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Original Article

Impact of Beta carotene and antioxidant minerals supplementation in combination on the exercise induced oxidative stress in sedentary females.

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Abstract

Background: Physical exercise may be associated with a 10-20 fold increase in whole body oxygen uptake. Oxygen flux in the active peripheral skeletal muscle fibers may increase by as such 100 to 200 fold during exercise. Studies during the past 2 decades suggest that during strenuous exercise, generation of reactive oxygen species (ROS) is elevated to a level that overwhelms tissue antioxidant defense systems. The result is oxidative stress. The present study aims to investigate the effect of supplementation of beta carotene, minerals and antioxidants in combination on the endogenous antioxidant system of the body (viz., glutathione peroxidase GP_X, super oxide dismutase SOD and catalase CAT) of hundred females of 18-21 years age, and comparable height, and weight. **Methods:** Endurance capacity (min) of each subject was determined through exercise on a Magnetic Break Bicycle Ergometer at a fixed workload of 600 kgM/min till exhaustion. GP_X, SOD and CAT levels were analyzed at pre and post exercise levels. The subjects (N=100) were divided into two groups – control group (n=50) receiving placebo supplementation and the experimental group (V_{M+Se}) receiving supplementation of beta carotene and antioxidant minerals combinedly in capsular form (n=50) for 15 days. The same experimental procedure was repeated after supplementation. **Results:** Results indicate that significant increase in endurance capacity ($P<0.001$) and post exercise significant decrease in GP_X ($P<0.05$) and SOD ($P<0.001$) and no significant changes in CAT ($P>0.05$) after beta carotene and antioxidant minerals combinedly supplementation. **Conclusion:** Beta carotene and antioxidant minerals seem to be beneficial to control oxidative stress and enhances endurance capacity.

Key Words: Exercise, female, endurance capacity, beta carotene and antioxidant minerals, antioxidant enzymes

Introduction

Free radical is any species of various groups of atoms capable of independent existence, and contains one or more unpaired electrons. This makes the species highly reactive, such as superoxide (O_2^-), singlet O_2, H_2O_2, peroxide anions and hydroxyl radicals.

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There highly reactive free radicals of oxygen species (ROS) are deleterious to our body.[1] Oxidative stress is the result of an imbalance between the generation of reactive oxygen species (ROS) and their elimination by antioxidant mechanisms.[2]

The production of free radicals can be countered by natural defense system present within the cell. These are antioxidant scavenger enzyme such as catalase (CAT), glutathione peroxidase (GP_X), superoxide dismutase (SOD).[3]

Studies reported an adequate intake of vitamins and minerals through a varied and balanced diet remains the best approach to maintain an optimal antioxidant status. Antioxidant supplementation may be warranted in particular conditions, when athletes are exposed to high oxidative stress or fail to meet dietary antioxidant requirements.[4]

In this particular study, female subjects were chosen, where estrogen plays a crucial role as an antioxidant hormones.[5] The study aims in assessing of beneficial effects if any, of Beta- carotene and antioxidant minerals supplementation on exhaustive exercise induced oxidative stress in females.

Aims and Objectives:

The aims of the study are to reveal:

1. The correlation between the immediate effect of physical exercise on the enzymatic antioxidant system and the first hand effect of beta carotene and antioxidant minerals supplementation in combating exercise- induced oxidative stress.
2. Whether any improvement in endurance (min) at all, occurs after supplementation.

Materials and Methods

Ethical Clearance: This study was conducted in Index Medical College, Hospital and Research Center, Department of Physiology. Ethical clearances were obtained from the Institutional Ethical Committee before carrying out the study.

The entire study procedures were explained and written consent was taken from each volunteer, individually for doing the study. Consent was also taken for publishing this article.

Hundred (N=100), physically fit (no cardiorespiratory disorder, no menstrual disorder) , females of Index Medical College, Hospital and Research Center students of comparable height and weight and aged 18 to 21 years were selected for the study. The subjects were subdivided into two groups – the first group (n=50) served as the control with placebo supplementation for 15 days and the second group was experimental group (V_{M+Se}) (n=50) was given beta carotene and antioxidant minerals (Oxidon Plus, by Micro Labs) supplementation in capsular form for the same period of time. The subjects had no history of any major disease, were not undergoing physical conditioning training. The composition of the Beta carotene, minerals and antioxidants capsules (Oxidon Plus by Micro Labs) was as follows:

Manganese 2.0mg + Zinc 27.5 mg + Beta carotene 30.0mg+ Copper 1.0mg+ L-Selenium 200mcg

Before the actual experiment, the details of the experimental procedure were explained to the subjects to allay apprehension. They were asked to refrain from eating at least for an hour prior to the test and allowed to take complete rest for half an hour before the actual experiment, so that the heart rate could settle to a constant value.

Before the actual exercise, height(Cm), weight(Kg), oral temperature (in°F by clinical thermometer) and blood pressure(mmHg) by sphygmomanometer (Auscultatory method) and pre- exercise heart rate by feeling carotid artery pulsation in beats/min were recorded. Endurance capacity (min) of each subject

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was determined through exercise on a Magnetic Break-Bicycle Ergometer with a fixed workload of 600KgM/min till exhaustion.

At the end of exercise, endurance capacity, peak heart rate, recovery heart rate upto 30 minutes of recovery heart rate upto 30 minutes of recovery period were recorded. The entire procedure was repeated on each subject before and after beta carotene, minerals and antioxidants supplementation.

Blood (5ml) was collected from the ante cubital vein before and after exercise for the determination of CAT[6], GP_X[7], SOD[8]. Blood Hb[9] was determined only before exercise.

The room temperature varied between 20°C- 24°C and the relative humidity was about 77%.

Statistical Analysis:

A two-tail 't' test by difference method was asked for testing the significance of difference between the sample means.

Results:

The physical parameters of the female subjects are shown in Table 1.

Endurance capacity (min) of the exercising female showed a significant increase ($P<0.001$) after beta carotene and antioxidant minerals supplementation, as shown in Table 2.

1st, 2nd, 3rd and 4th min recovery heart rates showed significant ($P<0.001$) decrease after beta carotene and antioxidant minerals supplementation, as shown in Table 3. Recovery of heart rate after exercise is a good indicator of physical fitness and faster the recovery is, the better fitness is reflected.

There were insignificant effects of beta carotene plus antioxidant mineral supplementation on resting hemoglobin concentration, as shown in Table 4.

In Table 5., it was found that the exercise caused a significant ($P<0.05$) decrease in CAT level, significant increase in serum SOD ($P<0.001$) and significant increase in GP_X level ($P< 0.05$) before beta carotene and antioxidant minerals supplementation. But this decrease in CAT level did not change significantly ($P>0.05$) after supplementation of beta carotene and antioxidant minerals supplementation. On the other hand GP_X ($P<0.05$) and SOD ($P<0.001$) was significantly changes after exercise after beta carotene and antioxidant minerals supplementation for 15 days (Table 5.).

Discussion:

Free radicals, produced in endurance exercise have lethal effects in our body, causing loss of membrane fluidity, its natural function and membrane integrity. These reactive oxygen species (ROS) increase red cell deformability and promotes hemolysis of erythrocyte by oxidative damage.[10] This causes the reduction in the activity of the antioxidant enzyme CAT.

The study reflects that Beta carotene, minerals and antioxidants is highly effective in combating exercise- induced oxidative stress as the study shows that Beta carotene, minerals and antioxidants is highly effective in reducing the change (increase in GP_X level, increase in SOD level) in the endogenous antioxidant enzymatic concentration following exhaustive exercise.

Previous experiment cohesively suggested that post exercise GP_X level increases[11] and dietary complexes trace mineral supplementation may be useful for improving antioxidant capacity during exercise.[12]

After exercise, Beta-carotene and antioxidant minerals combination combats against the negative effects of ROS and thus the action of SOD and GP_X were improved significantly.

Beta carotene, Manganese, Selenium, Copper, Zinc act synergistically in the form of antioxidant chain reaction. So, Beta- carotene, Manganese, Selenium, Copper, Zinc at a measured dose might be advocated to the exercising females for a better healthy life and for better performances.

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Tables:

Table 1. Physical Characteristics of Female Participants (Mean±SD)

Groups	Age (Years)	Height (cm)	Weight (kg)
Placebo (n=50)	21.4±0.63	154.78±3.06	60.69±5.85
V_{M+Se} (n=50)	21.6±0.74	156.17±5.22	50.90±8.27

Table 2. Endurance Capacity of Female Participants before and after Beta-carotenes, Minerals and antioxidants supplementation (Mean±SD)		
Groups	**Endurance Capacity**	
	Before Supplementation	**After Supplementation**
Placebo (n=50)	7.56±0.72	7.71±0.73 NS
V_{M+Se} (n=50)	7.27±0.71	15.18±0.72**
NS = Not Significant **p<0.001 = Significant		

Table 3. Recovery Heart Rates (Beats/min) of Female Participants before and after Beta-carotenes, Minerals and antioxidants supplementation (Mean±SD)						
Group		**1st min**	**2nd min**	**3rd min**	**4th min**	**5th min**
Placebo (n=50)	**Before Supplementation**	160.0±3.78	148.13±2.77	124.00±2.00	109.33±2.47	99.2±2.37
	After Supplementation	160.27±2.60 **NS**	149.20±2.70 **NS**	124.53±2.45 **NS**	109.47±2.56 **NS**	99.47±2.88 **NS**
V_{M+Se} (n=50)	**Before Supplementation**	159.73±2.71	148.0±2.51	123.87±2.33	109.47±2.33	99.6±3.04
	After Supplementation	151.47±1.51 **	140.2±1.52 **	109.33±2.19 **	92.9±1.97 **	84.13±1.92 **
NS = Not Significant **p<0.001 = Significant						

Table 4. Hemoglobin Concentration (g%) of Exercising Female Participants before and after Beta-carotenes, Minerals and antioxidants supplementation (Mean±SD)			
Groups	**Hemoglobin Concentration (g%)**		**Level of Significance**
	Before Supplementation	**After Supplementation**	
Placebo (n=50)	12.33±0.11	12.40±0.11	NS
V_{M+Se} (n=50)	12.10±0.16	12.04±0.17	NS
NS = Not Significant			

Table 5. Exercise induced changes in Oxidative stress Markers (Vitamin, Glutathione Peroxidase, Catalase, Glutathione Peroxidase, Catalase and Glutathione) level of Exercising Female Participants before and after Beta-carotenes, Minerals and antioxidants supplementation (Mean±SD)

Enzyme Parameters	Groups								Level of Significance
	Placebo (n=50)				V_{M+Se} (n=50)				
	Before Supplementation		After Supplementation		Before Supplementation		After Supplementation		
	Pre-Exercise	Post-Exercise	Pre-Exercise	Post-Exercise	Pre-Exercise	Post-Exercise	Pre-Exercise	Post-Exercise	
GPx Level (n mole NADPH oxidized/min/g of Hb)	24.23±2.67	28.4±2.58	24.09±2.39	28.32±2.73	24.0±2.44	28.1±2.65	24.9±2.19	25.8±2.07	1 & 2 = p<0.05 3 & 4 = p<0.05 5 & 6 = p<0.05 2 & 4 = NS 6 & 8 = p<0.05
	1	**2**	**3**	**4**	**5**	**6**	**7**	**8**	
CAT level (K X 10^{-2} units/g of Hb)	10.35±0.32	9.12±0.38	10.33±0.36	9.10±0.40	10.48±0.13	9.29±0.10	10.17±0.12	9.20±0.10	9 & 10 = p<0.05 11 & 12 = p<0.05 13 & 14 = p<0.05 14 & 16 = NS
	9	**10**	**11**	**12**	**13**	**14**	**15**	**16**	
SOD level (units/mg protein)	0.32±0.06	0.56±0.06	0.31±0.07	0.53±0.06	0.33±0.08	0.56±0.09	0.33±0.08	0.46±0.09	17 & 18 = p<0.001 19 & 20 = p<0.001 21 & 22 = p<0.001 22 & 24 = p<0.001
	17	**18**	**19**	**20**	**21**	**22**	**23**	**24**	

NS = Not Significant, p<0.001 highly significant, p<0.05 Significant.

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Conflict of Interest:

We the authors of this study, hereby declare that no financialandnon-financialrelationshipsandactivitiesrelatedtothemanuscriptwithentitiessuchasgovernmentagencies,foundations,commercialsponsors,academicinstitutions. We also disclose that, no resources have been receivedeitherdirectlyorindirectly(viainstitution)relatedtothemanuscriptfromthetimeofinitialconceptionanddplanningoftheworkto the present. We also disclose that there are no relationships or activities that are topically but not directly related to the work. This is for a complete listing thathelps to reassure readers of authors' commitment to transparency.