Effect of a Long Low-Power γ-Irradiation and β-Carotene on Lipid Metabolism in the Rat Thymocyte Nuclei

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The effect of γ-irradiation in a dose of 1.5 Gy at a dose intensity of 3 sGy/day on the rat thymocyte nuclear lipids and of β-carotene diet on the metabolism of thymocyte nuclear lipids is studied in intact and γ-irradiated rats. The irradiation activated the incorporation of 2-^{14}C-acetate in the total lipid fraction and in cholesterol and suppressed the label incorporation in sphyngomyelin. The β-carotene diet decreased the content of cholesterol and monoglycerides in the thymocyte nuclei of intact and irradiated rats. In irradiated rats β-carotene increased the label incorporation in the total phosphatidylserine+phosphatidylinositol fraction.

Key Words: *chronic γ-irradiation; thymocyte; nuclei; lipids; β-carotene*

A single or prolonged exposure to low-dose ionizing radiation impairs the immunity in mammals [7]. The molecular mechanisms of the effects of low-dose and low-dose intensity ionizing radiation on cellular metabolism in the thymus, the central organ of the immune system, are little known. At present, the attention of scientists is focused on lipid metabolism in cell nuclei and their role in the function of cellular genetic system [1]. Studies of the effects of immunostimulants involved in natural regulation of lipid metabolism, specifically, carotenoids, on the metabolic effects of low-dose radiation, are particularly interesting.

Carotenoids, primarily β-carotene (BC), exhibit immunostimulating [9], antioxidant [11], and provitamin [13] activities. It was reported that BC possesses radioprotective activity [5] which probably results from its antioxidant and immunostimulating properties. In the rodents, intact BC is deposited in many organs [15]. The addition of BC to diets of rats irradiated in a dose of 6 Gy prevents the loss of the thymus weight [2].

We examined the effect of prolonged (the whole life) γ-irradiation of low intensity not affecting the life span and negligibly modifying the hemopoiesis [6] on the metabolism of thymocyte nuclear lipids and the effect of BC-containing diet on nuclear lipid metabolism in rats exposed to γ-radiation.

MATERIALS AND METHODS

Four groups of male Wistar rats weighing 80-90 g were used: 1) intact controls, 2) control+BC, 3) irradiation, and 4) irradiation+BC. Chronic irradiation in a dose of 1.5 Gy at a dose intensity of 3 sGy/day was carried out in a special γ-chamber with ^{137}Cs as the source of radiation. Animals of groups 2 and 4 were fed synthetic BC with curds for 50 days, groups 1 and 3 were fed curds. In a daily dose of 3 mg/kg BC produces an antitumor effect [12]. After a total dose of 1.5 Gy had been attained, the animals were decapitated, the thymuses were removed, and a thymocyte suspension was prepared. The cells were incubated with 2-^{14}C-acetate. The conditions for preparation of thymocytes, their

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TABLE 1. Levels of Neutral Lipids in Thymocyte Nuclei of Intact and γ-Irradiated Rats Fed BC Diets (mg lipid/mg nuclear protein, *M±m*, *n*=3)

Lipid fraction	Control	Control+BC	% of control in paired comparison	Irradiation	Irradiation+BC	% of control in paired comparison
Cholesterol	8.3±1.1	6.2±0.9	75±2.5*	7.3±0.3	5.8±0.5	79±3.7*
Monoglycerides	9.0±1.1	6.7±1.7	73±3.5*	7.5±0.4	5.6±0.2	78±3.9*
Diglycerides	4.2±0.6	3.7±0.3	92±8.0	4.3±0.2	4.1±0.4	96±14
Fatty acids	29.7±3.6	27.4±8.8	88±17	28.1±3.9	34.7±9.9	127±26

Note. *$p<0.05$ *vs.* control.

incubation, isolation of the nuclei, extraction and chromatography of neutral lipids, methods of measuring the amounts and radioactivity of lipids were described previously [3,4]. Phospholipids were chromatographed as reported previously [14].

RESULTS

Chronic γ-irradiation in a dose of 1.5 Gy had no effect on lipid content in the thymocyte nuclei (Tables 1 and 2). Radiation-induced activation of 2-^{14}C-acetate incorporation in the total lipid fraction and cholesterol was observed (Table 3). Previously, we demonstrated activation of cholesterol production in lymphoid cells after acute γ-irradiation in a dose of 4 Gy, which was regarded as a cell response to injury, directed at membrane repair [10]. Activation of the label incorporation in the thymocyte nuclei cholesterol during chronic irradiation in a dose of 1.5 Gy was similar to the effect of acute irradiation in a dose of 4 Gy.

Irradiation affects the production of nuclear phospholipids. Judging from the level of total radioactivity (Table 3), the depression of the label incorporation in nuclear sphyngomyelin (SPM) is negligible. The level of specific radioactivity (55±4 cpm/mg lipid in the control and 70±4% of control in paired comparison) indicates a significant ($p<0.05$) decrease in the label incorporation in SPM. Acute irradiation of rats in a dose of 10 Gy resulted in a significant suppression of 2-^{14}C-acetate incorporation in the thymocyte chromatin SPM [3]. This indicates that both acute and chronic γ-irradiation modify the nuclear SPM metabolism. Chronic irradiation of rats in a dose of 1.5 Gy at a dose intensity of 3 sGy/day seems to trigger the same reactions of thymocytes involving cholesterol and nuclear SPM metabolism as acute sublethal and lethal irradiation.

Administration of BC diets resulted in a significant decrease in the levels of cholesterol and monoglycerides in the thymocyte nuclei (Table 1). The decrease in the content of cholesterol in the nuclei induced by BC diet agrees with publications on the hypercholesterolemic effect of BC [8]. The effect of BC on the levels of cholesterol and monoglycerides was not modified by γ-irradiation, because the content of these lipids in the nuclei of irradiated rats was decreased by BC similarly as in control animals. The decrease in the content of cholesterol in thymocyte nuclei of rats fed BC diets modified the nuclear membrane. This confirms the hypothesis that the protective effect of BC is mediated through alteration of membrane viscosity [13]. The specific effect of BC on metabolism of thymocyte nuclei phospholipids in irradiated rats consists in the development of a tendency toward an increase in the amount of SPM (Table 2) and a significant activation of the label incorporation in the total phosphatidylserin+phosphatidylinositol fraction (Table 3), the most important nuclear lipid regulators.

Changes in cholesterol and SPM metabolism in rats exposed to chronic γ-irradiation in a dose of

TABLE 2. Levels of Phospholipids in Thymocyte Nuclei of Intact and γ-Irradiated Rats Fed BC Diets (mg lipid/mg nuclear protein, *M±m*, *n*=3)

Lipid fraction	Control	Control+BC	Irradiation	Irradiation+BC
Total phospholipid fraction	35.4±3.3	33.1±3.8	39.1±4.9	34.1±3.0
Sphyngomyelin	1.1±0.2	1.3±0.02	1.3±0.01	2.5±0.3
Phosphatidylcholin	14.6±1.5	15.0±1.8	15.4±1.5	18.3±0.9
Phosphatidylserin+phosphatidylinositol	5.9±1.4	3.4±1.4	5.0±1.3	5.8±0.4
Phosphatidylethanolamine	6.5±1.5	4.3±0.6	6.1±2.1	5.9±0.7
Cardiolipin	1.1±0.3	1.4±0.2	1.4±0.3	2.1±0.4

TABLE 3. Effect of γ-Irradiation and BC on 2-^{14}C-Acetate Incorporation in Rat Thymocyte Nuclear Lipids ($M\pm m$, n=3)

Lipid fraction	Control	Control+BC	Irradiation	Irradiation+BC
Total lipid fraction	5550±830	7460±1730	7320±1590*	5050±550
Cholesterol	139±4	437±254	221±7**	344±186
Monoglycerides	634±30	796±142	686±96	477±79
Diglycerides	42±13	48±10	41±7	36±6
Fatty acids	137±6	159±22	186±49	157±26
Total phospholipid fraction	1800±290	1590±260	1680±30	1870±220
Sphyngomyelin	61±5	91±16	49±1	76±10
Phosphatidylcholin	1010±43	872±152	868±62	1370±200
Phosphatidylserin+phosphatidylinositol	102±12	103±3	96±12	141±3***
Phosphatidylethanolamine	176±8	168±8	165±11	179±10
Cardiolipin	23±5	21±5	20±9	15±6

Note. $p<0.05$: *vs. control (130±4%) in paired comparison; **vs. control; ***vs. control+BC.

1.5 Gy indicate a high sensitivity of thymocyte nuclear lipids to irradiation. Cholesterol and SPM metabolism are the most sensitive targets of γ-irradiation and BC.

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