STIMULATION OF PLANT GROWTH BY EXPOSURE TO LOW LEVEL y-RADIATION AND MAGNETIC FIELD, AND THEIR POSSIBLE MECHANISM OF ACTION

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KUZIN A. M., VAGABOVA M. E., VILENCHIK M. M. and GOGVADZE V. G. *Stimulation of plant growth by exposure to low level y-radiation and magnetic field, and their possible mechanism o faction.* **ENVIRONMENTAL AND EXPERIMENTAL** BOTANY 26, 163-- 167, 1986.--Chronic T-irradiation *ofCucumis sativus* seedlings with a cumulative dose of ~ 0.02 Gy over 7 days stimulated plant growth. A time-varying magnetic field (the strength of the stationary magnetic field varied from 100 G to approximately that of geomagnetic field levels within \sim 3 sec) also stimulated seedling growth of *C. sativus* and *Raphanus rafantinum.* It is suggested that a modulation of free radical reactions in plant cells and tissues, which initiates changes in the structure and function of cell membranes, is responsible for the effects observed. Ionizing radiation induces the formation of free radicals while the magnetic field is postulated to modulate the rate of their recombination during normal metabolism.

INTRODUCTION

PLANT seedlings have been used to study weak effects of various physical factors. We have previously shown^{(6)} that the development of seedlings shielded against natural y-radiation background was delayed. We proposed, therefore, to determine whether a slight increase (by 1-2 orders of magnitude) in the natural radiation background level would produce a stimulatory effect on seedling development. We also proposed to investigate the influence of a time-varying lowintensity magnetic field on seedling growth. In this report, we register growth stimulation produced by both factors and propose a hypothesis of their possible common mechanism of action.

MATERIALS AND METHODS

Seeds of *Cucumis sativus* and *Raphanus rafantinum* were used in our experiments. Seedlings were kept

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under strictly identical conditions (temperature, illumination, and moisture content). A special controlled environment chamber, with an outside γ -radiation source (¹³⁷Cs), was designed (Fig. 1) in which constant temperature $(26 \pm 0.1^{\circ}C)$ and illumination (3 klx, 10 hr/day) were maintained by conventional means. The rate of y-radiation in chamber section 1 was equal to that of the natural radiation background level. The dose rate of yradiation in chamber sections 2, 3, and 4 exceeded that of section 1 by 8, 100, and 400 times, respectively, as measured by a standard ionization counter.

Seeds were soaked overnight in tap water (19 22° C). The seedlings were rolled into filter paper, put into glasses of water and placed into a corresponding chamber section. Seedling growth was measured after 7 days of chronic irradiation with an accuracy of up to 1 mm ; 240 seedlings (60 seedlings per section) were used in each experiment undertaken 7 times.

Fio. 1. Schematic representation of the controlled environment chamber in experiments with different dose-rates (viewed from above). (A) Controlled environment chamber in which all experimental seedlings are placed at constant temperature, moisture content, and illumination. (B) Lead collimator. (C) $137Cs \gamma$ radiation source. (D) Dose-rate regulating screens. (E) Sections with different dose-rates: E_1 , natural radiation background (control); E_2 , E_3 , and E_4 , dose-rates exceeding that of E_1 by 8, 100, and 400 times, respectively. (F) Seedlings under study.

To study the effect of a time-varying magnetic field on plant growth, 2-day soaked seedlings were placed on moistened filter paper in Petri dishes and exposed to a variable magnetic field in the device schematically represented in Fig. 2. The treatment lasted for 1-14 min at room temperature under conditions recommended by Professor Danilov and co-workers⁽⁴⁾. Petri dishes with the

Fio. 2. Schematic diagram of the device used for exposure of seedlings to a timq-varying magnetic field. (I) Continuous tape with constant magnet bars fastened on its surface. (2) Magnet bars of $5 \times 1 \times 0.5$ cm are placed, at a distance of 15 cm from each other, so that their north (N) and south (S) are alternately directed outside. (3) Device providing a uniform movement of the tape at a rate of 1 m/min. (4) Petri dish with seedlings: the distance from the magnet surface is 0.2 cm.

control seedlings were kept under similar conditions, that is, for the same time, at the same temperature and illumination, but at a distance of 2 m from the device; in other words, they were outside the magnetic field. Each passage of a magnet under the Petri dish was considered as one exposure. The magnetic field varied linearly from 100 G to about that of the geomagnetic field within 3 sec, as measured by a standard magnetometer.

The control seedlings and those exposed to the magnetic field were rolled into filter paper, put into glasses with tap water, and placed into a controlled environment chamber at $26+0.5^{\circ}$ C. After 6 days of growth under strictly identical conditions, the lengths of roots and the heights of seedlings were measured; 160 seedlings (4 rolls) were used in each experiment. Statistical significance among regimens was estimated by two methods: a paired comparison test and a nonparametric paired comparison test.⁽¹⁾

RESULTS

The growth indexes of the experimental plants consistently exceeded those of the controls in each of 7 experiments conducted (Table 1). Statistical significance of the differences, as measured by the non-parametric paired comparison test, was P < 0.05. To insure that the growth increases in chamber sections 2, 3, and 4 were associated with chronic y-irradiation and not with some conditions not taken into account, the experiments were repeated using the same chamber without the $137Cs$ - y-radiation source; seedling growth in sections 2, 3, and 4 was the same as that in section 1 (control), as is seen in Table 1. Since the radiation doses applied were small (the cumulative dose delivered for 7 days did not exceed 0.02 Gy) we concluded that the duration of exposure, not the dose, was responsible for the observed growth stimulation. The stimulatory effect was absent in the control experiments when the same cumulative dose was delivered for 10 min. For instance, the main root and epicotyl lengths of the controls were 104 and 44 mm, respectively, and after single exposure to 0.02, 0.01, and 0.005 Gy radiation (which corresponded to a cumulative dose given for 7 days of chronic exposure) they

	Chamber section number						
Growth index		2	3				
Main root length (mm) $\%$ Control (section 1) P	$93.6 + 4.8$ 100	$113 + 3.5$ 120 < 0.01	$111 + 7$ 118 < 0.05	$114 + 6.8$ 121 < 0.02			
Epicotyl length (mm) $\%$ Control (section 1) P	$33 + 4$ 100	$49.3 + 6$ 149 < 0.05	$46.1 + 5$ 139 < 0.1	$47.3 + 6$ 142 < 0.1			

Table 1. Effect of low-level chronic y-radiation on the development of Cucumis sativus *seedlings*

Development of Cucumis sativus *seedlings under similar conditions but without 7 irradiation*

were 108, 105, and 109 mm, and 44, 48, and 47 mm, respectively.

The results of the studies on the effect of a timevarying magnetic field on seedling growth are shown in Table 2. The magnetic field did not influence the morphological development of plants but an increase in the number of exposures

up to 48 resulted in a significant stimulation of plant growth similar to that caused by chronic exposure to very low γ -radiation doses (Table 1). A further increase in the number of exposures did not lead to an increase in the stimulatory effect. With dry seeds treated in the same way no effect on plant growth was noted.

Organism	Number of exposures	Number of experiments in a series	Number of experiments		Differences between the			
			effect	with without stimulatory effect	exposed and control seedlings, $\%$ of control	Standard deviation	t	P
Raphanus rafantinum								
Root length								
measurement	7	10	6	4	-1.1	$+1,0$		
	48	10	θ	10	$+24.1$	± 9.6	2.5°	${<}0.02$
	48	20	$\bf{0}$	20	$+24.3$	$+2.0$	12.	< 0.001
	96	17		16	$+9.2$	$+1.7$	11.3	< 0.001
Epicotyl								
measurement	48	20	$\mathbf 0$	20	$+11.7$	$+1.4$	8.6	< 0.001
Cucumis sativus Root length								
measurement	48	39	ı	38	$+7.5$	$+1.3$		5.8 < 0.001

Table 2. Effect of a time-varying magnetic field on plant growth

DISCUSSION

As was previously shown, (7) the stimulatory effect of chronic low-level radiation is caused by maintaining at an elevated level the concentration of free radicals (of the semiquinone nature, in particular) which are formed in biomembranes and produce a "trigger effect" changing the structure and the lipid bilayer of a membrane. A membrane-bound adenylatecyclase is activated and the level of cyclic AMP increases, leading to gene activation and further stimulation of development.^{(7)} Under normal conditions, free radicals are also formed due to the activity of lipoxygenases and lipid peroxidation in membranes of plant and animal cells, and are known to have a role⁽²⁾ in the manifestation of regulatory properties of membranes. It may also be assumed that, on the one hand, the level of free radicals in biomembranes depends on the rate of their recombination and, on the other hand, the reactions of radical recombination in plant cells are modulated by the magnetic field.⁽⁹⁾

A magnetic field of $100-1000$ G can affect^(5,8) radical recombination due to a change in spin multiplicity (of the initial radical pairs) originated from the hyperfine coupling between the unpaired electron spins and nuclear spins within each radical. The difference in the Zeeman energy⁽⁸⁾ of electrons in a radical pair, i.e. the $g_1 BH-g_2 BH$ difference, where g_1 and g_2 are the values of the g-factors of the unpaired electrons of a radical pair, is another mechanism $(8,10)$ by which a magnetic field may modulate radical recombination. From this it follows that the rate of radical recombination under the effect of a magnetic field may be increased when the first mechanism is predominant, or decreased when the second mechanism prevails. The effect depends on the nature of the radicals and the strength of the field. Under defined conditions, when the rate of $S-T$ conversions of electrons in a radical pair is different, radical recombination under the effect of a magnetic field is less probable. This will inevitably lead to an increase in the level of radicals, normally found in biomembranes, which is similar to the increase caused by low radiation doses, (7) and leads to the acceleration of radical-induced biochemical processes.

In studying the photochemical processes in solid bodies, liquids and photosynthetic sys-

 $tems^(3,8,11)$ it was demonstrated that a magnetic field of the order of 100-1000 G influenced the recombination of radical pairs and a corresponding change in the processes under study. In the present study we noted a pronounced stimulatory effect in actively metabolizing seedlings subjected to the effect of a magnetic field, but not in resting seeds. This observation supported our postulate that natural free radicals stimulate growth.

It follows from the proposed physical mechanism of action of a magnetic field on the level of free radicals that the effect of a magnetic field on sensitive plant seedlings causes stimulation of their growth in a manner similar to that caused by low-level ionizing radiation exposure.

REFERENCES

- 1. BAILEY N. T.J. (1961) *Statistical methods in biology.* The English University Press, London.
- 2. BOUONITSKAYA E. V. (1982) About the physiological biochemical role of lipoxygenases in plant and animal organisms. *Usp. biol. Khim. 22,* 152-158 (in Russian).
- 3. BOXER S. G., CHIDSEY C. E. D. AND ROELOFS M. G. (1982) Anisotropic magnetic interaction in the primary radical ion-pair of photosynthetic reaction centers. *Proe. natn. Acad. Sei., U.S.A.* 79, 4632- 4636.
- 4. GOVORUN R. D., GOLOVACHEV N. A., DANILOV V. I., *et al.* (1982) Some biological effects of magnetic fields. Pages 385-391 *in Materials of the 4tk Meeting on the uses of new nuclear-physical methods in solving scientific, technical and economical problems.* Dubna, Joint Institute for Nuclear Research (in Russian).
- 5. KAPTEIN R. (1971) Page 210 *in Chemically induced dynamic nuclear polarization.* University of Leiden, The Netherlands.
- 6. KUZIN A. M., VAGABOVA M. E. and PRIMAK-MIROLYUBOV V. N. (1977) On the role of natural radiation background in the initial development of plants. *Radiobiologiya* 17, 37-40 (in Russian).
- 7. Kuzin A. M. (1980) Different major molecular mechanisms of action of high- and low-level radiation. *Izv. Akad. Nauk SSSR,* Seriya biologicheskaya *(Biol. Bull. U.S.S.R. Acad. Sci.* 6, 883- 890 (in Russian).
- 8. SAGDEEV R. A., SALIKOV K. M. and MOLIN Yu. N. (1977) Influence of a magnetic field on the processes involving radicals and triplet molecules in solutions. *Usp. Khim. (Adv. Chemi.* **46,** 569-601 (in Russian).
- 9. VILENCHIK M. M. (1983) *Radiobiological effects and environment.* Moscow, Energoatomizdat (in Russian).
- 0. VILENCHIK M. M. (1982) Magnetic susceptibility of rhodopsin molecules. *Biofizika* 27, 31-36 (in Russian).
- 11. VOZNYAK V. M., GANAGO J. B., MOSKALENKO A. A. and ELFIMOV E. J. (1970) Magnetic-fieldinduced changes in fluorescence yield of chlorophyll-protein complexes enriched with photosystem 1. *Studia biophys., Berlin,* pp. 13-20.