EFFECT OF VIBRATION, AS A FACTOR ASSOCIATED WITH COSMIC FLIGHT, ON A LYSOGENIC CULTURE OF E. coli K-12 (λ)

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The biological space research program envisages the study of the conditions of life in cosmic space by biological and, in particular, by microbiological methods and the investigation and the genetic effects produced by the action of the factors of flight in cosmic space.

Investigations to study the genetic effects of cosmic flight were undertaken with lysogenic bacteria. In the lysogenic strain E. coli K-12, carrying prophage λ, a genetic reaction (the formation of induced phage) is produced by tenths of a roentgen. It could therefore be used in all cases when it was necessary to determine how cells react to ionizing radiation. The use of a lysogenic system is particularly promising for the elucidation of the genetic effects of agents acting collectively: ionizing radiation and factors associated with cosmic flight, for example vibration, acceleration, and weightlessness.

In the present experiments, carried out during the flights of ships "Vostok-3,""Vostok-4," "Vostok-5," and "Vostok-6" [1,2], a statistically significant increase was observed in the induced phage production of E. coli K-12 (λ) in the experimental specimens by comparison with the controls remaining on the cosmodrome.

TABLE 1. Effect of Vibration of Different Frequency on Phage Production of the Lysogenic Culture E. coli K-12 (λ)

$R\left(\frac{Nr}{Ns}\right)$	Frequency and duration of vibration					
	18 cps 15 min	35 cps 30 min	75 cps 30 min	100 cps 15 min	700 cps 15 min	700 cps 60 min
R_1	1,1	1,2	0,9	0,7	0,6	0,7
R_2	0,6	1,3	1,8	0,6	0,8	0,7
R_3	—	1,1	0,9	0,8	1,0	0,9
R_4	—	1,3	0,9	1,1	1,0	0,9
R_5	—	0,6	0,8	—	—	—
R_6	—	0,5	1,5	—	—	—
R_7	—	0,6	0,5	—	—	—
R_8	—	0,7	0,9	—	—	—
R_9	—	2,8	1,6	—	—	—
Mean	0,85	1,1	1,1	0,8	0,85	0,8

Legend: Nr) number of phage particles induced in experiment: Ns) number of phage particles formed spontaneously.

Phage production in the specimens exposed in V. Bykovskii's cabin was 3.67 times greater than the control value, and in those exposed in V. Nikolaeva-Tereshkova's cabin it was 1.7 times greater.

Special experiments showed that an increase of three times in phage production corresponds to the inducing effect of acute γ-ray irradiation in a dose of ≈ 50 R. The same degree of phage induction may be produced by the action of chronic irradiation on lysogenic bacteria in a dose of 3.2 R at a dose rate of 4 R/day in frozen culture conditions [1, 2]. Analysis of the readings of physical apparatuses shows that a dose of 50 R is approximately three orders higher than the dose during orbital flights. It thus follows that the induction of lysogenic bacteria observed in the experiments during flight of the ships "Vostok-3," "Vostok-4," "Vostok-5," and "Vostok-6" was not entirely due to the action of cosmic radiation. The genetic-inducing effect observed could have been the result of the action of other factors operating in cosmic flight: weightlessness or mechanical agencies—acceleration and vibration.

TABLE 2. Results of an Investigation of the Influence of the Combined Action of Vibration and γ-Irradiation on the Phage-Producing Ability of E. coli K-12 (λ)

No.	Conditions	No. of seedings	Total No. of phage particles, x 10^3	Mean No. of phage particles of samples (M ± m)	R	P
1	Vibration–irradiation–vibration	5	12,667	2533.4 ± 34.54	9.0	< 0.001*
2	Vibration–irradiation	5	9,877	1975.4 ± 8.56	6.9	< 0.01†
3	Irradiation–vibration	5	9,009	1808 ± 23.10	6.4	
4	Irradiation	5	9,108	1821.6 ± 24.02	6.4	
5	Vibration	5	872	174.4 ± 10.50	0.6	< 0.001‡
6	Control	5	1,417	283.4 ± 22.70	–	

Remarks. The index R for each type of factor was calculated in relation to a control culture not exposed to vibration.

* Comparison between 1st type of procedure and 4th.

† Comparison between 2nd type of procedure and 4th.

‡ Comparison of results of exposure to vibration and control.

The object of the present investigation was to determine the effect of vibration on a lysogenic strain of E. coli among the whole group of factors of cosmic flight.

EXPERIMENTAL METHOD

In all the experiments the lysogenic culture was taken after incubation for 5 h in optimal conditions. A suspension of the bacteria was prepared in physiological medium and poured into special flasks. Samples of the culture of E. coli K-12 (λ) in terrestrial conditions were exposed to vibration in a pure form and in various combinations with γ-irradiation from a Co^{60} source (dose 100 R). Vibration with frequencies of 18, 35, 75, 100, or 700 cps was generated on a vibrator for 15, 30, or 60 min (overloading of 10 g).

The number of viable cells and of bacteria producing phage was determined simultaneously in the control and experimental samples.

The experimental results were assessed with the aid of the index R, characterizing the relationship between induced and spontaneous phage production.

RESULTS

The degree of induction obtained in a series of experiments with exposure to vibration in various ranges of frequencies (from 18 to 700 cps) and for various periods is given in Table 1. This table shows that the value of R in these ranges of vibration was approximately the same (0.8-1.1). The number of phage particles in the experimental samples did not exceed the level of spontaneous phage production in the controls, and in fact it was slightly smaller in some experiments.

The results in Table 2 show the effect of the combined action of vibration with a frequency of 700 cps (amplitude of oscillations 0.05 mm, overloading 10 g, time 15 min) with γ-irradiation in a dose of 100 R (dose rate 21 R/min). It is clear from Table 2 that in the samples of the lysogenic culture exposed to vibration and irradiation for 15 min, followed by vibration alone for 15 min, the number of induced bacteria was 40% greater than the number of induced cells observed after γ-irradiation alone. This increase is statistically significant.

A slight intensification of the inducing action of γ-rays was observed also following a single exposure to vibration 1.5 h before irradiation. However, vibration given 1.5 h after irradiation of the lysogenic culture was ineffective. The number of induced phage particles in this case was almost the same as in the samples exposed to irradiation alone.

It may be concluded from the results described above that vibration increases the sensitivity of lysogenic bacteria to the subsequent action of ionizing radiation, although by itself this factor does not induce phage formation, but on the contrary, inhibits it. This is confirmed by the degree of induced phage production in the samples of

E. coli K-12 (λ) exposed to vibration compared with that in controls not exposed to vibration (see Table 2). The value of R calculated by comparing the results of the experimental and control series is statistically significant.

It may be postulated on the basis of this investigation that the inducing effect discovered after exposure of E. coli K-12 (λ) on the ships "Vostok-3," "Vostok-4," "Vostok-5," and "Vostok-6" was associated with the combined action of the factors of cosmic flight. Exposure to vibration evidently causes sensitization of the lysogenic culture to the action of cosmic radiation, and this causes the appearance of induction of the bacteria after exposure to small doses amounting to several tens of millirads. As previously mentioned, other sources of genetic mutations may be mechanical factors associated with acceleration, and also weightlessness. The solution of these important problems requires special experiments to examine the biological effects of these factors.

SUMMARY

In experiments staged in space ships "Vostok-5" and "Vostok-6" a significant excess over induced phage production of E. coli (3.67 and 1.7 times respectively) was noted.

It was established under laboratory conditions that a three-fold increase in phage production corresponds to the inducing effect of radiation in a dose of 58 R, which is approximately by three orders higher than the physical dose of radiation in an orbital flight. The established inducing effect might be associated with other space flight factors (weightlessness, accelerations, vibration). To verify this supposition laboratory experiments were made in order to ascertain the influence of vibration in the total complex of space flight factors. A culture of E. coli K-12 (λ) was subjected to vibration in pure form or in combination with γ-irradiation (dose of 100 R). Vibration was produced on a vibrostand at a frequency rate of 18, 35, 75, or 700 cps during 15-60 min at overload of 10 g.

It was found that the vibration effect itself did not increase the number of phage particles in experimental samples as compared to the level of spontaneous phage formation. During the influence of vibration and radiation with subsequent vibration the number of induced bacteria was 1.4 times larger than that of induced cells which were observed in the case of γ-irradiation alone.

A slight intensification of the inducing effect of γ-rays was observed also with a single vibration dose 1½ h before irradiation. In vibration applied after irradiation of the culture the number of induced phage particles was almost the same as in the samples subjected to irradiation alone.

The findings obtained indicate that inducing effect in exposure of E. coli K-12 (λ) carried in "Vostok-5" and "Vostok-6" space ships was connected with a complex influence of space flight factors – radiation and vibration.

LITERATURE CITED

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