## Total Antioxidant Capacity of Blood Plasma from Healthy Donors Receiving Vitamin and Mineral Complex

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Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 137, No. 5, pp. 524-526, May, 2004 Original article submitted December 29, 2003

The total antioxidant capacity (TEAC assay) of blood plasma from healthy donors was studied before and after 2- and 4-week consumption of the vitamin and mineral complex containing the major antioxidant vitamins and cofactors of antioxidant enzymes. Before the treatment the donors were divided into 2 subgroups with initially high and low total antioxidant capacity. After 2-week course of vitamin and mineral complex the total antioxidant capacity considerably decreased in group 1 donors, but increased in group 2 donors. After 4-week course this index increased in donors of both subgroups and slightly surpassed the baseline level typical of individuals with high total antioxidant capacity.

**Key Words:** healthy donors; blood; antioxidant capacity

Modern notions of the etiological and pathogenetic role of oxidative stress determine high interest in clinical and preventive treatment, search, and study of antioxidants [8].

There are a variety of methods for measuring antioxidant activity. Previous experiments evaluated antioxidant activity of natural and synthetic compounds [1,2,4,7,8,10]. However, their effect on the system of antioxidant protection is poorly understood. These studies characterized only changes in one or several endogenous antioxidants. Until recent times it was difficult to evaluate the total antioxidant capacity (TAC) of the organism due to the absence of reliable methodical approaches. Specific diet-related and/or nutriceuticproduced changes in TAC remain practically unknown [3,5-7,9,11].

Here we studied TAC of blood plasma from healthy donors receiving a vitamin and mineral complex

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(VMC) for 2 and 4 weeks. The complex included the major antioxidant vitamins and cofactors of antioxidant enzymes.

## MATERIALS AND METHODS

We examined 15 healthy donors (8 women and 7 men) at the age of 32.5±8.9 years (women, 32.4±8.2 years; men, 32.7±10.3 years). They continued to receive a conventional diet in various periods of observations.

Cubital blood samples from fasting donors were placed in standard tubes with heparin (Vacutainer). The plasma and formed elements of the blood were separated by centrifugation on a Beckman J-6 centrifuge. The plasma was stored at -18°C for 14 days.

The blood was taken 3 times: before and after consumption of VMC for 2 and 4 weeks. VMC was given in a daily dose of 2 tablets in the morning and evening.

Treatment with 2 tablets of VMC provided vitamins, microelements, and other biologically active substances (percent of recommended daily dose is shown in brackets): vitamin A,  $\beta$ -carotene, 12,000 U (240%); vitamin C, 90 mg (150%); vitamin E, 100 U (333%);

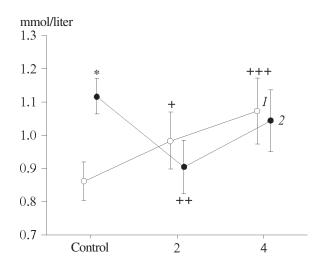
vitamin  $B_1$ , 20 mg (1333%); vitamin  $B_2$ , 20 mg (1176%); vitamin  $B_5$ , 30 mg (300%); vitamin  $B_6$ , 12 mg (600%); vitamin  $B_{12}$ , 100 µg (1666%); vitamin D, 600 U (150%); vitamin D, 20 µg; niacin amide, 50 mg (250%); biotin, 60 µg (20%), folic acid, 800 µg (200%); chromium, 13 µg; calcium, 300 mg (30%); copper, 2 mg (100%); iron, 8 mg (45%); magnesium, 100 mg (25%); molybdenum, 50 µg; nickel, 2.5 µg; potassium, 60 mg; selenium, 40 µg (80%); silicon, 20 µg; vanadium, 20 µg; manganese, 3 mg; phosphorus, 230 mg (23%); zinc, 15 mg (100%); spirulina; bee-queen milk; chlorella; and alphalpha (50 mg).

TAC was estimated in the Trolox equivalent antioxidant capacity assay (TEAC, Randox Laboratories Ltd.) [10]. Laboratory procedures were performed according to manufacturer's recommendations. Spectrophotometry was conducted on a Stat Fax 1904 Plus photometer.

The results were analyzed by parametric and non-parametric tests.

## **RESULTS**

Under control conditions TAC was far below the reference limit typical of West Europeans and derived in studies of the commercial set (1.28-1.83 mmol/liter) [10]. This disagreement can be related to considerable dietary differences between West Europeans and Russians. The latter people are characterized by low supply with antioxidant nutrients [3].



**Fig. 1.** Antioxidant capacity of blood plasma from healthy donors of groups 1 (1) and 2 (2). Data are represented as  $M\pm95\%$  confidence limit. Abscissa: period of treatment with the vitamin and mineral complex (weeks). Ordinate: total antioxidant capacity. \*p<0.001 compared to group 1; \*p<0.05, \*p<0.005, and \*p<0.001 compared to the control.

VMC containing considerable amounts of antioxidant vitamins and cofactors of antioxidant enzymes had no effect on the mean index of plasma TAC in various periods of observations (Table 1). Our results are consistent with published data that peroral administration of dietary antioxidants does not modify TAC in the plasma from healthy donors (TEAC and other assays) [5,6,11].

TABLE 1. Plasma TAC in Healthy Donors (mmol/liter, M±SEM)

Donor, No.		Control	2nd week of consumption	4th week of consumption
Group 1	4	0.93	0.93	1.06
	7	0.92	0.99	1.06
	9	0.83	0.95	1.02
	12	0.90	1.01	1.07
	13	0.88	1.12	1.06
	14	0.79	0.87	1.17
	15	0.78	1.01	1.07
Total index (n=7)		0.860±0.023	0.960±0.033	1.070±0.018
Group 2	1	1.08	0.64	0.94
	2	1.22	0.91	1.08
	3	1.25	0.85	0.97
	5	1.07	0.95	1.12
	6	1.12	1.05	0.86
	8	1.03	0.91	0.97
	10	1.12	1.01	1.39
	11	1.05	0.92	1.02
Total index (n=8)		1.120±0.027	0.900±0.044	1.040±0.057
<b>Total</b> ( <i>n</i> =15)		0.998±0.038	0.930±0.028	1.060±0.031

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It should be emphasized that donors were divided into 2 groups. In group 1 donors the baseline level of TAC was high, but decreased by the end of 2-week treatment with VMC. Group 2 donors had low baseline level of TAC, which increased after consumption of VMC for 3 weeks (Fig. 1). This method was sufficiently sensitive to detect significance differences between the baseline level of TAC in group 1 and 2 donors. By the end of 2-week treatment TAC significantly decreased in group 1 donors, but increased in group 2 donors. No between-group differences in TAC were revealed after consumption of VMC for 2 weeks.

Further observations showed that TAC was similar in group 1 and 2 donors receiving VMC for 4 weeks. In this period, TAC slightly surpassed that observed in group 1 donors before VMC course.

Our results indicate that other investigators made a hasty judgment on low sensitivity of the method for measurement of plasma TAC [10]. Previous studies revealed no changes in the test indexes under the influence of exogenous antioxidants. Most likely, administration of antioxidants in excessive amounts produces complex changes in TAC. We found that TAC decreases, but not increases in some donors receiving antioxidants. Changes in TAC produced by exogenous

antioxidant complexes of different qualitative and quantitative composition should be studied in details.

This work was supported by the Russian Foundation for Basic Research (grant No. 03-04-48591a).

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