes of the laboratory control, no mutations were observed. The data on frequency of recessive fatal mutations of males is presented in table 2.

As is apparent from table 2, the statistically verified data of relative frequency of fatalities in the sperm and spermatids are absent. However, the frequency of fatal mutations in the spermatagonia is fairly high. During radiation tests, as is known, sometimes one does not obtain an "output" of fatalities in the spermatagonia higher than 1%, inasmuch as /7 when increasing the dose of radiation the species are sterilized. The 11 fatalities recorded by us in this experiment, in all probability, are a cluster at the beginning of which a single mutation event took place.

	Number Analyzed	Number of fatal	<u>-</u> <u>/6</u> % <u>+</u>
Mature sperm	642	3	0,46 <u>+</u> 0,27
Spermatatids	462	3	0,64 ± 0,36
Retarded spermata- gonia	468	II	2,35 <u>+</u> 0.70

[Commas in the tabulated material are equivalent to decimal points]

Table 2

Upon completion of the experiment, the American specialists <u>/7</u> were given 130 flies of each group (that is, imago, hatched during flight, imago, placed on board at young and average age; correspondingly, the same number of flies was sent from the synchronous experiment). The material was delivered to Ames Scientific Research Center (California USA) 11 days after landing of the biosatellite, on September 2, 1978. Consequently, the age of the flies at the beginning of the study was for the imago hatched in weightlessness, 31-35 days, and for those placed on board at young and average age, 36-40 and 61-65 days, respectively. The age of the flies of the first group was counted from the moment of Lalaying the eggs; the age of the two other groups, from the moment of hatching.

As was noted above, the American scientists were interested in behavior, survival rate, characteristics of morphology, anatomy and biochemical processes in the fruit flies. The set of data obtained, in the opinion of the American colleagues, must have characterized the rate and intensity of the aging process. The results obtained were summarized in the form of a report compiled by J. Miguel, D. E. Filpott, R. Binnard and S. Turnbill and transmitted to the USSR in accordance with the existing agreement on carrying out the experiment in January, 1978.

First of all, from the report it follows that the embryonic development of the fruit flies in weightlessness does not differ from that occurring on Earth. In the microphotographs obtained

using a scanning electron microscope, it is apparent that all of the organs of the eye studied, the balancers, legs, etc., in the flies hatched in flight hardly differ from the standard.

When planning the experiment, we asked our Americal colleagues to devote attention to the fine structure of the muscles of the fruit flies, which provide for locomotion. The electronic microphotographs obtained indicate that when the fruit fly develops in weightlessness changes do not occur in the ultrastructure of the muscle tissue. In particular, in muscular cells of the test materials, gigantic mitochondria were observed typical for the wing muscles of insects. In the fruit flies put on board at the imago stage, mechanical damage to the wings was detected which, in the opinion of J. Miguel, showed a breakdown in the capability to control flight in a state of weightlessness. However, this effect was not observed in the flies who developed from eggs in conditions of weightlessness.

The viability of the flies was characterized by the value of negative geotaxis and the capability to mate. The intensity of the negative geotropic reaction was sharply decreased in flies placed in flight in the imago stage in comparison with the control. The intensity of sexual behavior was noticeably decreased in them both in the relative total number of matings and in the latent period during mating.

In the flies of all groups found on board, the average duration of life was decreased as described by the curves of the survival rate. Thus, the data of relative viability of the fruit flies indicates that the aging processes are accelerated in weightlessness conditions. This fact corresponds to J. Miguel's initial hypothesis. Earlier in the laboratory, the author had indicated that the return of the fruit fly imago /9 to horizontal clinostatus causes a strengthening in locomotion and a tendency to concentrate the flies in the region of the axis of rotation. Similar behavior in fruit flies was accompanied by a decrease in average length of life by approximately 13% in comparison with the lifetimes of flies returned to vertical clinostatus. As the American authors propose, the cause for speeding up the aging processes is nonspecific stress caused by weightlessness as a result of which general activity increases, including motor activity, and consequently, the requirement In our opinion, one should not exclude the idea for oxygen. that acceleration of the aging process involves the effect of increased temperatures on the imago (up to 30° and higher), because all of the effects observed, decrease in the length of life, decrease in intensity of negative geotaxis and particularly the intensity of sexual behavior could well occur due to the effect of increased temperature on the flies.

However, the data which attest to the acceleration of aging processes obtained by the American specialists at the present time using behavior tests have not been subjected to the results of biochemical analysis. These studies are not yet finished.

Cytogenetic studies in this complex experiment were carried out on crepis seedlings (Crepis capillaris).

The purpose of the experiment was to study the possible effect of weightlessness on the spontaneous mutation process in the cells of vegetation seedlings grown from air dried seeds on board the Kosmos-936 biosatellite. The seedlings were grown in a special "biofiksator-I" instrument whose design made it possible to automatically moisten the seed and fix the material. The instrument consisted of ten vegetation chambers with a volume of about 60 $\rm cm^3$ each. The chambers had three containers: two with a volume of 3 ml for water /10 and one with a volume of 5 ml for the fixing fluid. Feeding the water and fixer into the vegetation chambers was done automatically according to a prescribed cyclogram at determined time intervals, namely: the first and tenth days water was supplied in all chambers simultaneously; on the second, fourth, sixth, seventh and tenth, the sixteenth and nineteenth days, the fixer was fed correspondingly into the first, second, third, fourth, fifth, sixth, seventh, eighth and ninth chambers. The seedlings in the tenth chamber were returned to Earth unfixed. But because the flight lasted 18.5 days instead of the planned 20, the material in the ninth chamber was fixed in the satellite landing area. This cyclogram was applied in two other similar instruments where experiments were carried out with higher vegetation and lower fungi; the results are described below. The temperature inside the instruments for the entire flight was constant (25 ± 0.5°C). The synchronous control experiment was carried out on Earth in a mock-up of the biosatellite with all other conditions equal. In ali, 2,000 seeds were selected for the experiment, (1,000 each for the test and control). In each chamber, 100 air dried seeds were placed on filter paper. Part of the seeds for five days before flight were treated with an 0.3% solution of colchicine for obtaining a division in the seedling cells which was maintained at the metaphase stage with chromosomes typical for this stage in the K state. The nontreated part of the seeds were intended for studying at the anaphase stage. As is known, the population of Crepis seeds is divided into 3 fractions according to their growth period: early germination seeds every 20 hours after moistening. average, every 25 hours and late, every 30 hours and more. The conditions for carrying out such an experiment did not permit sing- /11 ling out one of these fractions; however, thanks to the use

of colchicine we were able to determine the time periods for the first and subsequent mytoses.

hours after landing of the biosatellite. an 3.5 inspection of the material was made in the field laboratory, a count of germination of the seeds and morphometry. Then. the material was transferred to a fresh fixer and sent to a permanent laboratory for cytogenetic study. Temporary "pressed" preparations colored with acetocarmine were prepared from the rootlets and whole seedlings with the cotyledons. In the course of analysis, the possibility was discovered of calculating damage to the chromosome in the cells of the inserted meristem of the primary procambium. Moreover, in the meristem cells, the mitotic index was taken into account. We did not have the problem of tracing the formation of cellular differentiation at the early stages of development of the seedlings. However, the precision of the observed cytologic pictures made it possible for us to record this process. Of 1,000 seeds in flight, 104 grew whereas in the control 542 grew which is a noticeable excess of the control over the test. The length of the seedlings at the end of the experiment, that is, on the nineteenth day, in the test was on the average 4.5 cm and in the control, 2.5 cm. In the seedlings grown in weightlessness, the roots and shoots went in different directions and made up a straight axis whereas in the terrestrial experiment, the shoots with the cotyledons bent upward and the rootlets spread along the surface. These data attest to the manifestation by the sprouts of a negative geotropic reaction in the ground control experiment and to the absence of this in weightlessness; this confirms the results of preceding experiments on the Kosmos-782 biosatellite in 1975. Moreover, one can suppose that the greater length of the seedlings grown in weightless-/12 ness is due to the more intense stretching of the cells, the quantity of divided cells in weightlessness not only was not increased but was somewhat decreased in comparison with the control. The shape of cells which were grown in weightlessness was circular and differed from the ordinary. Apparently, outside the field of gravity, the mechanical pressure of the cells on each other decreases considerably. In judging the possible causes of such a type of difference in configuration and dimensions of the cells, we will turn to a consideration of the results of electron microscopic analysis.

In the first half of the experimental period, the rate of growth of the seedlings was much larger both in the test and in the control. This principle was observed in another experiment with corn seedlings. In all probability, this is due to the use of oxygen. The data obtained during cytologic inspection of the meristem cells of the root are presented in table 3.

Fixa- No. tion of root- lets con- sid- ered	No. of	No. of cel considere	ls d	M±m%	No. of root-	No. of cel considere	ls d	
	root- lets con- sid- ered	Stan- Break- dard down		lets con- sid- ered	Stan- Brea dard down	.k- M <u>+</u> m;	%	
				letaphases				
2	5	423	I	0,2 <u>+</u> 0,14	16	645	I	0,15+0,1
		(First Division)		\ ·		(First Division)		
		44	-			·I38	-	_
GINAL PA POOR QU	GE IS	(Second Division)				(Second Division)		
• . •				Anaphase	S		•	
4,5, 6,7, 8.	35	168	I	0,5 <u>+</u> 0,3	38	545	I	0,18 <u>+</u> 0,17

[Commas in the tabulated material are equivalent to decimal points]

Modification of the colchicine method was successfully /12 realized. In the rootlets fixed on the second day, all of the divided metaphase cells were found in K mitosis, that is, they were spiral and twisted under the effect of colchicine and were found in a first or second cellular division. In the test material, neither in the first nor in the second mitoses did one observe an excess of mutations over the spontaneous level; their numerical value consisted, respectively of 0.15% and 0.20%. In the test material, a total of a single microfragment was observed, and in the control -- chromatic deletion of the long arm of the A chromosome. Anaphase analysis made at later fixation time periods showed 0.6% breakdown in the test and 0.5% in the control which indicates the absence of any kind of difference between the test and the control. In the tip (apical) meristem, when examining 172 cells in the control. one chromosome aberration was detected and in the test (98 cells examined) no aberrations were detected. When examining

88 cells of the procambial system of the test materials and 115 cells of the control material, not one cell with aberrations in the chromosome either in the first or second case was recorded.

A calculation of the mytotic index indicated that in the cells of control variants it is higher for all time periods of fixation except the last, that is the statokinetic index (second fixation) (table 4). The differences in all of the

Гa	b	1	е	4
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Mytotic Index in Meristem Cells of Rootlets

		TEST				CONT	ROL			
Fixation	Number of rootlets examined	Number of interphases	Number of divided cells	M ± m%	Fixation	Number of rootlets examined	Number of interphases	Number of divided cells	%	
2 4 5 6 7 8	6 9 5 3 4	4349 4918 5629 2091 2954 2633	679 155 64 1 11 70	I3,5 <u>+</u> 4,4 3,5 <u>+</u> 2,7 I,I <u>+</u> 0,7 0,04 <u>+</u> 0,0I 0,5 <u>+</u> 0,I 2,5 <u>+</u> 0,6	2 4 5 6 7 8	7 9 6 5 3 4	3087 11034 1420 8998 813 2210	8I5 II77 30 68I - 6I	T -	20,8 <u>+</u> 12,4 9,7 <u>+</u> 8,4 2,I <u>+</u> I,7 6,3 <u>+</u> 5,0 - 2,7 <u>+</u> 0,7

[Commas in the tabulated material are equivalent to decimal points]

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cases are unreal as a result of the large changeability of these indices in separate roots. Thus, the mitotic index in the test and in the control were the same statistically speaking.

On board the Kosmos-936 biosatellite, an experiment was carried out for the first time in space biology whose purpose was to study the peculiarities of the formation and development of the ultrastructural organization of vegetation cells.

Meristem cells of corn rootlets (Zea mays) were selected /15 as the object of the study. This experiment was carried out using the Biofiksator-I instrument which we have mentioned above. The main task of the experiment was to discover whether or not the conditions of space flight, basically weightlessness, affect the character, rate of development and internal organization of the cells and their basic components. Moreover, it was proposed using a method of electronic microscopy to confirm or disprove data on changes, shapes and dimensions of cells which develop in weightlessness obtained by anatomical and morphological methods.

Air dried seeds of the Sterling brand of corn, in a quantity of 50 pieces, were selected, disinfected and placed in the ten chambers of the Biofiksator-I (5 in each). The containers for the fixer and the water were filled with 4.5 ml of 2% gluteraldehyde, 6 ml (2 x 3 ml) of water, respectively. As has already been mentioned, there was a common cyclogram for all the instruments. The instrument was disassembled at the landing area for the satellitc an external inspection was made and a morphometric analysis of the material obtained.

As the examination showed, without exception all of the seed had grown in all of the chambers. The commands for supplying the water and the fixer had been given and realized. The material in the ninth chamber was fixed in the landing area (in the tenth chamber, as had been designed in the program, no fixer was used). After a morphometric analysis of the rootlets, they were separated from the seeds, put into containers with a fresh fixer and sent to the permanent laboratory where, simultaneously with control material, obtained in a synchronous experiment, they were prepared for electron microscopic analysis. /16 The three day rootlets were crushed on a cover glass in a drop of fixer, put into 1% 03 04, contrasted with uranyl-acetate and then put into a growing concentration of alcohol from 20% to absolute, holding at each for 30 minutes. The dehydrated material was placed in a mixture of acetone with Epon-812 epoxy resin, held in the pure resin, poured into gelatin capsules and polymerized for three days. Ultrathin sections were obtained

on the ultramicrotome. Photographs were made on the YeM-100 $^{\rm M}$ microscope.

The results of morphometric analysis are presented in figure 1. As is apparent from the drawing, the growth of the roots both in the test and in the control experiments was normal in the first 6-7 days, that is, in chambers 1, 2, 3, and 4. Then, growth was retarded and after 10 days, lengthening of the rootlets and sprouting of shoots (coleoptile) was not noted.

In our opinion, the main reason for this unequal growth was inadequate supply of oxygen (partial hypoxia) as a result of the limited volume of the chamber (about 60 cm³). A similar picture obtained in the synchronous (ground) experiment can be used as confirmation of this, that is, all of the remaining parameters except for temperature were the same in the tests and in the control groups; consequently, the conditions for developing the control and test seedlings was uniform.

An electron microscope anaylsis of the material, obtained in the experiment, was made in cooperation with the Institute of Biological Physics of AN USSR.

The preliminary results obtained at the present time make it possible to establish certain peculiarities of characteristic development of a cell and its components.

Figure 2 shows electronograms of sections of tissue from $\frac{17}{17}$ the growth zones of the rootlets grown in the test and control variation. First of all, from the photographs taken it is apparent that the dimensions of the cells which were grown in weightlessness on board the biosatellite considerably exceed the dimensions of cells grown in the ground experiment. As was pointed out above, this principle was noted during anatomical study of the flight and synchronous material in the Crepis seed-lings by a method of light microscopy.

Electron microscopic analysis confirmed the fact that the cells which form in weightlessness have a more circular shape. These differences apparent when studying the anatomy of tissue of the root meristem of the Crepis and supported by a method of electron microscopy on cells of the corn roots give us the basis for proposing that such an architecture is characteristic for cells which develop in weightlessness and is explained, apparently, by the absence of tension on the cellular wall, that is, in other words it is possible that the growth of cells in conditions of a gravitational field is accompanied by mutual mechanical pressure against each other which determine their shape and dimensions.



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Figure 2. Overall view of cells in the growing zone on the $\frac{/25}{1000}$ tenth day in corn roots. Electronogram Mag. 5000 X.



a) Test/weightlessness



b) Synchronous control on Earth.

However, how does weightlessness affect the internal structure of the basic organella (nuclei, mitochondria) in developed cells? Although the data obtained up to the present time by electron microscopy is preliminary and does not make it possible to draw final conclusions as to the principles of these changes and how important they are for functioning of the cells, nevertheless, the material existing in this arangement of materials (an adequate number of microphotographs) makes it possible to state several hypotheses. First of all, in our cpinion, one must check on the structures responsible for energy provision of the cells. In laboratory studies which we made recently on the effect of the changed force of gravity on /18 oxygen activity of the mitochondria (Tairbekov et al., 1978) a decrease was pointed out in the endogenic breeding of these organelia separated from cells in the seedlings cultivated from corn seed in conditions of continuous a) clinostatic separation (hypogravitation) and b) centrifugal separation (hypergravitation), in comparison with the activity of mitochondria of cellsor ordinary seedlings. However, their capability to oxidize malate, malonate) < additional substrata from within (NADN , does not differ from the control. Naturally, in connection with this there was interest in how these organella in the cells which developed in weightlessness look. Figure 3 shows electron microphotographs of mitochondria in the cells of the flight and synchronous (ground) variations. As is seen from the photo, the structure of these organella in the cells grown in weightlessness differ from the control. The mitochondria appears as swellings with a small number and small orderliness of internal These changes are typical manifestations of a nonmembranes. specific response reaction (more precisely, it is the initial easily reversible stage). The characteristic changes which occur in the mitochondria were noted by us during interaction of different factors of the environment (Tairbekov, 1973). As is known, the energy system of cells is more sensitive to changes in the parameters of the ambient atmosphere. The completeness and normal activity of this system are a guarantee for retaining internal homeostasis of the cell. Consequently, changes which occur in the mitochondria are the first signal for possible breakdown in the metabolism of the system. The change in structure of other cellular organellae: nuclei, dictyosis, endoplasmic reticulum are secondary and occur as the result of a shift in the energy process. What can the mechanism of the possible effect of weightlessness be in the formation of cellular organellae?

Undoubtedly, each factor of the growing medium of the organism, varying in its force, intensity, duration of effect and direction besides that within the cells of the information studied plays a determining role in the life activity of the organism. The force of gravity is such a factor, in a growing medium. This factor of effect on the organism, which in the process of evolution constantly exerts its influence, is adapted to it and extracts from it a benefit for itself (A. Brown, 1971). From this, one can conclude that the absence of the effect of a given factor can be perceived by the organism including vegetation, as a condition undesirable for its normal existence. Moreover, another no less important aspect exists which is less developed which must be taken into account when working out the theory of the question of the effect of the factor of weightlessness on the organism.

It is possible to propose that living organisms which have developed on Earth, that is, in conditions of the constant effect of the forces of gravity, have developed and secured a genetic type mechanism for whose start one requires a certain energy reserve which compensates for the predominance of the gravitational factor. In conditions of weightlessness, this reserve is not realized because it is not used.

In this case, we find evidence of a situation where the energy system in the cell and in the organism as a whole operates with a certain lack of load which makes an impression on it, in particular, on the characteristics of the morphologic state of the mitochondria. It stands to reason that the thesis presented is still very hypothetical although it is not devoid of a basis.

In the opinion of A. Brown, (1971) the development of space /20 biology, in particular, gravitational, is at that stage where the possible effective factors of space flight are considered at a level not only of the organism but at the cellular, subcellular and even molecular. One should note that a deep understanding of the questions involved in the effect of the forces of gravity is determined by the level of biological organization because the higher the level at which the study is carried out the smaller is the amount of data we obtain. One must find out, continues the author, that many of the most interesting biological phenomena involving the effect of the force of gravity, especially in the vegetable world, were studied primarily at the level of the organism and therefore there are still many "blank spots" in this region.

The functional approach developed in recent years toward the study of the biological effect of a change in gravitation, in particular, weightlessness, which we obtain in "pure form" in experiments conducted on biological satellites and manned spacecraft, are based on the study of aspects involving a change in the energy of cells.

A solution of the disputed questions relating to the theory of gravitational biology is discussed in this article can be

realized when fulfilling studies in a flight experiment on successive biological Earth satellites.

A study of the morphogenesis of lower vegetation in experiments on artificial Earth satellites is of a certain interest from the point of view of the effect of weightlessness on biological systems.

In the present experiment, as in the preceding on the Kosmos-782 in 1975, the object of the research was cultivation of a representative of one of the types of mucorin phycomycetes fungi (Phycomyces blackesleeanus). The experiment prepared and /21 conducted by us on the Kosmos-936 biosatellite was a sequential stage in the study of the biology of phycomycetes fungi and observation was planned of the basic stages of growth and development of this organism beginning with the moment of penetration (feeding) of the fungus spore in the nutritive substratum. For this purpose, as in the experiments with higher vegetation, the Biofiksator-I instrument was used. In the preflight preparation of the instrument, it was sterilized, filled with a nutritive medium, and charged with a suspension of spores and fixer. The initial concentration of spores was 50,000 units/ml of suspension. The nutritive medium was a 2% solution of must-agar. 2% glutagaldehyde was used as the fixer. The final assembly of the instrument before putting it on board was done in a sterile The seeding was carried out on the first days of flight box. simultaneously in all ten chambers. Fixation was accomplished according to the cyclogram presented above. Upon completion of the experiment, in the landing area, an examination was made of the instrument for visual evaluation of the material obtained. When the culture chambers were opened it was found that the material fixed only on the tenth day of the flight (chamber No. 5) was suitable for analysis. The colonies of fungi grown in this chamber reminded one in external appearance of a matted deposit of a grayish yellow color and occupied four fifths of the surface of the substratum. The airborne mycelia, rich in sporangia carriers, were 1-1.5 mm above the substratum. Approximately one third of the sporangia were mature, of a brown or black color. There were many immature sporangia which were white, bright yellow, yellow brown or of a yellow green color. The airborne mycelia carrying the sporangia, are branched, and from the substratum. In the synchronous bent away experiment, the formation of sporangia carriers with sporangia was not noted. The presence of a large number of sporulating /22 mycelia in the test variation, if you please, can be explained by the fact that the fungus spores found in an aqueous suspension for three days before launch had a temperature of approximately 28-30°C, grew, and on the tenth day of flight of the biosatellite, the mycelia reached the third and fourth phase of growth.

The mycelia hypha branched freely and extended along the nutritive medium; the maximum thickness in the test was equal to 33 m and in the control -- 38.5 m, although they seemed less mature. However, the thickness of branching of the mycelia in flight was somewhat smaller than in the synchronous experiment. In the flight variation, growth of the primary end of the hypha was predominant whereas branching was suppressed. Certain biological characteristics of the fungus are presented in table 5.

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Bilogical Indices	FI	Light		Synchron Experime		
	Stat Ir	istical ndices	• 	Stati Ind		
	M	m_	<u> </u>	M	_ m _	n
Value of the angle of - branching (in degrees)	84	0,46	90	79,5	0,4I	90
Diameter of mature sporangia (in M)	125	3,5	20	Not fo	ormed	
Diameter of immature sporangia (in M)	74	2,6	20	Not fo	ormed	

[Commas in the tabulated material are equivalent to decimal points]

The granular protoplast of the cells broke away from the sheathings and in certain areas accumulated into small clumps; as a result of this, unequal distribution of it ocurred in the cells. However, the microstructure was not broken down. In our opinion, this effect can be the result of long term storage of the material in a fixer. The size of the sporangia in flight varied from 66 m (immature) to 143 m. For a quantitative evaluation of the degree of orientation of the fungus in conditions of weightlessness, measurements were made of the angles of branching of the hypha of the mycelia of different magnitudes. The average branching angle of the mycelia in the flight material amounted to $84^{\circ} \pm 0.46^{\circ}$, and the synchronous control -- 79.5 ± 0.41 .

Thus, summarizing consideration of the preliminary results $\frac{23}{23}$ obtained in the experiment on the Kosmos-936 biosatellite, with

different biological objects (insects, higher and lower vegetation), made using modern methods of study (molecular genetics, cytogenetics, electron microscopy) can, with a certain authenticity, show that with a relatively long period of the life organisms remaining in conditions of weightlessness, coinciding with the active stage of their formation, a number of insignificant changes occur in the structure and functional peculiarities of their development (frequency and rate of the division of cells, their shape, dimensions and the ultrastructure of the cellular organellae).

However, according to our convictions, these changes have a physiological character, that is, they are easily reversible, do not leave a structural trace and do not affect the genetic apparatus of the cell.

Consequently, relying on the data obtained in this article and calling attention to results of previously conducted studies (Kosmos-605, 690, 782) one can propose that weightlessness as a factor of the ambient atmosphere cannot be the principal obstacle for normal occurrence of basic biological processes in the developing organism.

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Figure 3.

Mitochondria in a cell from the tensile zone of the tenth day of corn roots. Electronogram mag. 20,000 X.



a) Test/weightlessness



b) Control synchronized on Earth

Summary

The biological experiments on board the Kosmos-936 satellite were carried out in order to further and more deeply study the mechanism of the effect of weightlessness on the character of formation and development of the basic components of the cells, primarily, the genetic structure and energy apparatus.

Genetic studies were carried out in cooperation with American specialists on the fruit fly (Drosophila melanogaster) of the Oregon-R strain. The possible effect of weightlessness was studied on the structural changes in the chromosomes and the appearance of gene mutations, features of morphology, anatomy, and also on the behavioral reactions and degree of survival rate in the insects.

In the experiments on higher vegetation, the research objects were sprouts of Crepis (Crepis capillaris) and corn (Zea mays) grown from air dried seed on board the biosatellite.

Cytogenetic studies were made on the crepis seedlings. The purpose of the study was to discover the possible effect of weightlessness on spontaneous mutagenesis in cells of root meristems. For studying the characteristics of formation and development of the ultrastructure of vegetative cells in weightlessness, a comparative electron microscopic analysis of the material was carried out (cells of the root meristems) grown on board the biosatellite and in a ground control.

The experiment with lower phycomycetes fungi (Phycomyces blackesleanus) was carried out for studying the characteristics of morphology and shape formation in weightlessness.

The results of studies in general biology obtained in the process of analysis of the flight material and ground control indicated that weightlessness does not cause significant changes in the processes of development and formation of the basic biological processes and consequently, cannot be the principal barrier for normal development.

Modeling of Ectopic Osteogenesis in Weightlessness

The effective factors of space flight on the skeletal bone tissue is receiving fixed attention. It is well known that in weightlessness, weakening of the process of bone formation (osteoporosis) occurs and the loss of calcium salts (Yagodovskiy et al., 1977, Vogel, 1973, Whedon, 1972, Mack et al., 1971). A breakdown in skeletal tissue can be considered as a "barrier to weightlessness" (according to the descriptive expression by Hatner and McMillan, 1968), which can significantly limit the duration of space flights. Similar changes in bone tissue were observed during hypokinesis (Rokhlin, et al., 1975, Vogel et al., 1970). The similarity in development of the character of change of bone tissue in weightlessness and during hypokinesis gives us a basis for assuming that a breakdown in bone formation is the result of the deficiency of support muscle load. However, the absence of mechanical load cannot be the only cause of breakdown of osteogenesis. One must not exclude the fact that in conditions of space flight, a breakdown in the process of bone formation can involve changes in the system (neurohumoral) which regulate the formation and remodeling of the bone. Besides the support role and the participation of the bone in mineral exchange, it also fulfills a hemopoietic function (Korzhuev, 1971), that is, the bone tissue participates in histogenesis of the bone marrow. Data on cultivation in vitro of bone marrow indicates this on the bony stroma previously developed (Luriya, 1972). In connection with this, one should expect a decrease in the hemopoietic function of the bone marrow in weightlessness inasmuch as there is no necessity for it to be retained in /29 the same volume as in ground conditions. The function of the bone marrow in these conditions must be through the hemopoietic function of the bone and at the same time can be caused by shifts in the bony tissue itself (osteoporosis, loss of Ca. etc.

For testing the expressions indicated above it is advantageous to cause development of the bone outside the skeleton (ectopic), that is, in locations where bony tissue develops neither in ontogenesis nor in phylogenesis. A convenient model for ectopic osteogenesis is implantation in the forward wall of the abdomen of a decalcified and lyophilized bony matrix separated during its resorption of the osteogenesis inductor (Urist, et al., 1965, 1974). It is essential that ectopic bone has all of the morphologic and histochemical traits which exist in skeletal bone tissue (Fridenshteyn, 1963).

In this section, induced activity of a bony matrix was studied in different types of animals. Obtaining these materials was proof of the adequacy of our using the bony matrix for studying ectopic osteogenesis and blood circulation in conditions of weightlessness.

I. Ectopic Osteogenesis as a Model for Studying the Processes of Histogenesis of the Bone and Bone Marrow in an Experiment.

Many attempts to cause ectopic osteogenesis in young animals by transplantation of both live and dead bony tissue are well known (Fridenshteyn, 1963). They demonstrated the undoubted capability of skeletal tissue to induce ectopic osteogenesis /30 having advanced two concepts as to the mechanisms of this effect. One of the experimenters connected the development of new bone to participation of cells of granulated and connective tissue of the host and considered this phenomenon as the induction Other scientists connected the ectopic osteogenesis process. with activity of the bone transplant itself. The first point of view has been further developed being supported by many tests, particularly by experiments with induction of the bone using transplants from dead skeletal tissue (Uris et al., 1965, 1967, 1968). These experiments have raised the question as to the nature of the inducer which stimulates connective tissue of the host and the cell predecessors which perceive this inducer. We will not discuss all of these questions because they are well documented in a number of works (Fridenshteyn, Lalykina, 1937, Urist et al., 1968). Summarizing the results of tests of different authors, one can with authenticity say that the high percentage and rapidity of obtaining ectopic osteogenesis under the effect of deceased bone tissue confirms that in these tissues a chemical substance exists which induces osteogenesis.

The purpose of this series of tests was to reproduce the Urist method (Urist, 1965) in growing ectopic bone using a bony matrix in different species of animals.

Fourteen rabbits, 12 guinea pigs and 30 Wistar rats were used in the tests. Bones of the rear extremities (femur and tibia) were removed from intact donors in sterile conditions. The bone marrow and muscle tissue were thoroughly removed. The bone was cut into sections so that the length of the diaphysial sections amounted to 1-2 cm; they were decalcified in a titrated 0.6 N solution of hydrochloric acid taking into account that 1 gram of bone requires 100 ml of solution (Urist, 1967). The acid was washed off in a sterile 0.15 M flow of NaCl solution for 24 hours. After this, the bony matrix was frozen in dry ice at -50°C and dried in vacuum equipment. Implantation was done under the fascia of the rectus muscle of the animal. The duration of the experiment varied from 25 to 100 days. The transplants with the surrounding tissue were fixed in 96° alcohol to cold (+4°C) or in alcohol formol (9:1). Decalcification of the test samples was carried out in 5% nitric acid or in 25% trilon. The material, after washing in

alcohol, was poured into celluloid paraffin. Sections with thickness 5-8 microns were colored according to Mallory, Van-Gizon, with hematotoxylin-eosin, and also on alkali phosphatase according to Gomor, on polysaccharides (ShLK-method) on Ca salts according to Koss.

The results of the tests are summarized in table 1; from this it is apparent that induction of osteogenesis was obtained in conditions of homotransplantation in rabbits, guinea pigs and rats. Each recipient was implanted with six samples of

Table 1

Induction of New Bone in Different Species of Animals

Decalcifica- tion Time (Hours)	Species of Animal	Location of Implan tation	Days N - of test A	Number of nimals	Removal of the ectopic bone (%)	
18 (20 ⁰ C)) ŕabbit	under the fascia	ື25 - 100	5	1 00	
· · · ·		subcutanèo	ous	5	100	
	ir	tramuscula	ar	4	100	
12 (20°C)	guinea pig	under the fascia	45	6	0	
6 (20 ⁰ C)	n <u>n</u>	11 11	34	6	100	i
50 (2 ⁰ C)	rat	11 11	28	12	96	·
48 (2 ⁰ C)	'rat.	<u>n n</u>	18	18	78	

bony matrix. From the table one finds that for the factor of /31 bony induction decalcification time of the initial material was not different. Thus, decalcification of the skeletal tissue of guinea pigs for 12 hours makes the osteogenic inductor inactive. Such transplants are reabsorbed after 45 days not causing an induction effect. If the duration of decalcification of the same material had been six hours, then one would have observed intense bone formation reaction after 34 days. Thus, the optimum duration of decalcification can vary. In rabbits, in dis- /32 tinction from guinea pigs, the decalcification time equaling 18 hours is the most suitable for induction of osteogenesis.

The location of the transplantation did not play a role in rabbits because implantation of the matrix subcutaneously, under the fascia or in vagina of the rectus muscle always is accompanied by the development of new bone.

The morphology of induced bone formation, as a whole, was similar in all of the groups of animals studied. The earliest stages of induction were not studied. The transplants were fixed at a moment when they became the densest by feel and contained as was seen during microscopic studies, recognizable bony tissue. A transplanted bone matrix, in all cases, was fatal with an excess of osteocytes and osteoblasts; this basic substance gave a reaction to polysaccharides, had a negative reaction to alkali phosphatase and Ca salt. Beginning with 31 days, the implants are greatly loosened, filled with histeocytes, the cells of foreign bodies, granulated and loose connective tissues, and are rich in small blood vessels. A reactive connective tissue consisting of fibroblasts and histeocytes develops around this implant. On the interior surface (on the endosteal side) one noted intense formation of bone tissue. It consisted of typical bony pieces having a coarse fibrous structure. In some cases, newly formed bony tissue directly adjoined the dead bone and bordered its lines of ossification. In others, the newly formed bone was separated by a layer of cellular fibrous tissue consisting of fibroblasts and osteoblasts. The induced bone, in all cases, had a typical structure. It consisted of several layers of osteoblasts and basic substance giving a positive reaction to the alkali phosphatase and polysaccharides. In the internal regions of the newly formed bone, there were deposits of calcium salts. Alkali phosphatase was localized in the zone of active proliferation of osteoblasts and was absent in the zone of formation of calcified bony tissue. Control for alkali phosphatase and the Koss reaction clearly indicate the absence of calcium salts in the zone of activity of the osteoblasts.

On the 35th day after implantation, and in a number of cases earlier, one observed the development of hemogenic tissue between the bone pieces. The latter consisted of all cellular elements encountered in the bone marrow of skeletal bone. At the early stages of development of new bone (3-4 /34 weeks) local positioning of hemogenic islands was characteristic consisting of erythroid and myeloid cells. In the last time periods (6-7 weeks) the bone marrow filled a large space. Bone marrow developed most strongly in rabbits, less so in rats and very slightly in guinea pigs.

Thus, ectopic osteogenesis was obtained and reproduced according to Urist under the effect of a bony matrix in different

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species of animals. The osteogenic tissue obtained, in morphological structure and histochemical reaction, was identical to skeletal bone tissue. These materials are the basis for conducting experiments on rats irradiated on the Kosmos-936 biosatellite.

2. Modeling of Ectopic Bone in Weightlessness

In the tests, SPF rats were used weighing 100-120 grams in which, three weeks before flight, decalcified and lyophilized bone matrix of the femur and tibia bones were implanted under the fascia or the front wall of the abdomen (6 samples per rat) from a rat of the same strain weighing 200 grams. The test rats were made up of three groups: group PN2 (10 rats), SN2 (10 rats) and VK₂ (7 rats). Part of the animals (about 5 rats) from groups PN2 and SN2 were slaughtered on the day the test was organized (background control); the other animals, immediately (11-13 hours) after completion of the test. In the 10-13 days after implantation of the matrix, the animals of all groups (carriers of the bony matrix) were immunized with sheep erythrocytes (this program was carried out independent of ours). All of the animals before and after implantation were put on a "space diet".

In the animals killed on the day the biosatellite was launched (22 days after implantation) on the day SN₂ was put into the test (19 days after implantation), the bony matrix under visual examination was solid which indicates the accumulation of Ca salts in them. During microscopic analysis (in 92% of the cases) separate sections of coarse fiber bone were detected whose development was observed both in the locations of reabsorption of the cortical part of the matrix and within the cavity on the endosteal side of the old bone. In the bone marrow cavity of the old bone, as a rule, coarse fibrous connecting tissue developed in combination with delicate fiber penetrated by small blood vessels. In the cortical part of the matrix, the process of resorption was intense thanks to the presence of multinuclear symplasts similar in morphology to osteoplasts. Thus, in both groups of rats, before beginning of the test, the ectopic bone was well developed which indicated capability for use of the bony matrix to induce new bone. The bone marrow was not yet developed in this period.

After 18.5 days of flight (40.5 days after implantation) or completion of the SN_2 test (38.5 days after implantation), and also in rats from the VK_2 group (46.5 days after implantation) a further development of ectopic bone occurred whose volume was somewhat larger than in the prelaunch period. It is significant that that no important differences were detected in volume of new bone between rats of the PN_2 , SN_2 and VK_2 groups.

In space flight, as in the standard, the process of resorption of the old matrix was as intense as in the preflight period. In distinction from the preflight state, during flight, bony and cartilaginous tissue developed. One should note also the presence in the bone marrow cavities of old bone with strong growth of coarse fiber connective tissue and hemorrhage foci in it. In the location of the cortical bone of the matrix, and often in its cavity, besides the development of a new bone, bone marrow develops. The cellular make up of the extramedulla bone marrow in rats from the PN₂ group was less marked in comparison with the cellular content of bone marrow in the control rats. In rats from the SN₂ group, the bone marrow was exclusively fatty cells and very rarely erythroid and myeloid elements. Such a picture is similar to aplasia of bone marrow after the effect of large doses of radiation causing the development of the bone marrow syndrone.

Summarizing the results of this study, one can draw a conclusion which is still preliminary that the development of bone outside the skeleton is not subject to the effect of the factors of space flight. This makes it possible to propose that a breakdown in the development of the skeletal bone tissue which occurs in weightlessness involves, basically, a deficit in the support muscle apparatus. Apparently, the role of neurohumoral components in a change of skeletal bone is insignificant. Moreover, suppression of the extramedullary myelopolisis was observed in ectopic bone which attests to weakening (at the system level) of functions of the bone marrow in the organism.

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Summary

It was established that the development of ectopic bone is not subject to the effect of factors of space flight because extramedullary hemopoeisis is suppressed in ectopic bone.

Radiation-Physical Research

The main directions in which the experimental studies were made on the Kosmos-936 biosatellite were radiation dosimetry and protection from cosmic radiation. Part of the studies were carried out with the participation of American and French specialists.

The purpose of the experiment on radiation dosimetry was the study of dosimetric and spectrometric characteristics of cosmic radiation in near-Earth space, and also the study of the passage of contaminated particles of cosmic radiation through the protection material and biological tissue. Moreover, measurements were made of the spectra of linear energy loss, the charge composition, the dose characteristics of cosmic radiation within and outside the satellite. Special attention was given in the experiment to the study of the characteristics of heavy nuclei of galactic cosmic radiation for planning further radiobiological experiments in space and on accelerators.

The purpose of the experiment in protection from cosmic radiation was the study of the characteristics of elements of different kinds of electrostatic protection, vacuum and dielectric.

- 1. Radiation Dosimetry
- A. Dosimetric and Spectrometric Studies
- 1. Problems of the research

On the Kosmos-936 artificial Earth satellite, dosimetric and spectrometric studies were made which had been carried out on the Kosmos-782 AES (Ye. Ye. Kovalev et al., in the press; Ye. Ye. Kovalev, V. V. Markelov, 1978).

The purpose of the research was to obtain information on the dynamics of change of radiation conditions on the flight path, the measurement of doses and power of doses and to obtain the spectra of linear energy loss.

The use on the artificial Earth satellite of instruments with transmission of information according to a telemetry system made it possible to obtain data in a form differentiated according to time which makes it possible to present the picture of change of parameters along the flight path.

Measurement of doses, the power of the doses, the flows and spectra of linear energy loss of particles were measured

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in the S-I dosimetric instruments (Akatov, Yu. A. et al., 1978).

2. Methods of the Research

For correct processing of the readings of the dosimetric instruments, it is necessary to know the distribution of solid angles relative to the detector of the instrument according to thickness of the material of the equipment. Using an instrument for gamma rays (Gribov, B. S., 1977) the distribution of thicknesses was obtained for the S-I spectrometer, the Soviet-American K-206 container, the Bioblok container. Determination of the thickness of equipment is based on a comparison of intensity of gamma radiation passing through the equipment with intensity in its absence. The principle of action of the unit involves the fact that at a given point inside the equipment, there is an isotopic gamma source and recording of nonscattered radiation passing through the equipment is carried out by a collimated scintillation detector which moves outside the object of the coordination system along a spherical surface with the center at a given point. The overall dimensions of the measured object are up to 5 m and thickness up 50 g/cm². The solid angle for single measurement is $0.7.10^{-4}$ steradian, precision of measurement is \pm 0.3 g/cm² with thickness 1 g/cm² and \pm 0.5 g/cm² with thickness 10 g/cm². The duration of measurements in a /41 solid angle is 4-30 hours.

Table 1 shows distribution of solid angles according to thickness for a S-I spectrometer, the K-206 container and the Bioblok. Measurements were made within the limits of a solid angle of 3.2 steradians for the S-I (solid angle of the telescope of the spectrometer). The geometry of measurements for K-206 container and the Bioblok are presented in drawing 1.

The distributions presented make it possible to correctly interpret the results of measurement of the functionals of cosmic radiation and evaluate their functionals in open space.

Telescopes of Counters

In the experiment on the Kosmos-936 artificial Earth satellite, measurements were made of linear energy loss using two S-I analyzers one of which was mounted inside and the other on the outside of the surface of the satellite. A telescope was used in the S-I analyzer as the sensor; it consists of a silicon semiconductor detector with thickness of the sensitive region 500 microns \pm 3% and area 1 cm² and a scintillation counter with crystal of cesium iodide, thickness 10 mm and diameter 10 mm. The use of a protected detector, a system of coincidence and anticoincidence excluded the recording of particles passing at large angles to the axis of the telescope. The electron circuit of the analyzer produced an amplitude analysis

Table 1

Distribution of Solid Angles According to Thickness for the K-206, the Bioblok, and the S-I.

No	S'g/cm ²	4	W stera	adian	No	g/cm ²	4	W stera	dian
	•••••••	K-206	Bioblok	S-I.	-		K-206	Biøblok	S-I
I.	2,0	-	-	0,044	18.	IO-II	0,292	0,279	0,092
2.	2,0-2,5	-		0,060	19.	II-I2	0,258	0,664	0,186
з.	2,5-3,0	-	=	0,121	20.	12-13	0,220	0,631	0,193
4.	3,0-3,5	-	-	0,025	21.	13-14	0,197	0,586	0,164
5.	3,5-4,0	-	0,040	0,050	22.	14-15	0,187	0,302	0,091
6.	4,0-4,5	-	0,051	0,084	23.	15-16	0,198	0,299	0,101
7.	4,5-5,0		0,082	0,101	24.	16-17	0,199	0,304	0,101
8.	5,0-5,5	0,073	0,131	0,102	25.	17-18	0,186	0,297	0,103
9.	5,5-6,0	0,143	0,142	0,081	26.	18-19	0,175	0,291	0,106
10.	6,0-6,5	0,187	0,161	0,064	27.	19-20	0,179	0,279	0,107
II.	6,5-7,0	0,279	0,165	0,066	28,	20-25	0,564	0,702	0,281
12.	7,0-7,5	0,837	0,157	0,082	29	25-30	0,859	0,703	0,233
13.	7,5-8,0	0,337	0,125	0,104	30.	30-35	0,730	0,514	0,086
14.	8,0-8,5	0,370	0,II7	0,115	31.	35-40	0,640	0,543	0,011
15.	8,5-9,0	0,337	0,126	0,108	32.	40-45	0,360	0,368	-
16.	9,0-9,5	0,286	0,148	0,095	33.	45-50	0,330	0,419	-
17.	9,5-10,0	0,265	0,228	0,091	34.	50	0,470	0,085	

[Commas_in the tabulated material are equivalent to decimal points.]

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For the K-206, the solid angle $\Delta \omega_1 = 1.57$ steradians with thickness more than 5 g/cm² and $\Delta \omega_2 = 2.28$ steradians with thickness more than 20 g/cm² (ends of the satellite) were not measured.

For the Bioblok, the angle $\Delta \omega_1 = 2.46$ steradians with $\delta > 5 \text{ g/cm}^2$ and $\Delta \omega_2 = 1.20$ steradians with thickness $\delta > 20 \text{ g/cm}^2$ (ends of the satellite) were not measured.

Table 1, continued

Distribution of Thicknesses of the Relative Boundary of K-206

No: Bou arie	s: 0 o	φo	: S g/cm ²
I	90	90	8.5
2	. 90	180	II-O
3	90 .	270	22.0
4	90	0	55.0
5	180	-	20.0
6	0	••••	6.0

Boundary 1 was perpendicular to the cover of the hatch.

Table 1, continued

Distribution of Solid Angles According to Thickness for the K-206 Relative to Boundary 1.

0°	₽°.	S g/cm ²	Steradian
0.40	0 360	>5	I.57
40.140	0.360	5-60	8.71
140 180	0 360	>20	2.28

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Figure 1: Geometry of Measurements

of the pulses coming from the semiconductor detector, record- $\frac{41}{100}$ ing of pulses in the appropriate channels and transmission of information to the onboard memory device of the satellite and Earth according to the telemeasurement system. Then, the logic device of the analyzer provided recording only of those particles whose length exceeded the thickness of the sensitive region of the detector and energy generation in the scintillator was larger than the selected level of discrimination. The measured $\frac{42}{100}$ amplitude of the pulse in a silicon detector is proportional to linear energy loss of charged particles passing through the telescope. The coefficient of proportionality was determined with calibration of the analyzer for alpha particles (Pu-239) and for the accelerator of protons.

When processing the telemetric information for each recorded charged particle, the value of energy generation in the silicon detector was determined and the value of linear energy

loss was calculated in biological tissue. According to the data obtained, differential spectra of linear energy loss of charged particles was constructed and the value of the equivalent dose was calculated and then the average value of the coefficient of the quality of radiation was obtained.

Dosage Channels of the S-I Instrument

In both S-I instruments on the Kosmos-936 artificial Earth satellite, there were dosage channels with which information on the dynamics of the accumulation of the dose, power of the dose and flows were sent by the telemetric system to Earth. A flowpassage semiconductor detector of the telescope counters was used as the dosimetric sensor. The electronic block of instruments produced a total of amplitude of the pulses of the particles passing through the detector which provided measurement of the absorbed dose of cosmic radiation. A high dosage sensitivity of such a channel made it possible to obtain a picture of the change of power of the dosage along the turns of the flight. Calibration of the dosage channel was made according to the alpha source, the accelerators and the gamma sources.

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3. Results of Research with the S-I Instrument

Measurements of the dynamics of accumulation of doses along the flight path indicate a large change in the power of the doses in certain areas of space. The smallest value of power of the dose is observed in the equatorial region and has a value of 0.2 milliroentgen/hour inside the object. Here, the mean value of density of flow is 2 $\frac{\text{part} \cdot \text{s}}{\text{cm}^2}$. This value of power of the dose and flow is due to the maximum screened effect of the magnetosphere of Earth for pulse generator particles. In the central latitudes, the power of the dosage increases and reaches 1.5 mr/hr. The effect of radiation within the radiation bands of Earth is apparent in the periods when the AES passes through the region of the Brazilian magnetic anomaly of Earth. Then, the maximum value of the power of the dosage within the object can reach 200 mr/hr. In spite of the fact that of the 16 daily turns only six pass through this region, the contribution to the total dose of the protons within the Earth band is on the order of 60%. At latitudes of 60-65°, episodically one observes surges of power in the dosage caused by the electrons of the outer radiation band of Earth. The largest values of power of the dosages is recorded by the external instrument and reaches a value of 20 rad/hr. Inside the satellite, the increase in these periods is significantly smaller and amounts to 20 mr/hr.

The integral dose measured by the internal instrument for the entire flight amounted to 620 mrad. Dosimetric measurements along the flight path indicate very significant

nonuniformity of irradiation of bioobjects in time which, must, obviously, be taken into account when interpreting the biological data.

Measurements of the linear energy loss spectra by the internal S-I instrument indicated that the differential dosage spectrum on the LEL scale can be a graduated function with the index of the degree, 1.7. Calculation of the value of the factor of quality gives a value on the order of 1.5. The low value of the quality factor can be explained by the significant cutting off of the heavy components of the gamma ray cosmic radiation by the magnetosphere of Earth.

Processing of the information is continuing at the present time.

4. Nuclear Emulsions

The basic results according to nuclear photoemulsions result in the following: a sample display of the emulsion indicated that for 20 days of flight, the density of the tracks in the relativistic BR-2 emulsion is so great the measurement of the characteristics of separate tracks is extremely difficult. Also a significant gamma background is observed. Part of the layers were subjected to the effect of an electric field with the use of which one could successfully photograph gamma backgrounds and relativistic particles. Measurement of flows on heavy nuclei of gamma ray cosmic radiation was begun along this layer. Also, processing of flight and calibrated emulsions of the PR-2 type was begun according to a method of 2-potential developments. At the present time, the American nuclear emulsions from the general section of containers are being developed. The density of the track in the relativistic 2-5 emulsion are also very large. A less sensitive K-2 emulsion is fully suitable for the measurements. At the present time, processing is continuing.

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SUMMARY

Dosimetric studies were made on the Kosmos-936 AES using telescope sensors, nuclear photoemulsions and thermoluminescent dosimeters. The size of doses, power of doses, LEL spectra of cosmic radiation were measured on the flight path of the Kosmos-936 AES. The value of the coefficient of quality of radiation was obtained, equaling 1.4. Information was obtained on the dynamics of change of the power of doses along the turns, on the contribution to the integral dose of gamma ray cosmic radiation and the internal radiation band of Earth, and on the dynamics of accumulation of the general dose. Measurement was made of the distribution of solid angles according to thickness in material of the equipment relative to the radiation detectors.

The upper estimates of flows of neutrons were obtained in the joint Boviet-American dosimetric K-206 experiment, measured using dividing detectors made of neptunium, uranium and bismuth. Flows of heavy particles of gamma ray cosmic radiation were measured using plastic detectors. A series of ground calibrated experimental detectors was produced for accelerators of protons and multicharged ions in the USSR and USA.

The processing of the results is continuing.

B. Studies of Flows of Heavy Nuclei of Cosmic Radiation Using Dielectric Track Detectors.

Studies which have been made on the Kosmos-690 and Kosmos-782 AES were continued on the Kosmos-936 (Marennyy, A. M. et al., 1977).

1. Tasks of the Research.

A number of detectors intended for solving the following problems were placed on board the Kosmos-936 AES:

- measurement of the planar flow of heavy nuclei of space radiation at distant points on the object, that is, for different protective layers; 150

- obtaining the integral spectrum of LEL inside the object;
- obtaining charged and energy spectra of heavy nuclei within the object;
- detection of biological objects damaged by heavy nuclei of cosmic radiation;
- a comparison of methods of identification of particles used by different groups;
- testing of new methods and materials for detectors.
- 2. Methods of Experimental Research.

Blocks of detectors were placed in special containers at different points on the object.

1. The KNA containers were placed on the external surface of the descent vehicle; blocks and separate layers of detectors were placed in them intended basically for recording the low energy heavy nuclei of gamma ray cosmic radiation.

2. A container with equipment from the Soviet-American K-206 /51 experiment contained four large blocks in a system or orthoganol tracking detectors (DTD and YaFE); the location of the unit is inside the satellite directly behind the paneling. Charged energy spectra of heavy nuclei within the objects, LEL spectra, angular distribution of flows of particles and density of the particle units will be obtained upon completion of processing the results of measurements.

3. The block of detectors with thickness 0.7 g/cm^2 is located along the axis of the S-I instrument located inside the object. This combination makes it possible to "prolong" the integral spectrum of linear energy loss obtained using the S-I in the field of linear energy loss values up to approximately $1000 \frac{\text{KeV}}{\text{microns}}$.

4. The container with the equipment of the Soviet-French Bioblok-SF-2 experiment is located close to the geometric center of the object. A block with bio-objects, a block of detectors and a system of orthogonal detectors (DTD and YaFE are located in the container.

Lavsan detectors, nitrocellulose KNTs detectors and Kodak make up the composition of different blocks. Several methods will be used for identification of particles: according to etching rate, according to the full etched length of the tracks and according to the difference in rates of etching at two points of the particle trajectory.

3. Results of Experimental Studies.

Up to the present time, the following results have been obtained: planar flow of high energy heavy particles from $\text{LEL}_{1000} \ge 200 \frac{\text{KeV}}{\text{microns}}$ on the external surface of the object -- (13.4 ± 0.6) cm⁻², inside -- (2.9 -- 7.6) cm⁻². The planar flow of heavy particles with $\text{LEL}_{350} = 30 \frac{\text{KeV}}{\text{microns}}$ at the center of the Soviet-American container changes from 12 to 32 cm⁻² depending on direction. 107 damaged bio-oblects were discovered when scanning the Bioblok.

Processing of the results is continuing.

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Summary

The planar flow of high energy heavy nuclei with $LEL_{1000} \ge 200 \text{ KeV/microns}$ was determined on the external surface of the object and within. During scanning of the Bioblok, 107 damaged bio-objects were discovered. The flow of heavy particles of cosmic radiation within the AES is smaller, by at least, 5 times than on the surface of the spacecraft on the flight path of the Kosmos-936 AES.

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C. Dosimetri: Measurements Using Thermoluminescent Detectors. /54

When carrying out biological experiments in space an important parameter for evaluating the effect of factors of space flight on biological objects is the value of the absorbed dose of ionizing radiation. The value of this parameter cannot be indicated ahead of time with adequate precision because the radiation condition in space is strongly varied. The protective properties of the space objects and the position of the experimental samples on board also have a large effect on the forming of this dose. In many experiments on AES, with manned space flights, for measuring the absorbed dose, thermoluminescent detectors were successfully used (Akatov, Yu. A. et al., 1971; Grigor'yev, Yu. G.; Kovalev, Ye. Ye. et al., 1976).

When realizing the scientific program on the Kosmos-936 biological satellite, measurements of integral doses by thermoluminescent detectors was part of the complex dosimetric experiment which included the Soviet-American K-206 experiments.

1. Research tasks.

The research tasks were determining the values of integral doses of ionizing radiation in the areas where the Iondoz (K-206) was located, the S-I, the biological objects and other experimental samples placed both inside the satellite and on its exterior surface in KNA type container boxes.

In addition to a similar experiment carried out on the Kosmos-605, 690 and 782 satellites, on the Kosmos-936 satellite, the problem of studying attenuation of cosmic radiation was set up for passage of thin layer; of protective material. /55

2. Methods of the Experimental Research

In the experiment, thermoluminescent glass detectors of the TLS-2 type were used with dimensions 13X13X4 mm, 126 pieces, of which 54 pieces were mounted inside the satellite and 72 in the external containers. The detectors were placed in locations where the control objects were mounted or were directly attached to them. In the external containers, the detectors were located in boxes in two layers of four pieces each. Then, each first layer on the upper side of the half space was screened by different thicknesses of protective material in a range from 0.015 to 1.2 g/cm².

Taking readings from the TLS-2 detectors was done after completion of the experiment in laboratory conditions on the IT-1 unit.

3. The Results of Experimental Studies.

As a result of processing, values were obtained of integral doses which are characteristic for different locations inside the satellite and on the external surface. The values of integral doses at the Iondoz unit position were: 0.68 ± 0.04 rads -- on the upper surface of the assembly; and 0.57 ± 0.06 rads -- on its lower plane.

A dose of 0.52 \pm 0.01 rads was recorded on the S-I instrument position.

The values of integral doses on the external surface of the satellite depend strongly on the degree of screening of the detectors and are within a range of 0.87 to 20.5 rads.

An evaluation of the thickness of the screens of the assemblies was made. A table was drawn up according to the results of the relationship of the value obtained of the integral dose on the thickness of the screen over the detector in the upper half space.

Table

Protecti g·cm ⁻²	on 0,015	0,027	0,98 I	,02÷I,I3 I,	,2I÷I,34	3-7 Inside the object
Dose, rad	.20,5 <u>+</u> 2,9	15,4 <u>+</u> 1,6	I,I2 <u>+</u> 0,06	0,94 <u>+</u> 0,03	0,88 <u>+</u> 0,0I	0,52÷0,68

On the side of the assemblies turned toward the object (lower half space), protection can be evaluated as not less than 10-15 g/cm². The value of the dose with minimum protection is 0.015 g/cm² obtained according to the 4-m TLS-2 detector. The relatively large spread of their readings $(\pm 14\%)$ can be due to different screening of the detectors of of the metal net which covers the assemblies and also shading of the assembly by the container covers. The value of the dose for the shield is 0.027 g/cm² obtained according to 8 TLS-2 detectors with a spread of $\pm 10\%$.

Thus, the absolute values of the size of the dose for small thicknesses of protection are relatively great for near Earth orbit which indicates the necessity for further study of attenuation of radiation outside the spacecraft in order to evaluate radiation danger for operation of the cosmonauts in open space.

The drop in the dosage value by 15-20 times when changing the shielding from 0.015 - 0.027 g/cm² to 1.0 g/cm² attests to the very soft spectrum of radiation outside the object. It is obvious that the study of the curve of attenuation of the dose must be made with the finest detectors possible.

The values of the doses inside the object depend on the actual screen to a much lesser degree. The maximum drop amounted to about 30%.

The presence of dosimeters for control made it possible to determine the value of dosages received by biological objects and other samples and, thus, to correlate the studied effect with radiation effects.

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Summary

Measurements were made of integral doses of cosmic radiation on the Kosmos-936 biosatellite. Thermoluminescent glass TLS-2 type detectors were used. The value of the dose inside the object was 0.52-0.68 rads for 20 days of flight. The value of the dose outside the satellite depends strongly on the thickness of the screening layer over the detectors. For a thickness of 0.015 g/cm² and 4.27 g/cm⁻², values were obtained for doses of 20.5 and 0.88 rads, respectively. Further study of attenuation of the doses outside the space objects in near Earth orbit must be made in order to evaluate the radiation danger for cosmonauts working in open space.

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