

The association between food diversity and serum antioxidant indices in cataract patients compared to healthy subjects

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Background: Cataract is a chronic disorder that is related to antioxidant–oxidant imbalance situation. We aimed to investigate the association between food diversity and serum antioxidant and oxidant indices in cataract patients compared to healthy subjects. **Materials and Methods:** In this case–control study, ninety volunteers (aged > 50 years) were divided into the cataract ($n = 45$) and healthy control ($n = 45$) groups. Anthropometric variables, physical activity and stress levels, food diversity score, serum total oxidant capacity (TOC), and total antioxidant capacity (TAC) measurements were done for all participants. **Results:** Serum TAC, even after adjustment for stress level, was significantly higher in healthy people compared to cataract patients ($P < 0.001$). In addition, serum TOC was significantly lower in healthy controls compared to cataract patients ($P < 0.002$). In healthy group, there was a weak significant positive association between serum TAC and meats group diversity ($r = 0.149, P = 0.047$). In addition, there was a moderate negative association between meats group diversity and TOC in the healthy controls ($r = -0.712, P = 0.041$). In the cataract group, there was a significant negative association between serum TOC and diversity score of fruits ($r = -0.811, P = 0.017$) and meats group ($r = -0.926, P = 0.046$) as well as total score of food diversity ($r = -0.466, P = 0.003$). **Conclusion:** It seems that increase in total dietary diversity and food groups' diversity can have a beneficial effect on oxidant situation among cataract patients.

Key words: Antioxidant, cataract, diet, oxidant

How to cite this article: Heidari N, Nabie R, Jabbari M, Irannejad Niri Z, Zeinalian R, Asghari Jafarabadi M, *et al.* The association between food diversity and serum antioxidant indices in cataract patients compared to healthy subjects. *J Res Med Sci* 2021;26:59.

INTRODUCTION

Cataract, a chronic age-related condition, is one of the major causes of eyesight damage and blindness, which is responsible for about 48% of blindness in the world.^[1] Low-income countries have more than 80% of cataract patients, and it has been suggested that cataract is a leading cause of blindness (about 41.70%–42%) in Asia.^[2] Based on the data from meta-analysis of 27 studies, overall frequency of cataract in Iran was 9.27% with more frequency in females than in males (8.03 vs. 8.32%, respectively).^[2]

One of the major risk factors of cataract that has been proposed in recent studies is oxidative stress. It has been suggested that oxidative stress has a main role in the pathophysiology of age-related cataract.^[3] Then it is so important to investigate and identify various factors to reduce the severity of complications or delay the progression of the disorder.

Based on definition, oxidative stress is the disbalance between the oxidant situation (free radicals including reactive oxygen and nitrogen species are normally produced in cellular metabolic reactions) and antioxidant

Access this article online	
Quick Response Code: 	Website: www.jmsjournal.net
	DOI: 10.4103/jrms.JRMS_321_20

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Submitted: 05-May-2020; **Revised:** 29-Jun-2020; **Accepted:** 08-Mar-2021; **Published:** 30-Aug-2021

defense (enzymes including superoxide dismutase [SOD], catalase, and glutathione peroxidase [GPx] and water- and lipid-soluble antioxidants, such as glutathione, ascorbate [Vitamin C], α -tocopherol [Vitamin E], and β -carotene, and also endogenous antioxidant in the body).^[4] In this harmful situation, while free radical's concentration increases, the efficiency of antioxidant defense decreases. To investigate serum oxidant and antioxidant status, total antioxidant and oxidant capacities are measured.^[5] The total oxidant capacity (TOC) of biological samples is the amount of serum total oxygen and nitrogen free radicals. In addition, the total antioxidant capacity (TAC) is the amount of antioxidant compounds or enzymes with antioxidant function in the biological samples.^[6]

Previous researches have been suggested that oxidant-antioxidant balance in the body is related to the incidence and progression of many kinds of chronic diseases.^[6] On the other hand, dietitians are looking for finding beneficial effects of specific dietary patterns (which are main resources of many crucial macro/micronutrients and antioxidants) on the prevention or amelioration of many noncommunicable chronic diseases.^[7-11] In this meantime, food diversity has attracted much of interests for many research fields. Increment in the number of food groups' consumption during a day as-called food/dietary diversity could have beneficial health effects as suggested in the previous studies.^[12-14]

To the best of our knowledge, there is no previous study to evaluate the association of food diversity and serum TAC and TOC in cataract patients. In the present study, we hypothesized that total food diversity score and food groups' diversity are related to the levels of serum TAC and TOC. Hence, we aimed to investigate the association between food diversity and serum antioxidant and oxidant indices in cataract patients compared to healthy subjects.

MATERIALS AND METHODS

Study design and participants

In this case-control study, ninety volunteers (aged more than 50 years) were recruited based on convenient sampling method. Among who admitted to the Mehr Ophthalmological Center in Tabriz and their companions, forty-five nondiabetic cataract patients and 45 healthy people were included in the case and control groups, respectively. Inclusion criteria for case group were patients aged more than 50 years and with the confirmation of cataract diagnosis by an expert ophthalmologist. Exclusion criteria for case group were patients having any other chronic diseases (including type 2 diabetes mellitus, cancer, and gastrointestinal, kidney, and liver diseases), receiving any dietary supplements (micronutrients or phytoestrogens), and on steroid drugs over the past 6 months. Control group

was selected from apparently healthy people who were patients' companions came to the same ophthalmological center. They had not any obvious symptoms or diagnoses of diseases at the study time as reported by themselves. Cataract diagnosis in the case group has been done based on the opinion of the expert ophthalmologist according to slit lamp procedure.^[15] The study groups were matched for age (within 5-year age groups as 50-54, 55-59, 60-64, and 65-70), body mass index (BMI) (within five BMI groups as 18.5-24.9, 25-29.9, 30-34.9, 35-39.9, and 40-44.9 kg/m²), and sex (individual matching). The aim of the study was clearly explained for all participants. Before the start of the study, written informed consent was obtained from all subjects. The study protocol was approved by the Ethical Committee of Tabriz University of Medical Sciences, Tabriz, Iran (Ethics Code: IR.TBZMED.REC.1396.1138).

Sample size calculation

The study sample size was calculated based on Wang *et al.*'s study^[16] by considering mean \pm standard deviation (SD) of SOD, with power $1 - \beta = 0.80$ and type I error probability $\alpha = 0.05$. Based on the mentioned data, the sample size in each group was calculated (38 subjects in each group). Considering 20% sample dropout, the final study sample size was calculated (45 subjects in each group). The formula for calculating the sample size is given below:

$$N = \frac{(Z_{1-\alpha/2} + Z_{1-\beta})^2 (S_1^2 + S_2^2)}{(\mu_1 - \mu_2)^2}$$

Anthropometric measurements

All measurements were done by an expert individual. Body weight measurement was done using a "seca" scale in the accuracy of 0.1 kg, while the participants were wearing light clothes with no shoes. A wall-mounted stadiometer was used for height measurement to the nearest 0.5 cm when the participants were barefoot. BMI calculation was done by dividing body weight in kg by the square of height in m².

Physical activity and stress levels assessment

International Physical Activity Questionnaire short-form was used for the determination of physical activity levels in all participants.^[17] For this purpose, data were expressed as a standard metabolic energy turnover in MET-minutes/week. Then, physical activity levels in all participants were classified in three levels: low (<600 MET-minutes/week), moderate (from 600 to 2999 MET-minutes/week), and high (3000 MET-minutes/week or more). For investigation of stress levels, we used the Holmes and Rahe stress scale.^[18] Based on this scale, stress status was determined in four levels including mild (score <150), moderate (score = 150-200), high (score = 200-300), and severe (score > 300). Data were collected by face-to-face interview.

Food intake assessments

To assess the amount of the portion sizes of consumed food items in the past year, we used a self-administered semi-quantitative 147-item food frequency questionnaire (FFQ). Validity and reliability of this questionnaire in the Iranian community were determined previously.^[19] This FFQ contains questions about on the average consumption frequency during the past year for 147 food items. These food items including meats and their substitutions, eggs, dairy products, fruits, vegetables, whole and refined cereals, fats, simple sugar, salt and other food additives, beverages, and a variety of fast foods. Participants could indicate their answers in “never,” “times per day,” “times per week,” “times per month,” or “times per year.” Food preparation methods also were asked in the face-to-face interviews. Altogether, the habitual consumption of 147 food items was calculated from the obtained information. For determination of some food items’ portion size, the colored food album was proposed to participants.^[20] The average of daily consumption for each food item was calculated by multiplying consumption frequency of that specific food item to the standard amount of its substitution.

Determination of food diversity

We used Kant *et al.*'s method for the determination of food diversity in the present study.^[21] For calculating food diversity scores, consumed food items were divided into major five food groups and 23 subgroups including grains with seven subgroups (white bread, wholegrain biscuits, pasta, whole grain breads, noodle, rice, and barley), vegetables with seven subgroups (vegetables, potatoes, tomatoes, other starchy vegetables [corn, pea, eggplant, squash], legumes [pea, beans, mung beans, split peas, lentils], yellow vegetables [carrots and pumpkin], and other green vegetables [bell peppers, all kinds of cabbage, broccoli, celery, cucumbers, garlic, onion, green beans, zucchini, leeks, parsley, lettuce, radish, spinach, rhubarb, turnips]), fruits with two subgroups (berries and citrus, other fruits and juices), meats with four subgroups (red meat [cattle and sheep], poultry [hen and chicken], fish, and eggs), and dairy groups with three subgroups (milk [low-fat and full-fat], yogurt [low-fat and full-fat], cheese, or curd).^[22] Participants who consumed at least half serving of any food items in any five major food groups were considered in the food diversity score calculating. It has been allocated two scores to each food group. Food diversity score of each food group was calculated by dividing the number of consumed food items in each food group by all consumed food items in a food group and the multiplying obtained number to two. Sum of the all scores of food diversity of all food groups makes the total food diversity score for each participant.^[23]

Biochemical assessments

After an overnight fasting, 5 ml venous blood was collected

from all participants. Separation of serum from the whole blood was done by centrifugation of samples at 2000 rpm for 10 min at 4°C. Aliquots were stored at -70°C until analysis time. Serum TAC and serum TOC were determined by TAC (TAC assay kit, Zellbio GmbH, Ulm, Germany) and TOC (TOC assay kit, Zellbio GmbH, Ulm, Germany) ELISA kits, respectively. An Automatic ELISA Plate Reader (Bio Tek Instruments, Winooski, VT, USA) was used for absorbance reading at 450 nm.

Statistical analysis

Demographic characteristics of the participants were presented as mean \pm SD for continuous data or frequency (percentage %) for proportional data. Assessment of equality in the continuous data was assessed by Kolmogorov-Smirnov test. Independent sample *t*-test and Chi-square were used for comparison of between-group quantitative variables and comparison of between-group proportions. To adjustment for the effect of confounding variables, an analysis of covariance was used. The linear and binary logistic regression test were used for the assessment of relationship between dietary diversity score with TAC and TOC levels as well as the association between dietary diversity score and risk of cataract in the study groups, respectively. A $P < 0.05$ was considered statistically significant. IBM SPSS Statistics 19 Software (IBM Corp., Armonk, NY, USA) was used for all statistical analysis.

RESULTS

General characteristics of the study population

As shown in Table 1, there was no significant difference between the two groups in age, weight, height, BMI, stress, and physical activity levels ($P > 0.05$). However, serum TAC, even after adjustment for stress level, was significantly higher in healthy people compared to cataract patients ($P < 0.001$). In addition, serum TOC was significantly lower in healthy controls compared to cataract patients ($P < 0.002$).

Association between serum total antioxidant capacity, total oxidant capacity, and food diversity in the study population

The association between serum TAC and food diversity scores in the study population is shown in Table 2. After adjustment for stress levels, there was no significant association between serum TAC and food diversity scores neither in the healthy controls nor in the cataract group. However, in the healthy group, there was a weak significant positive association between serum TAC with meats group diversity ($\beta = 0.149$, $P = 0.047$).

As shown in Table 3, there was a moderate negative association between meats group diversity and serum TOC

in the healthy controls ($\beta = -0.712, P = 0.041$). In addition, in the cataract group, there was a significant negative association between serum TOC with diversity score of fruits ($\beta = -0.811, P = 0.017$) and meats ($\beta = -0.926, P = 0.046$) group as well as total score of food diversity ($\beta = -0.466, P = 0.003$).

Risk of cataract in the study population

As shown in Figure 1, there was no significant association between dietary diversity score and risk of cataract based on binary logistic regression test in the both crude and adjusted model (adjusted for stress and physical activity levels) in the study population (odds ratio = 1.506, 95% confidence interval: 0.539–4.207) among total dietary diversity tertiles.

Table 1: General characteristics of the study subjects (n=45)

Variables	The study group		P	
	Cataract patients	Healthy controls		
Age (years)	58.44±4.51	58.24±5.59	0.852 ^a	-
Weight (kg)	75.92±12.23	74.01±11.82	0.453 ^a	-
Height (m)	1.64±0.09	1.63±0.09	0.582 ^a	-
BMI (kg/m ²)	28.17±4.57	27.81±4.38	0.706 ^a	-
Stress	40.51±28.95	42.484±29.04	0.747 ^a	-
TAC (mmol/L)	1.00±0.22	1.19±0.24	<0.001 ^a	<0.001 ^b
TOC (μmol/L)	9.02±1.09	8.25±1.09	<0.001 ^a	0.002 ^b
Physical activity				
Low	36 (80)	32 (71.1)	0.327 ^c	-
Moderate	9 (20)	13 (28.9)		

^aP value was reported based on independent sample t-test; ^bP value was reported based on ANCOVA test after adjustment for stress levels; ^cP value was reported based on Chi-square test. BMI=Body mass index; TAC=Total antioxidant capacity; TOC=Total oxidant capacity

DISCUSSION

In the present study, serum TAC was considerably higher in the healthy people compared to cataract patients. In addition, serum TOC was meaningfully lower in healthy controls compared to the cataract patients. Rokicki *et al.* in their study showed that concentration of oxidative degradation products including malonyldialdehyde (MDA) and lipofuscin was significantly higher in glaucomatous cataract patients compared with nonglaucomatous.^[24] In addition, they reported that serum Mn-SOD and activity of total SOD were significantly lower in glaucoma patients. In another study, Selvi *et al.* compared serum TAC between cataract patients and healthy controls. They reported that serum TAC was significantly lower in cataract patients.^[5] Furthermore, the results of Türk *et al.*, in their study about the investigation of serum oxidant and antioxidant status of patients with mature and immature senile cataract, showed that oxidative stress indices in the aqueous humor of mature

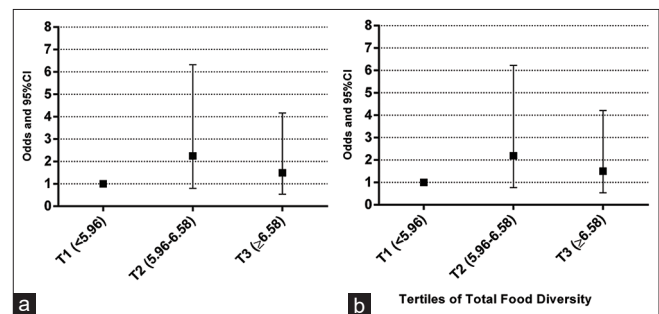


Figure 1: Risk of cataract in the study population among total dietary diversity tertiles. (a) Crude model, (b) Adjusted model

Table 2: The association between serum total antioxidant capacity and food diversity scores in the study population (n=45)

Variables (diversity in five major food groups)	Cataract patients		Healthy controls	
	Un-adjusted ^a	Adjusted ^b	Un-adjusted ^a	Adjusted ^b
Bread and cereals				
B	-0.065	-0.070	-0.269	-0.299
P	0.649	0.625	0.092	0.068
Vegetables				
B	0.037	0.034	-0.103	-0.098
P	0.707	0.732	0.351	0.382
Fruits				
B	0.134	0.136	-0.099	-0.095
P	0.056	0.055	0.510	0.528
Dairy				
B	0.071	0.065	0.045	0.051
P	0.388	0.438	0.507	0.460
Meat, poultry, fish, eggs				
B	0.170	0.166	0.149	0.146
P	0.073	0.084	0.047	0.055
Total				
B	0.063	0.062	0.014	0.015
P	0.055	0.065	0.679	0.673

^aData were reported based on linear regression; ^bData were reported after adjustment for stress levels

Table 3: The association between serum total oxidant capacity and food diversity scores in the study population (n=45)

Variables (diversity in five major food groups)	Cataract patients		Healthy controls	
	Un-adjusted ^a	Adjusted ^b	Un-adjusted ^a	Adjusted ^b
Bread and cereals				
B	-0.348	-0.331	1.062	1.175
P	0.616	0.637	0.147	0.119
Vegetables				
B	-0.570	-0.561	0.325	0.306
P	0.226	0.238	0.523	0.554
Fruits				
B	-0.805	-0.811	0.460	0.448
P	0.017	0.017	0.502	0.518
Dairy				
B	-0.512	-0.496	-0.299	-0.324
P	0.200	0.224	0.334	0.304
Meat, poultry, fish, eggs				
B	-0.938	-0.926	-0.719	-0.712
P	0.041	0.046	0.036	0.041
Total				
B	-0.468	-0.466	-0.117	-0.119
P	0.003	0.003	0.454	0.453

^aData were reported based on linear regression; ^bData were reported after adjustment for stress levels

cataract patients was higher while antioxidant capacity was lower. They proposed that elevated oxidative stress may had a crucial role in maturation and pathogenesis of senile cataract.^[25]

As mentioned above, the results of other related studies about oxidant and antioxidant status of cataract patients (serum or aqueous humor investigation) are in accordance with our results. The rising bodies of evidence have been suggested that oxidative stress is a trigger agent in cataract development.^[26,27] Furthermore, Vinson in his review claimed that the defect in the body antioxidant defense could be a cause to incidence and progression of cataract.^[28]

Given that lens cells are in constant exposure to H₂O₂ and other prooxidant agents, so it is an important issue to protect these sensitive proteins from the oxidation damages.^[29]

The glutathione (GSH) level (as the pivotal member of antioxidant defense in the body)^[30] is an indicator of cataract development.^[31] As oxidized form of glutathione (GSSG) rises, the GSH loss occurs. It has been reported that increment in GSSG and decrease in GSH levels are related to cataract development approximately in all *in vivo* cataract models.^[28]

In the present study, after adjustment for stress values, there was a meaningful negative association between meats group diversity score and serum TOC in the both healthy controls and cataract groups. In addition, in the cataract group, there was a considerable negative association between sera TOC with diversity score of fruits group.

Based on evidence from Root *et al.*'s study, there was a negative association between higher fruits and vegetables intake and lower serum inflammatory factors including IL-6, TNF- α , and CRP in general population.^[32] On the other hand, there was a positive association between fruits and vegetables intake and indicators of plasma antioxidant capacity including oxygen radical absorbance capacity (ORAC) and ferric reducing antioxidant power (FRAP). The relationship also was negative between intake of mentioned food groups and plasma F2-isoprostane (an oxidant status indicator) levels.^[32]

It has been hypothesized that elevation in iron intake following red meat consumption increases/exacerbates oxidative stress in the body. The proposed mechanism is that iron could be catalyze high reactive hydroxyl radicals generation (Haber-Weiss reaction).^[33] Dietary components could be the main resources of oxidants or antioxidants agents.^[34] *In vitro* researches showed that specific micronutrients, especially precursors of fat-soluble vitamins including tocopherols and carotenoids, could scavenge ROS from the body fluids.^[35]

It is worth noting Romeu *et al.* reported that the mentioned association between dietary iron and oxidative stress is just about heme iron not nonheme. Even they found that this association is negative in terms of nonheme iron.^[36] It could be inferred from these findings that nonheme iron dietary resources such as fruits and vegetables have beneficial effects on oxidative stress status, while heme iron dietary resources, especially red meat, exacerbate this situation in the body. However, in the same study, it has been suggested that serum ORAC has positive correlation with vegetables

and Vitamin C intake and negative correlation with meat and saturated fatty acid intake.^[36]

Despite other findings about the relationship between red meat intake and oxidant status, our findings showed a meaningful negative association between meats group diversity score and serum TOC in both healthy controls and cataract group. First of all, it is worth noting that meats group is different from red meat as a single food item and includes red meat, fish, poultry, and eggs. The composition and quantity of endogenous antioxidants and prooxidant can be affected by different species of meats, even among animals of a single species as well as muscle type.^[37] Furthermore, the pasture or grain of the diet of the animals can play a meaningful role in content and modification of fatty acids, prooxidant, and antioxidants in the meats. On the other hand, consumers prepare and process meats by means of wide range of thermal treatments and cooking procedures. These diverse and different cooking processes can change the composition and structure of meats' components and considerably change the antioxidant capacity of meats.^[37]

There is a growing body of evidence that meat proteins are a good source of antioxidant peptides.^[38] For example, the high available essential amino acids such as hydroxy-methyl-lysine and methyl-histidine that are specific to meats and not usually found in plants are some of these peptides. On the other hand, some notable antioxidant peptides, anserine and carnosine, exist in the muscle tissue, endogenously. These peptides act as scavengers of free radicals and chelators of metal ion.^[38] In the present study, we did not gather the detailed data about the cooking processes of meats group in the study subjects, but all of the mentioned reasons can be justifications for founded meaningful negative association between meats group diversity score and TOC in the both healthy controls and cataract groups. However, it needs more investigation to find more detailed mechanisms in this regard.

The novel finding of the present study was the association between total food diversity score and serum TOC in cataract patients. Based on the facts mentioned above, it seems that higher food diversity may be related to increased intake of foods containing antioxidant/proantioxidant nutrients (such as vegetables, fruits, fish, and poultry). On the other hand, it seems that concurrently decrease in consumption of foods with high content of oxidant/prooxidant nutrients (such as red and processed meats) is necessary to emerge beneficial effects of high food diversity on well-being. Altogether, this situation could help to efficient antioxidant defense and alleviate inflammation and detrimental oxidations in the body. In the other words, the total characteristics of diet, altogether, determine the extent of ROS production and oxidative stress or antioxidant situation.

There are many studies have been investigated the effect of only one or a few nutrients (variety of vitamins and minerals) on the eye health, simply.^[39-41] However, the effect of whole meals and foodstuffs intake has been ignored in these researches. It could not be denied the beneficial and health effects of whole meals and food/dietary diversity on human health, especially eye health, compared to simple nutrients intake.

Based on our knowledge, the present study is one of the limited researches on the association between food diversity and serum antioxidant indices in cataract patients compared to healthy subjects. However, there were some limitations in the present study including low sample size in each group and limitation in the measurement of other serum oxidant/antioxidant indicators such as MDA, GPx, and SOD. In addition, we did not gather the detailed data about the cooking processes of different consumed food groups, especially meats group, among the participants.

CONCLUSION

Based on the findings of the present study, it seems that increase in total dietary diversity and food groups' diversity can have a beneficial effect on oxidant situation among cataract patients. Although findings of the present study had limitations to infer this fact, it could be a start point for more detailed and large-scaled studies, even intervention studies, in this field. These findings could provide promising data for future studies in discovering specific dietary patterns to ameliorate or prevent cataract complications.

Acknowledgments

Data presented in this article are results of the MSc thesis in Health Sciences in Nutrition. This research was supported by School of Nutrition and Food sciences, Tabriz University of Medical Sciences (Ethics Code: IR.TBZMED.REC.1396.1138).

Financial support and sponsorship

This research was supported financially from Research Deputy of Tabriz University of Medical Sciences, Tabriz, Iran.

Conflicts of interest

There are no conflicts of interest.

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