In conclusion, we have shown that augmented TNF production correlates with increased lethality among mice with endotoxin shock after their sensitization with high-molecular-weight substances contained in the sera of tumor-bearing mice. Such TNF hyperproduction is shown by cells of the macrophage lineage from sensitized animals. Intact animals injected with sera from tumor-bearing mice become more susceptible not only to the toxic activity of LPS and MDP but also to that of the recombinant TNF. With the substances contained in the sera of tumor-bearing animals, hypersensitivity to LPS and MDP resulting in TNF hyperproduction can therefore be transferred to macrophagal elements of intact animals and, in addition, the sensitivity of the body systems acted upon by the TNF itself can be increased in these animals.

LITERATURE CITED

1. A.L. Rakhmilevich and M.S. Rakhimova, Byull. Eksp. Biol. Med., No. 4, 483-485 (1988).
2. B.B. Fuks, A.L. Rakhmilevich, A.A. Pimenov, et al., Byull. Eksp. Biol. Med., No. 10, 497-499 (1987).
3. B.B. Fuks, A.I. Shapoval, and I.M. Grzhebin, Byull. Eksp. Biol. Med., No. 7, 78-80 (1991).
4. J. Bartholeyns, M. Freudenberg, and C. Galanos, Infect. Immun., **55**, 2230-2233 (1987).
5. M.J. Berendt, M.F. Newborg, and R.J. North, Infect.Immun., **28**, 645-647 (1980).
6. J.W.B. Bradfield, T. Whitmarsh-Everiss, and D.B. Palmer, Brit. J. Cancer, **42**, 900-907 (1980).
7. E. Carswell, L. Old, R. Kassel, et al., Proc. Nat. Acad. Sci. USA, **72**, 3666-3670 (1975).
8. H. Fisch and G.E. Gifford, Int. J. Cancer, **32**, 105-112 (1983).
9. G.E. Grew, J.E. Teylor, D.O. Trop, et al., New Engl. J. Med., **320**, 1586-1591 (1989).
10. J.A. Krosnic, J.K. McIntosh, J.J. Mule, et al., Cancer Immunol. Immunother., **30**, 133-138 (1989).
11. M.M. Mustafa, O. Ramilo, X. Saer-Llorens, et al., Pediat. Infect. Dis., **8**, 921-922 (1989).
12. K. Nara, H. Odagiri, M. Fujii, et al., Cancer Immunol. Immunother., **25**, 126-132 (1987).
13. A.L. Rakhmilevich and R.J. North, Cytokine, **3**, 398-406 (1991).
14. E. Suter, G. Ullman, and R. Hoffman, Proc. Soc. Exp. Biol., **99**, 167-169 (1958).
15. T. Takashima, C. Ueta, I. Tsuyugchi, et al., Infect. Immun., **58**, 3286-3292 (1990).

0007-4888/93/0001-0063$12.50 ©1993 Plenum Publishing Corporation

Mammary Carcinogenesis Suppression by Ginseng Tissue Culture Biomass Tincture

V.G.Bespalov, V.A.Aleksandrov, V.V.Davydov, A.Yu.Limarenko, D.S.Molokovskii, A.S.Petrov, L.I.Slepyan, and Ya.G.Trilis

UDC 618.19.006.6.092:615.015.89

Translated from *Byulleten' Experimental'noi Biologii i Meditsiny*, Vol. 115, No. 1, pp. 59-61, January, 1993
Original article submitted June 30, 1991

Key words: *Bioginseng, mammary carcinogenesis*

Much attention has been paid of late to search for agents capable of suppressing carcinogenesis, since such agents appear to be promising in the primary prevention of cancer [1]. Some authors have reported that a natural ginseng root tincture inhibited the development of pulmonary adenomas induced by various carcinogens in mice [5,17]. An epidemiologic survey from South Korea, where ginseng preparations are widely used, has demonstrated by the chance control method that patients with malignancies use ginseng much more seldom than control patients with nononcologic diseases [16]. Hence, further research of the oncoprophylactic or anticarcinogenic characteristics of ginseng seems to hold promise.

The present research was aimed at experimental study of ginseng potentialities in the prevention of mammary carcinoma. Bioginseng, an officinal drug (Provisional Pharmacopeial Article No 42-1890.89 as of April 21, 1989), obtained from cultured cells of Panax ginseng C.A.Mey, was used in the study. The cultural bioginseng preparation is characterized by the same pharmacologic activity as natural ginseng root galenics [4], but it is more readily available for clinical practice.

Laboratory for Preclinical Trials, N.N.Petrov Research Institute of Oncology; Department of Physiology, Chemical Pharmaceutical Institute of the Ministry of Health of the Russian Federation, St.Petersburg
(Presented by Academician N.P.Bekhtereva, of the Russian Academy of Medical Sciences)

MATERIAL AND METHODS

Experiments were carried out on male Wistar rats. The carcinogen, N-methyl-N-nitrosourea (MNU), was synthesized by methods described previously [9]. Bioginseng tincture manufactured by the Kirish Biochemical Plant was used. Rats initially weighting 200-300 g were injected with MNU in the tissue of all 12 mammary glands (1 mg in 0.1 ml of normal saline per gland). One week after injection the rats were divided into 2 groups. The experimental animals were fed bioginseng 5 times a week at 2-day intervals orally via a tube in a dose of 0.5 ml per rat daily (2.5 ml/kg) till the end of the experiment. The agent was dealcoholized in vacuum rotor evaporizer directly before administration, and then diluted with water to the initial volume. The control animals were administered water orally via a tube, according to the same scheme. Intact control rats weighting 200-300 g were exposed to neither.

At the end of the experiment blood serum estradiol was radioimmunoassayed by a CIA-TRI (Sorin, Italy) kit with a Beckman (USA) γ-scintillator. Twenty-eight weeks after the start of the experiments the animals were killed by ether vapours and a complete autopsy was carried out. The tumor tissue was histologically treated according to standard methods with hematoxylin-eosin staining and examined under a light microscope. The results were statistically processed using the t, χ^2 , and U tests.

RESULTS

MNU induced multiple mammary tumors in the rats, mostly malignant ones (Table 1). Bioginseng reduced the incidence of these tumors by 44% and their multiplicity by 62% as against the control animals. Moreover, 22% of the MNU-injected controls developed malignant renal tumors, by 19 and 68%, respectively. The intact controls developed neither mammary nor renal tumors (Table 1).

Bioginseng had a marked inhibitory effect on mammary tumors induced by MNU injections in rats.

Table 2. Bioginseng Effect on Blood Serum Estradiol Level (M±m) In parentheses number of rant examined.

Group	Estradiol levels, 1/nM		
	in total group	in rats with mammary tumors	in rats without tumors
Intact controls	2,50±0,20 (12)	-	-
MNU+control	4,20±0,67* (23)	3,98±0,49 (17)	5,20±2,30 (6)
MNU+bioginseng	3,70±0,50* (18)	5,08±1,20 (6)	3,03±0,70** (12)

Note: In parentheses number of rats examined. * - difference from intact controls is reliable according to the t test ($p<0.05$); ** - difference from rats with tumors fed bioginseng is reliable according to the U test ($p<0.05$).

Adaptogenic [4] and immunostimulating [6] effects of ginseng were revealed. The ginseng preparations were able to normalize various hormonal and metabolic disturbances promoting tumor development; they enhanced the DNA repair processes [7] and showed interferonogenic [8], cytodifferentiating [10,14], and antioxidant [13] activities. The preparations were found to elevate the level of intracellular cyclic AMP [15] and to prevent the manifestation of biologic effects of crotonic acid, a carcinogenesis promoter [11]. These versatile mechanisms of action seem to underlie the revealed anticarcinogenic activity of bioginseng.

Table 2 shows that blood serum estradiol levels of rats exposed to MNU were significantly higher than in intact animals. This may be due to a reduced number of specific receptors in target organs in the presence of augmenting morphologic and functional dedifferentiation of cells, resulting from carcinogen action. The experimental and clinical findings demonstrate that an increased blood estrogen level is conductive to the development of mammary carcinoma [2]. Rats without tumors that were fed bioginseng had lower blood serum estradiol levels than the intact animals or the

Table 1. Bioginseng Effect on MNU-Induced Carcinogenesis in Rats.

Group	Number of rats in group	Absolute number and % of rats with tumors, number of tumors, and mean number of tumors per rat		
		all tumors	mammary tumors*	renal tumors**
MNU (control)	27	21 (78%)	21 (78%)	6 (22%)
		49	42	47
		1,8±0,2	1,6±0,2	0,3±0,1
MNU+bioginseng	29	10 (34%)****	10 (34%)****	1 (3%)****
		19***	17	1
		0,7±0,1****	0,6±0,1****	0,03±0,05****
Intact control	20	-	-	-

Note: *histologically verified adenocarcinomas, fibroadenomas in a few cases; **mesenchymal tumors; ***adenocarcinoma of the large intestine was found in one rat; **** the difference with the control MNU group is statistically reliable according to the t and χ^2 tests ($p<0.001$-0.05).

animals with tumors that were fed bioginseng (Table 2). Thus, bioginseng normalizes the blood estradiol level, this possibly being one more mechanism explaining mammary carcinogenesis inhibition by this drug.

Our finding demonstrate a new indication for the use of bioginseng tincture: it can be used for the primary prophylaxis of mammary carcinoma. Breast cancer is the most prevalent oncologic disease in women [3]. Antiestrogens, recommended for the prevention of mammary carcinoma, induce a number of noticeable side effects [3,12].

LITERATURE CITED

1. V.A.Aleksandrov and V.G.Bespalov, Vopr.Onkol., **37**, No. 4, 387-393 (1991).
2. V.M.Dilman, L.M.Bershtein, E.V.Tsyrlina, and S.Yu.Revskoi, Hormones in Experimental and Clinical Oncology. [inRussian], Moscow, VINITI (1990), **20**, Ser.Onkol., p. 113-139.
3. D.G.Zarigze, E.E.Yakovleva, Vopr.Onkol., **35**, No. 5, 519-528 (1989).
4. A.N.Kudrin, F.P.Krendal', I.K.Sokolov, et al., Farmatsiya, **31**, No. 5, 33-38 (1982).
5. N.V.Lazarev, Vopr.Oncol., **11**, No. 12, 48-54 (1965).
6. V.N.Chubarev, E.R.Rubtsova, I.V.Filatova, et al., Farmakol. i Toksikol., **52**, No. 2, 55-59 (1989).
7. K.V.Yaremenko, Adaptogenes: Perspektive Medical Agents. [inRussian] Izd.Tomsk.Univ., Tomsk, 96 (1990).
8. P.Devaleou, P. Lallouette, and A.M.Tessier, Planta Med., **40**, 49-54 (1980).
9. H.Druckrey, R.Preussmann, S.Ivankovic, and D.Schmahl. Z.Krebsforsch., **69**, 103-201 (1967).
10. X.-R.Jiang and H.-X.Lin, Anticancer Res., **10**, 1489 (1990).
11. W.-X.Juan, L.-H.Gui, J.-L.Zhou, et al., Acta Pharmacol.Sic., **4**, 124-127 (1983).
12. R.R.Love, J.Nat. Cancer Inst., **82**, 18-21 (1990).
13. S.Nakagawa, S.Joshida, J.Hirao, et al., Hiroshima J.Med.Sci., **34**, 303-309 (1985).
14. T.Ota, K.Fujikawa-Yamamoto, Z.-P.Zong, et al., Cancer Res., **47**, 3863-3867 (1987).
15. M.Yang and B.-X.Wang, Acta Pharmacol.Sin., **12**, 272-275 (1991).
16. T.K.Yun S.Y.Choi, Int.J.Epidemiol., **19**, 871-876 (1990).
17. Y.S.Yun, S.K.Jo, H.S.Moon, et al., Cancer Detect.Prevent., **10**, Suppl.1, 301-309 (1987).

0007-4888/93/0001-0065$12.50 ©1993 Plenum Publishing Corporation

MORPHOLOGY AND PATHMORPHOLOGY

Peculiarities of the Relief of the Mineralized Surface of Lacunae and Canaliculi in Lamellar Bone

A. A. Doctorov and Yu. I. Denisov-Nicol'skii

UDC 616.71-008.9-07

Translated from *Byulleten' Experimental'noi Biologii i Meditsiny*, Vol. 115, No. 1, pp. 61-65, January, 1993
Original article submitted July 15, 1992

Key Words: *bone tissue; lacunar-canalicular system; bone mineral crystals*

The term "bone fluid," whose circulation is observed mainly in the lacunar-canalicular system, is used at present in a number of morphological and physiological investigations of bone. In fact, the term is used to designate an interstitial fluid of bone which differs from blood plasma in its composition [4]. According to the Arnold-Frost model [7], during the blood-and-bone exchange the phenomenon of "bone fluid" filtration through the mineralized matrix is of prime importance. The extremely large surface of bone mineral crystals, even with a comparatively low flow of the fluid, provides a quick and effective equilibration of the Ca^{2+} and phosphate concentrations between the fluid and the mineral [6]. To determine the nature of the circulation processes in the mineralized bone matrix the microcorrosion method [2, 5] and various markers are used [6].

Department of Experimental Morphology, Resarch Center of Biological Structures, Moscow