

# Analysis of antimutagenic effect of a compositional preparation made from extracts of green tea leaves and Caucasian persimmon fruits

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## Abstract

Antimutagenic activity of a compositional preparation produced from extracts of green tea leaves and fruit of Caucasian persimmon was determined on the cell culture of human peripheral blood lymphocytes with modification of a mutation process induced by ionizing radiation and nitrosoguanidine. Antimutagenic efficiency of the preparation composites increases when the induced mutability is modified by factors of various physical and chemical nature. The comparative evaluation of their antimutagenic efficiency showed high antimutagenic activity of the composites of the preparation produced from extracts of green tea leaves and Caucasian persimmon fruit. Therewith the strength of this effect is dependent on dose intensity.

**Key words:** lymphocyte, mutagen, antimutagen, chromosome aberrations

## Introduction

Increase of the resistance of organisms to environmental pollution has become one of the challenges facing biological science. This issue is closely related to the accumulation of mutagenic and carcinogenic factors of different origin in the biosphere (Von Borstel *et al.*, 1996). These main factors cause the growing of mutational load of biological species and human body, including frequency of genetically determined diseases, oncological and other etiology diseases, and acceleration of aging (Durnev *et al.*, 1993).

Since the amount of contaminants in the biosphere increases and their mutagenic and carcino-

genic traits are revealed, this problem approaches its turning point. Furthermore, it is referred to irreversible environmental impact and that's why legislation measures of its prevention are insufficient nowadays. There are different protection methods against these factors though.

The development of theoretical and practical points of antimutagenesis is considered to be one of the real models of increasing organism genetic resistance, since it's based on universal effective and physiologic preparations which are capable to reduce mutation rate in essential extent within existing environmental mutagen level (Agabeyli *et al.*, 2006). Previous intensive researches have shown antimutagenic and anticarcinogenic efficiency of many extracts and endogenic metabolites originated from different natural origins, including high-polyphenol vegetable products and some vitamins (Huseynov *et al.*, 2005).

It's also determined that antimutagenic efficiency of the mixture exceeded that of its separate components. According to existing data, fruits of Caucasian persimmon are rich in tannins (61%) and vitamin C (Colborn *et al.*, 1996). Tea and its polyphenols are also considered to be high-effective antimutagens and anticarcinogens (Alekperov *et al.*, 1984).

Two factors are responsible for combining compositional preparations: first, high antimutagenic property of compositional preparation constituents; then enough storage compounds in each of the two constituents, respectively, and their wide practical use must be taking into consideration. In case of high-efficiency of both these factors providing compositional preparations (in comparison to their separate components) there are vast opportunities for

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gaining their advantages. The investigations made on human peripheral blood lymphocytes are necessary in order to take their wide practical use. Considering all of the above, antimutagenic effect of the compositional preparation produced from green tea extracts and Caucasian persimmon fruit was studied using human peripheral blood lymphocytes.

## Materials and methods

The experiments were carried out on peripheral blood lymphocytes of practically healthy donors. Ionizing radiation – gamma-rays (GR) at a dose 2.92 Gy (tele-gamma-therapeutic apparatus Rokus-M 60-Co, dose capacity 48.6 Gy/min) were used as physical mutagens, such a chemical as alkylating mutagen of direct action N-methyl-N'-nitro-N-nitrosoguanidine (MNNG) at concentration 5.5 mmol was used. During the experiments on cultures of human blood lymphocytes the analysis of chromosome aberrations was done in the metaphase (Moorched *et al.*, 1960).

As a mutation modifier we used compositional preparation produced from green tea leaves and fruit of Caucasian persimmon cultivated in the south of Azerbaijan.

According to previous experiments (Huseynov, 2009), green tea extract dose within mixture made 0.01 mcg/ml whereas another compound, i.e. Caucasian persimmon extract doses made 0.001, 0.01 and 1.0 mcg/ml.

The data received during the experiment were analyzed with the help of mathematical statistic methods (Lakin, 1990). Antimutagenic efficiency index (AEI) was determined as the difference between initial and modified levels of mutability divided by the initial one (Aleksperov, 1984).

## Results and discussion

Experimental results are given in Tables 1, 2. The data show that both MNU and SF used as inducers of mutations had high mutagenic efficiency. Considering given data it has been observed that chromosome aberration rate in human peripheral blood lymphocytes increased from  $1.93 \pm 0.96\%$  to  $10.50 \pm 2.17\%$  after MNU treatment, and up to  $11.23 \pm 2.21\%$  after SF.

Adding of antimutagenic preparation to the experiment resulted in significant decreasing of chromosome aberration rate. It must be noted that such

efficiency directly related to proportional ratio of the antimutagenic preparation constituents. The highest efficiency was observed when using 0.001 mcg/ml of green tea extract and 0.01 mcg/ml of Caucasian persimmon fruit. It concerned both mentioned mutagens. With treating by MNU, AEI equaled 0.53 r.u. and after SF it made 0.55 r.u. While changing proportional ratio of Caucasian persimmon fruit extract, general antimutagenic efficiency of the composition decreased as well. Discrepancy of proportions of compositional components is more than likely responsible for it. Nonetheless, in spite of the reducing of antimutagenic activity, statistically significant difference was performed in all the experimental variants. The comparative analysis of AEI in variants showed composition efficiency of 40% and 34% in proportional ratio of the constituents 0,01+0,001 mcg/ml, 48% and 42% in 0,01+0,1 mcg/ml, and 27% and 25% in 0,01+1,0 mcg/ml, respectively.

In experiments such a high antimutagenic activity of extracts is directly dependent on content of bioactive substances. Green tea leaves are known to contain catechine, tannin, tanstuffs, different vitamins and other similar bioactive substances. Caucasian persimmon fruits are rich in vitamins C and B, such microelements as zinc, copper, iron, chrome, etc. Thus, we conclude that the mixture of these substances causes antimutagenic activity of given extract composition.

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**Table 1. Chromosome aberration rate in human peripheral blood lymphocytes treated by MNU.**

Experiment variants	Extract from green tea leaves, mcg/ml	Extract from CP fruit, mcg/ml	Chromosome aberration rate, %			Antimutagenesis efficiency factor
			M±m	td	P	
Control	-	-	1.93±0.96	-	-	-
MNU	-	-	10.50±2.17	-	-	-
Extracts	0.01	0.001	6.34±1.70	2.13	<0.01	0.40
	0.01	0.01	4.95±1.53	3.07	<0.001	0.53
+ MNU	0.01	0.1	5.50±1.61	2.67	<0.01	0.48
	0.01	1.0	7.69±1.84	1.35	>0.05	0.27

**Table 2. Chromosome aberration rate in human peripheral blood lymphocytes treated by SF.**

Experiment variants	Extract from green tea leaves, mcg/ml	Extract from CP fruit, mcg/ml	Chromosome aberration rate, %			Antimutagenesis efficiency factor
			M±m	td	P	
Control	-	-	1.93±0.96	-	-	-
SF	-	-	11.23±2.21	-	-	-
Extracts	0.01	0.001	7.46±1.85	1.81	<0.05	0.34
	0.01	0.01	5.03±1.55	3.41	<0.001	0.55
+ SF	0.01	0.1	6.50±1.74	2.38	<0.01	0.42
	0.01	1.0	8.37±1.94	1.32	>0.05	0.25