

Evaluation of Oxidants, Antioxidants and Some Biochemical Parameters in Vegetarianisms and Ova-lacto vegetarianisms

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Abstract

The study was conducted in Ninawa governorate to show the evaluation of oxidants, antioxidants and some biochemical parameters in vegetarianisms and ova-lacto vegetarianisms parameters which include: vitamin E, vitamin A, β -carotene, vitamin C, ceruloplasmin (Cp), total protein, albumin, calcium, total bilirubin, uric acid, creatinine, total iron binding capacity (TIBC), iron, glutathione(GSH), malondialdehyde (MDA), peroxynirate (ONOO^-), cholesterol, glucose, glutathione S-transferase (GST) . The study included (182) persons represented in three groups (Male and nonsmokers): the first group as a vegetarianism (54), the second group represented for ova-lacto vegetarianisms (62) and the three group omnivores (Non-vegetarianisms) (66) .

The results showed a significant increase in vegetarianisms in comparison with non-vegetarianisms for: vitamin E, vitamin A, β -carotene, vitamin C, total protein, albumin, calcium, TIBC, GSH and glucose. While a significant decrease in total bilirubin, MDA, ONOO^- , cholesterol, GST and body mass index(BMI). The results also showed a significant increase of ova-lacto vegetarianism in: vitamin E, β -carotene, vitamin C, GSH and glucose when compared with non-vegetarianisms. While a significant decrease in: Cp., total protein, calcium, TIBC, MDA, ONOO^- , GST and BMI. On the other hand, the results revealed a significant increase of vegetarianism most antioxidants when compared with ova-lacto vegetarianisms, while a significant decrease in oxidants compounds

In conclusion, vegetarianisms and ova-lacto vegetarianisms persons had an increase (Vegetarianism Especially) antioxidants levels compared with omnivores leading to a decrease in their oxidative stress, disease and its complication from oxidants.

Keywords: Vegetarianisms, Ova-lacto vegetarianisms, Antioxidants, Oxidants.

الخلاصة

شملت الدراسة معرفة مستويات الاكسدة ومضاداتها وبعض المتغيرات الكيموحيوية لدى النباتيين والنباتيين الذين يتناولون البيض والحليب في محافظة نينوى، إذ تضمنت تلك المتغيرات: فيتامين E، فيتامين A، بيتا كاروتين، فيتامين C، سيرولوبلازمين (Cp.)، البروتين الكلي، ألبيومين، كالسيوم، البليروبين الكلي، حامض اليوريك، كرياتينين، سعة الارتباط الكلي للحديد (TIBC)، الحديد، كلوتاثيون (GSH)، المالوندايالديهايد

(MDA)، بيروكسي نيتريت (-ONOO)، كوليستيروول، كلوكوز وكلوتاثايون S- ترانسفيريز (GST). استخدمت في الدراسة (182) شخصا" مثلوا بثلاث مجاميع (ذكر وغير المدخنين): المجموعة الاولى مجموعة نباتيين (54) والمجموعة الثانية مثلت بالأشخاص النباتيين والذين يتناولون البيض والحليب في وجباتهم (62) والمجموعة الثالثة غير المعتمدين على النبات بشكل أساس في التغذية (يتناولون النباتات واللحوم) (66). أظهرت النتائج زيادة معنوية لدى النباتيين عند مقارنتهم مع غير النباتيين للمتغيرات: فيتامين E، فيتامين A، بيتا كاروتين، فيتامين C، البروتين الكلي، الألبومين، الكالسيوم، TIBC، GSH، والكلوكوز، بينما ظهر هناك انخفاض معنوي للبيروكسي الكلي، MDA، -ONOO، الكوليستيروول، إنزيم GST وقيمة BMI. فضلا عن ذلك، لوحظ زيادة معنوية لدى النباتيين (الذين يتناولون البيض والحليب) في المتغيرات: فيتامين E، بيتا كاروتين، فيتامين C، GSH، وكلوكوز عند مقارنتهم مع غير النباتيين. بينما لوحظ ان هناك انخفاض معنوي في المتغيرات: Cp، البروتين الكلي، الكالسيوم، TIBC، MDA، -ONOO، إنزيم GST وقيمة منسب كتلة الجسم (BMI). من جانب اخر، لوحظ زيادة معنوية لدى النباتيين في اغلب مستويات مضادات الاكسدة عند مقارنتهم مع النباتيين الذين يتناولون البيض والحليب وانخفاض معنوي في مستويات الاكسدة. يستنتج من ذلك، ان النباتيين والنباتيين من الذين يتناولون البيض والحليب أيضا تزداد لديهم مستويات مضادات الاكسدة (وبالأخص النباتيين) أكثر من الأشخاص الذين يتناولون النباتات واللحوم وبالتالي يكونون اقل عرضة للإصابة بالأمراض المختلفة او مضاعفة الأمراض نتيجة الأكسدة .

الكلمات الدالة: النباتيين، النباتيين الذين يتناولون البيض والحليب، مضادات الأكسدة، الأكسدة.

Introduction

Evidence that diet is a key environmental factor affecting the incidence of many chronic diseases is overwhelming⁽¹⁾. Antioxidant substances in such a diet enhance the DNA, protein and lipid protection by increasing the scavenging of radical oxidative species that occur during metabolic reactions. The a imbalance between the amount of "unhealthy" and "healthy" food leads to the accumulation of oxidants damage, initiating DNA instability and inducing cancer development⁽²⁾.

Three degrees of vegetarianism present, each one distinguished by what's allowed in addition to fruits, grains and veggies; degree one is a plant-based diet for people who don't eat meat but do eat fish and poultry or just fish, degree two is a plant-based diet for people who don't eat meat, fish, or poultry but do eat other animal products such as eggs and dairy products. Vegetarians who follow this

regimen are called ovo-lacto vegetarians (or Lacto-ova vegetarians) (ovo = egg; lacto = milk) and degree three is a diet for people who eat absolutely no foods of animal origin. Vegetarians who eat only plant foods are called vegans^(3,4).

Organisms are continually exposed to a number of reactive oxygen species (ROS) and reactive nitrogen species (RNS) that react with intracellular lipids, proteins, and nucleic acids, leading to loss of biological function, various forms of ROS/RNS are present as pollutants in the atmosphere, are formed during exposure to irradiation, or occur as by-products of normal metabolic processes and in inflammatory, drugs and other chemicals also can increase formation of oxidants compounds (ROS and RNS in cells. Whereas organisms contain numerous antioxidants that can convert ROS/RNS to inactive derivatives, their antioxidant efficiency dependent on

their intracellular concentrations and on the rates of ROS/RNS generation, which depends on the magnitude of the oxidative stress⁽⁵⁾. Antioxidant substances in diet enhance DNA, lipid and protein protection by increasing the scavenging of free radicals⁽⁶⁾.

Because of increased vegetarians with increased the poor in Iraq after occupation in (9/4/2003), an attempt to show the evaluation of oxidants, antioxidants and some biochemical parameters in omnivores, vegetarians and ova-lacto vegetarians in Ninawa governorate.

Materials and Methods

The study included (182) persons (Male and nonsmokers) classified in three groups: the first group as a vegetarianism (n=54), the second group represented for ova-lacto vegetarians (n=62) and the three group non-vegetarians (Omnivores)

(n=66). For venipuncture, 10 ml sterile syringes equipped with (22G x 1.25) syringe needles were used and put of blood in dry and clean plain tube. After coagulation, it was centrifuged at 4,000 x g for 15 minute. Serum was transferred into plain tube equipped with tight-fitting caps by disposable tips, then stored at -20 °C⁽⁷⁾ until use.

Kits for determination of total Protein, albumin, calcium, total Bilirubin, iron, Total Iron Binding Capacity(TIBC), Glucose, Cholesterol, were obtained from BIOLABO kits, France, But other biochemical parameter (vitamin E, Vitamin A, β-carotene, vitamin C, ceruloplasmin (Cp.), malondialdehyde (MDA), glutathione(GSH), peroxy-nitrate, uric acid and creatinine and glutathione-S-transferase (GST)) determination used manual methods (Table 1).

Table 1: Methods used to determination of biochemical parameters.

No.	Parameters measured	Method used
1	Vitamin E	Emmerie-Engel reaction ⁽⁸⁾
2	Vitamin A	Needld-Pearson method ⁽⁹⁾
3	B-carotene	Needld-Pearson method ⁽⁹⁾
4	Vitamin C	2,4-dinitrophenylhydrazine derivatization method ⁽¹⁰⁾
5	Cp.	p-Phenylenediamine oxidase method ⁽¹¹⁾
6	Total protein	Biuret methods ⁽¹²⁾
7	Albumin	Bromocresol green method(dye binding method) ⁽¹³⁾
8	Calcium	Methylthymol blue method ⁽¹⁴⁾
9	Total bilirubin	Diazo method ⁽¹⁵⁾
10	Uric acid	Phosphotungstic acid method ⁽¹⁶⁾
11	Creatinine	Jaffé method ⁽¹⁷⁾
12	TIBC	Ramsay method ⁽¹⁸⁾
13	Iron	Bathophenanthroline method ⁽¹⁸⁾
14	GSH	Modified procedure utilizing Ellman's reagent. ⁽¹⁹⁾
15	MDA	Thiobarbituric acid method ⁽²⁰⁾
16	Peroxy-nitrate	Modified procedure ⁽²¹⁾
17	Cholesterol	Cholesterol esterase methods ⁽²²⁾
18	Glucose	Glucose oxidase method ⁽²³⁾
19	GST	1-chloro-2,4-dinitrobenzene(CDNB)conjugation ⁽²⁴⁾

For determination of Body Mass Index (BMI) calculated using the formula as weight (kg)/height²(m²)⁽²⁵⁾.

Results and Discussion

1- Compared of vegetarianism with non-vegetarianism group(omnivores):

The results of biochemical parameters for vegetarianism and non-vegetarianism group were listed in Table (2).

The results showed a significant increased in: vitamin E (P=0.041), vitamin A (P=0.03), β -carotene (P=0.022), vitamin C (P=0.0001), total protein (P=0.034), albumin (P = 0.047), calcium (P=0.035), TIBC(P=0.042), GSH (P=0.0001) and glucose (P=0.01). While a significant decrease in vegetarians when compared with non-vegetarians for: total bilirubin (P=0.036), MDA (P=0.0001),

cholesterol (P=0.045), GST(P=0.033) and BMI (P=0.045) which were similar to other reported results^(26, 27, 28, 29, 30, 31, 32, 33).

Vegetarian dietary practices have been associated with many health benefits, including lower death rates from ischemic heart disease, diabetes, and certain forms of cancer^(34, 35) and lower risks of dyslipidemia, hypertension, and obesity⁽³⁶⁾. Most vegetarians have higher intakes of fruit and vegetables, dietary fiber, antioxidant nutrients, phytochemicals, and folic acid than do nonvegetarians, and lower intakes of saturated fat and cholesterol⁽³⁷⁾, which have been related to lower risk of chronic disease⁽³⁸⁾.

Table 2: The biochemical parameters of vegetarians and non-vegetarianism.

Parameters	Non-vegetarianism (n=66)		Vegetarianism (n=54)		t-test
	Mean	SD	Mean	SD	P-value
Age (year)	30.04	1.54	34.1	1.91	0.11
B.M.I (k.g./m ²)	24.43	1.01	23.45	1.21	0.045*
Vit.E (mg/dl)	1.14	0.09	1.88	0.11	0.041*
Vit.A (μ g/dl)	47.66	3.1	56.49	8.3	0.03*
β -carotene (μ g/dl)	59.08	8.6	84.47	11.18	0.022*
Vit.C (mg/dl)	0.77	0.07	1.09	0.09	0.0001**
Cp. (mg/l)	145.29	14.05	112.9	10.56	0.6
T.p. (gm/dl)	5.03	0.24	5.93	0.17	0.034*
Alb. (gm/dl)	3.92	0.1	4.74	0.16	0.047*
Calcium (mg/dl)	9.20	0.19	10.07	0.69	0.035*
Total Bilir. (mg/dl)	0.32	0.01	0.24	0.02	0.036*
Uric acid (mg/dl)	4.94	0.1	4.87	0.18	0.431
Creatinine (mg/dl)	3.66	1.4	2.75	0.11	0.442
TIBC (μ g/dl)	290.97	35.37	415.32	84.64	0.042*
Iron (mg/l)	1.27	0.23	1.52	0.21	0.411
GSH (μ mol/l)	10.84	0.73	15.8	0.4	0.0001**
MDA (μ mol/l)	14.46	2.04	5.01	0.35	0.0001**
Peroxy nitrate	98.64	7.05	77.45	6.23	0.018*
Cholesterol (mg/dl)	181.97	7.00	163.87	5.14	0.045*
Glucose (mg/dl)	67.62	3.72	82.99	7.05	0.01*
GST(U/l)	36.12	6.12	19.29	3.16	0.033*

* Different Significantly at $p \leq 0.05$.

** High Significantly at $p < 0.0001$.

Oxidative stress, antioxidant status and their relation to diet is a subject of interest in recent years⁽³⁹⁾. In vegetarianism a high levels of antioxidants compounds (vitamin E, vitamin A, β -carotene, vitamin C, albumin and GSH) compared to non-vegetarianism were observed and these are give protection against the oxidants compounds for vitamin E is an efficient antioxidant and nonenzymatic terminator of free radical chain reactions⁽⁴⁰⁾. When Vitamin E donates an electron to a lipid peroxy radical, it is converted to a free radical form that is stabilized by resonance⁽⁴¹⁾.

Carotenoids is a term applied to β -carotene these compounds can exert antioxidant effects, as well as quench singlet 1O_2 (singlet oxygen). The relative efficiency of individual species at preventing lipid peroxidation chain reactions. Beside of the carotenoid radicals can be reduced by both α -tocopherol and ascorbic acid, which might account for the observed synergistic interactions between these three important vitamins (A, C, and E)⁽⁴²⁾.

Albumin, one of the most important protein in human plasma, is able to bind to Cu^{++} tightly and with iron weakly. Copper bond to albumin is still effective in generating radicals species (Hydroxyl radicals) in the presence of hydrogen peroxide by Fenton reactions. Therefore increasing it to protect human from oxidants compounds formation⁽⁴³⁾.

Increase of calcium in vegetarians because increasing consume enough leafy greens, which are sources of abundant calcium. Some sources of calcium include collard greens, bok choy, kale, and turnip greens. Spinach, swiss chard and beet greens are high in calcium, but the calcium is bound to oxalate and therefore it is poorly absorbed⁽⁴⁴⁾.

The results observed an increase in GSH, which is one of the most important antioxidants that occur in almost all organisms removed of oxidants by direct or indirect mechanism. Its used as a substrate in glutathione peroxidase and glutathione S-transferases and other enzymes⁽⁴⁵⁾. Therefore an increase in the defenses against to oxidants and detoxifications compounds might be inter in human body.

Bilirubin is a powerful lipophilic antioxidant that protects membranes from lipid peroxidation and protects membrane proteins from oxidation, Much of the power of bilirubin as an antioxidant comes from the extreme rapidity with which biliverdin (oxidized bilirubin) is converted to bilirubin by biliverdin reductase⁽⁴²⁾. A small concentration of bilirubin can protect against a 10,000-fold greater concentration of hydrogen peroxide due to the abundance and rapid action of biliverdin reductase in all tissues⁽⁴⁶⁾. Therefore the results showed a significant decrease in vegetarianisms in comparison with non-vegetarianisms, because of present antioxidant compounds (for example vitamin antioxidants Table(2)). On other hand, decrease in GST in vegetarianisms led to a decrease of detoxification and oxidants compounds. The GST increase when present is best known for its role in detoxification of environmental carcinogens. However, they also catalyze specific reactions in a number of biosynthetic and catabolic pathways and play an important role in defense against oxidative stress by reducing peroxides and dehydroascorbate⁽⁴²⁾.

In addition a decrease in cholesterol and (BMI) in vegetarians Table (2), leads to a decrease of ischemic heart disease, less incidence of heart disease, hypertension, type 2 diabetes, renal disease, osteoporosis,

dementias such as Alzheimer's disease and other disorders⁽⁴⁷⁾. Mortality was might be observed probably, and lower prevalence of obesity. Probably an ample consumption of fruits and vegetables and not the exclusion of meat make vegetarians healthful⁽⁴⁸⁾. A diet with a strong vegetal component, without being vegetarian, provides a very broad range of antioxidants and thus appears to be an important defense against oxidative stress⁽⁴⁹⁾.

Malondialdehyde(MDA) is an organic compound with the formula $\text{CH}_2(\text{CHO})_2$. The structure of this species is more complex than this formula suggested. This reactive species occurs naturally and is a marker for oxidative stress and a significant decreased in vegetarianisms when compared with non-vegetarianisms that refer to not occur oxidative stress⁽⁵⁰⁾. On the other hand a significantly decrease in peroxynitrate (ONOO-) indicator to less superoxide radical anion in vegetarianisms accompanied with nitric oxide radical.

Lower intakes of saturated fatty acids, and cholesterol in vegetarians than in omnivores were also found in several other studies^(37, 51, 52). A higher intakes of dietary fiber in vegetarians

than in omnivores⁽⁵³⁾. Overall, the vegetarians had nutrient intakes closer to those recommended for healthy eating than did the omnivores. Literature brings evidence of the association between cholesterol high serum levels and the prevalence of arterial diseases, especially atherosclerosis, which might lead, among other problems, to myocardial infarction and cerebral vascular accidents⁽⁵⁴⁾. Recent evidences has suggested that increase cholesterol levels have also been found to be a risk factor for Alzheimer disease⁽⁵⁵⁾.

2-Compared of ova-lacto vegetarianisms with non-vegetarianism group (omnivores):

The results showed a significant increase of ova-lacto vegetarianism in: vitamin E (P=0.034), β -carotene(P=0.039), vitamin C (P=0.004), GSH (P=0.0001) and glucose (P=0.023) when compared with non-vegetarianisms. While significant decreased in: Cp. (P=0.016), total protein (P=0.001), calcium (P=0.046), TIBC (P=0.033), MDA (P=0.029), peroxynitrate (P=0.011), GST (P=0.041) and BMI (P=0.034) Similar results were published by other investigators^(56, 57).

Table 3: The biochemical parameter of ova-lacto vegetarianisms and non-vegetarianisms

Parameters	Non-vegetarianism (n=66)		Ova-lacto vegetarianisms (n=62)		t-test
	Mean	SD	Mean	SD	P-value
Age (year)	30.04	1.54	30.96	1.33	0.037*
B.M.I (k.g./m ²)	24.43	1.01	26.25	1.21	0.021*
Vit.E (mg/dl)	1.14	0.09	1.51	0.18	0.034*
Vit.A (µg/dl)	47.66	3.1	57.55	3.52	0.809
β-carotene (µg/dl)	59.08	8.6	76.14	6.47	0.039*
Vit.C (mg/dl)	0.77	0.07	0.84	0.1	0.004*
Cp. (mg/l)	145.29	14.05	158.01	15.59	0.016*
T.p. (gm/dl)	5.03	0.24	5.81	0.18	0.001*
Alb. (gm/dl)	3.92	0.1	4.55	0.19	0.263
Calcium (mg/dl)	9.20	0.19	12.77	0.13	0.046*
Total Bilir. (mg/dl)	0.32	0.01	0.155	0.02	0.2
Uric acid (mg/dl)	4.94	0.1	5.43	0.29	0.55
Creatinine (mg/dl)	3.66	1.4	2.48	0.19	0.224
TIBC (µg/dl)	290.97	35.37	480.47	111.97	0.033*
Iron (mg/l)	1.27	0.23	1.08	0.1	0.114
GSH (µmol/l)	10.84	0.73	12.65	0.66	0.0001**
MDA (µmol/l)	14.46	2.04	7.27	1.09	0.029*
Peroxynitrate	98.64	7.05	105.13	7.05	0.011*
Cholesterol (mg/dl)	172.97	7.00	171.16	5.63	0.189
Glucose (mg/dl)	73.62	3.72	55.03	5.8	0.023*
GST(U/l)	36.12	6.12	25.28	5.08	0.041*

* Different Significantly at $p \leq 0.05$.** High Significantly at $p < 0.0001$.

Ovo-lacto vegetarians consume more fiber and antioxidants when compared to meat eaters. They also consume more phytochemicals (special compounds found in plants)⁽⁵⁸⁾. Many phytochemicals have a wide range of protective effects in the human body. They help to detoxify carcinogens, stimulate the immune system, regulate cell growth (which helps to fight cancer), and reduce some of the harmful effects of excessive exposure to hormones. Vegetarians do not eat red meat. Red meat stimulates the production of mutation-causing compounds in the colon and is linked to increased damage to DNA⁽⁵⁹⁾. Therefore observed to significantly increase to vitamin E, β-carotene, vitamin C, GSH and decreased in

MDA, Cp. and GST. The antioxidant status could be used as a biomarker to assess chronic disease risk and diet can modulate antioxidant defence⁽⁶⁰⁾.

Proteins are composed of amino acids, and a common concern with protein acquired from vegetable sources is an adequate intake of the essential amino acids, which cannot be synthesised by the human body⁽⁶¹⁾. While dairy and egg products provide complete sources for lacto-ovo vegetarians. However, the essential amino acids can also be obtained by eating a variety of complementary plant sources that, in combination, provide all eight essential amino acids (e.g. brown rice and beans, or hummus and whole wheat pita, though protein combining in the same meal is not

necessary). A varied intake of such sources can be adequate⁽⁶²⁾.

A lacto-ovo vegetarian typically consumes less saturated fat and cholesterol than someone who eats meat. A high-fiber, low-fat vegetarian diet, combined with other lifestyle changes, might even reduce atherosclerosis, or hardening of the arteries. Incorporating soy based foods, whole grains, and a wide variety of vegetables may lower glucose without medication⁽⁶³⁾.

Increase of calcium in ova-lacto vegetarianism because of the calcium-rich plant foods such as dark-green vegetables and from milk was observed⁽¹⁾.

2-Comparison between vegetarians and ova-lacto vegetarians:

The results showed a significant increase of vegetarianism in: vitamin E (P=0.001), β -carotene (P=0.016), vitamin C (P=0.005), total bilirubin (P=0.002), GSH (P=0.044). While significant decreased in: Cp. (P=0.041), calcium (P=0.046), TIBC (P=0.031), MDA (P=0.001), peroxynitrate (P= 0.001), GST (P=0.031) and BMI (P=0.021) when compared with ova-lacto vegetarians, these results were similar to other reported results^(56, 64, 65)

Ovo-lacto vegetarian diets omit all animal and animal derived foods except for milk, milk products, and eggs. These differences among vegetarian diets could vary greatly in energy, fat, and cholesterol content. These diets require careful planning as dietary fat, saturated fat, cholesterol (except in vegan diet), and energy content can easily exceed recommended intake levels if nuts, coconut, cooking oils, eggs, butter, cheese, full-fat milk products are frequently and abundantly consumed as part of the vegetarian diet⁽⁶⁶⁾.

It was observed from the data listed in Table (4) that there were significant increase of antioxidant compounds(vitamin E, β -carotene, vitamin C, total bilirubin and GSH) in vegetarians and significant decrease in oxidant compounds (MDA and peroxynitrate) when compared with ova-lacto vegetarians refer this results a decrease of oxidative stress in vegetarians more than ova-lacto vegetarians and Oxidative stress can be defined as an imbalance between the amount of oxidants (Reactive oxygen species (ROS), Reactive nitrogen-oxygen species(RNOS) and others) with the intracellular and extracellular antioxidant protection systems (Figure1)⁽⁴²⁾.

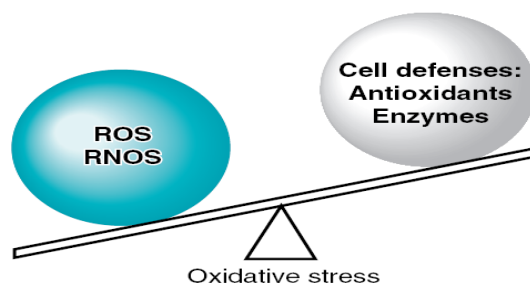


Figure 1:Oxidative stress occurs when the rate of ROS and RNOS production overbalances the rate of their removal by cellular defense mechanisms.

Oxidative stress is involved in the process of aging and various chronic diseases such as atherosclerosis, diabetes, and eye disease. Oxidative DNA damage might be important in mutagenic, carcinogenic, and aging processes^(34, 35). Although it is plausible that antioxidant vitamins might reduce oxidative DNA damage, evidences for favorable effects of vitamin

supplements in human studies are partly inconsistent^(67, 68). It is evident that antioxidant intake through consumption of antioxidant- rich foods was more effective at lowering DNA damage than supplementation with single antioxidants. The benefits of fruit and vegetable intake became evident with the consumption being at least three servings/day⁽⁶⁹⁾.

Table 4: The biochemical parameter of vegetarianisms and ova-lacto vegetarianisms.

Parameters	Vegetarianism (n=54)		ova-lacto vegetarianisms (n=62)		t-test
	Mean	SD	Mean	SD	P-value
Age (year)	34.1	1.91	30.96	1.33	0.024*
B.M.I (k.g./m ²)	23.45	1.21	26.25	1.21	0.021*
Vit.E (mg/dl)	1.88	0.11	1.51	0.18	0.001*
Vit.A (µg/dl)	56.49	8.3	57.55	3.52	0.251
β-carotene (µg/dl)	84.47	11.18	76.14	6.47	0.016*
Vit.C (mg/dl)	1.09	0.09	0.84	0.1	0.005*
Cp. (mg/l)	112.9	10.56	158.01	15.59	0.041*
T.p. (gm/dl)	5.93	0.17	5.81	0.18	0.861
Alb. (gm/dl)	4.74	0.16	4.55	0.19	0.711
Calcium (mg/dl)	10.07	0.69	12.77	0.13	0.046*
Total Bilir. (mg/dl)	0.24	0.02	0.155	0.02	0.002*
Uric acid (mg/dl)	4.87	0.18	5.43	0.29	0.29
Creatinine (mg/dl)	2.75	0.11	2.48	0.19	0.478
TIBC (µg/dl)	415.32	84.64	480.47	111.97	0.031*
Iron (mg/l)	1.52	0.21	1.08	0.1	0.641
GSH (µmol/l)	15.8	0.4	12.65	0.66	0.044*
MDA (µmol/l)	5.01	0.35	7.27	1.09	0.001*
Peroxynitrate	77.45	6.23	105.13	7.05	0.001*
Cholesterol (mg/dl)	163.87	5.14	171.16	5.63	0.235
Glucose (mg/dl)	82.99	7.05	55.03	5.8	0.12
GST(U/l)	19.29	3.16	25.28	5.08	0.031*

* Different Significantly at $p \leq 0.05$.

** High Significantly at $p < 0.0001$.

The health status of vegetarians can be assessed through several approaches. Dietary intake and nutritional status can be measured and compared with reference values. Other indexes of health can be measured, such as body mass index (BMI), serum lipid concentrations, and blood

pressure, and the likely effect on health and disease risk can be estimated⁽⁷⁰⁾, therefore the vegetarians more healthy compared with ova-lacto vegetarians because normal BMI value⁽⁷¹⁾.

The results of present study suggest that an increase of oxidative

damage in diseases, pollutions, aging and others might be prevented by vegetarian nutrition rather than ova-lacto vegetarian nutrition or non vegetarian nutrition respectively.

References

- 1-Key T. J., Appleby P. N., Rosell M. S. Health effects of vegetarian and vegan diets. *Proceedings of the Nutrition Society*, 2006, **65**, 35-41.
- 2- Kapiszewska M. A vegetable to meat consumption ratio as a relevant factor determining cancer preventive diet. The Mediterranean versus other European countries. *Forum. Nutr.* 2006, **59**, 130-153.
- 3- Rinzler C. A. Nutrition For Dummies. 4th Ed. Published by Wiley Publishing, Inc. Indianapolis, Indiana . 2006. pp.330.
- 4- Forrest, J. (December 18, 2007). Is Cheese Vegetarian?. Serious Eats.
<http://www.seriousseats.com/2007/12/is-cheese-vegetarian.html>. Retrieved 9 July 2010.
- 5- Laskowska-Klita T., Chelchowska M., Ambroszkiewicz J., Gajewska J., Klemarczyk W. The effect of vegetarian diet on selected essential nutrients in children. *Med. Wieku. Rozwoj.*, 2011, **15**(3):318-25.
- 6- Krajcovicová-Kudlácková M., Valachovicová M., Pauková V.,(2008). Effects of diet and age on oxidative damage products in healthy subjects. *Physiol. Res.*, 2008, **57**(4), 647-651.
- 7- Liang, L., D'Haese, P., Lamberts, L.V. , DeBroe, M.E.(1989). Direct determination of iron in urine and serum using graphite furnace atomic absorption spectrometry. *Analysis*, 1989, **114**, 143-147.
- 8- Emmerie, A. ,Engel, C. *Nature*, 1938, **142**, 873. Cited by Glick D. .Methods of biochemical analysis .volume (2).John Wiley and Sons, Inc. 1967, p.18.
- 9- Neeld, J.B. ,Person, W.N. *J. Nutr.*, 1963, **79**,454. Cited by Glick D..Methods of biochemical analysis .volume (15).John Wiley and Sons ,Inc.1967, p.18.
- 10- Roe, J. H. , Kuther, C.H. *J. Biol. Chem.*, 1943, **147**, 399. Cited by McMornick, D.B. , Wright, L. D. Methods in enzymology. Volume (62)“(Vitamins and co enzymes” .Part D. Academic press,Inc., 1979, p.7.
- 11- Sunderman, F.W., Nomato, S.Measurement of human serum ceruloplasmin by its para phenylenediamine oxidase activity. *Clin. Chem.*, 1970, **16**(11), 903-910.
- 12- Kingsley, G.R. The direct biuret method for the determinations of serum proteins as applied to photoelectric and visual colorimetry. *J. Clin. Lab.*, 1942, **27**, 840-847.
- 13- Doumas, B.T., Waston, W.A., Bigg, H.G. Albumin standards and the measurement of serum albumin with BCG. *Clin Chim Acta.*, 1971, **31**, 87-96.
- 14- Rbertson, W.G. , Marshall, R. W.Calcium measurments in serum and plasma : total and ionized. *Crit. Rev. Clin. Lab. Sci.*, 1979, **11**, 271-304.
- 15- Toro, G., Ackermann P. G. Practical clinical chemistry. Little , Brown and Company(Inc.).USA., 1979, pp.497-506.
- 16- Varley, H. Practical clinical biochemistry.4th ed. The White Friars Press Ltd.UK., 1967, p.82.
- 17- Jaffě, M.Uber,denNiederschlag, Wwlchen Pikrinsaure in normalem harn erzeugt and uber

- eine neue reaktion des creatinine. Hoppe- Seylers Z. *Physiol. Chem.*, 1886, **10**, 391.
- 18- William, L., White, J.M., Flashka, C. M. The measurment of iron and total iron binding capacity by using calorimetric test (ferrozine). *Clin. Chem.*, 1977, **23**, 237-239.
- 19- Sedlak, J. , Lindsay, R.H. *Anal. Biochem.*, 1968, **192**.Cited by Al-Zamely , O.M. , Al-Nimer M.S., Al-Muslih R.K. Detection the level of peroxyinitrite and related with antioxidant satus in the serum of pateints with acute myocardial infarction. *Nation. J.Chem.*, 2001, **4**, 625-637.
- 20- Lunec, J. *Review Articale, Ann. Clin. Biochem.*, 1990, **27**, 173.Cited by Muslih R. K. , Al-Nimer M.S. ,Al-Zamely O. M.Y.The level of malondialdehyde after activation with (H₂O₂ and CuSO₄) and inhibition by desferoxamine and molsidomine in the serum of patient with acute myocardial infarction. *Nation. J. Chem.*, 2001, **5**,139-148.
- 21- Vanuffelen, B. E., Van Derzec, J., Dekoster, B. M. *Biochem J.*, 1998, **330**,719. Cited by Al-Zamely et al. 2001.
- 22- Richmond, W. Preparation and properties of a cholesterol oxidase from Nocardia Sp. And its application to the enzymatic assay of total cholesterol. *Clin. Chem.*, 1973, **19(12)**, 1350-1356.
- 23- Trinder, P. Determiation of glucose in blood using glucose oxidase with an alternative oxygen acceptor. *Ann. Clin. Biochem.*, 1969, **6**, 24-27.
- 24- Habig, W. H.; Pabst, M. J.; Jakoby, W. B. Glutathione-S-transferase, the first enzymatic step in mercapturic acid formation. *J. Biol. Chem.*, 1974, **249(22)**,7130-7139.
- 25- Al-Abbad, F. A.; Al-Sowielem, L. S. Prevalence of obesity. *Saudi Med. J.*, 1998, **19(5)**, 608.
- 26- Gorczyca, D., Prescha, A., Szeremeta, K., Jankowski, A. Iron status and dietary iron intake of vegetarian children from Poland. *Ann. Nutr. Metab.*, 2013, **62(4)**, 291-7.
- 27- Chełchowska M., Ambroszkiewicz J., Klemarczyk W., Gajewska J., Ołtarzewski M., Laskowska-Klita T. Influence of vegetarian diet on serum values of homocysteine and total antioxidant status in children. *Pol. Merkur. Lekarski.*, 2010, **29(171)**,177-180.
- 28- De Biase S.G., Fernandes S.F, Gianini R.J., Duarte J.L. Vegetarian diet and cholesterol and triglycerides levels. *Arq. Bras. Cardiol.*, 2007, **88(1)**, 35-39.
- 29- Kazimírová A., Barancoková M., Krajcovicová-Kudlácková M., Volkovová K., Staruchová. The relationship between micronuclei in human lymphocytes and selected micronutrients in vegetarians and non-vegetarians. *Mutat. Res.*, 2006, **611(1-2)**, 64-70.
- 30- Szeto Y.T., Kwok T.C., Benzie I.F. Effects of a long-term vegetarian diet on biomarkers of antioxidant status and cardiovascular disease risk. *Nutrition.*, 2004, **20(10)**, 863-6.
- 31- Rauma A.L., Mykkänen H. Antioxidant status in vegetarians versus omnivores. *Nutrition*, 2000, **16(2)**, 111-119.
- 32- Krajcovicová-Kudlácková M., Simoncic R., Béderová A., Klvanová J. Lipid and antioxidant blood levels in

- vegetarians. *Nahrung*, 1996, **40(1)**, 17-20.
- 33- Pronczuk A., Kipervarg Y., Hayes K.C. Vegetarians have higher plasma alpha-tocopherol relative to cholesterol than do nonvegetarians. *J. Am. Coll. Nutr.*, 1992, **11(1)**, 50.
- 34- Li, D. Effect of the vegetarian diet on non-communicable diseases. *J. Sci. Food Agric.*, 2013, Aug 21.[Epub ahead of print]
- 35- Fraser G.E., Lindsted K.D., Beeson W.L. Effect of risk factor values on lifetime risk of and age at first coronary event. The Adventist Health Study. *Am. J. Epidemiol.*, 1995, **142**, 746-58.
- 36- Appleby P.N., Thorogood M., McPherson K., Mann J.I.(1995). Associations between plasma-lipid concentration and dietary, lifestyle and physical factors in the Oxford Vegetarian Study. *J. Human Nutr. Diet*, 1995, **8**, 305-14.
- 37- Kim, M.K., Cho, S.W., Park, Y.K. Long-term vegetarians have low oxidative stress, body fat, and cholesterol levels. *Nutr. Res. Pract.*, 2012, **6(2)**,155-61.
- 38- Pietinen P., Rimm E.B., Korhonen P. Intake of dietary fiber and risk of coronary heart disease in a cohort of Finnish men. The Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study. *Circulation*, 1996, **94**, 2720-7.
- 39- Somannavar M.S., Kodliwadmth M.V. Correlation between oxidative stress and antioxidant defence in South Indian urban vegetarians and non-vegetarians. *Eur. Rev. Med. Pharmacol. Sci.*, 2012, **16(3)**, 351-4.
- 40- Wang, Y., Yang, M., Lee, S.G., Davis, C.G., Koo, S.I., Chun, O.K. (2012). Dietary total antioxidant capacity is associated with diet and plasma antioxidant status in healthy young adults. *J Acad Nutr Diet*, 2012, **112(10)**, 1626-35.
- 41- Yu, L. Wheat antioxidants. John Wiley and Sons, Canada., 2008, pp.46,48,237.
- 42- Banerjee, R. Redox biochemistry. by John Wiley and Sons, Inc. New Jersey, Canada., 2008, pp.11,183.
- 43- López-Tinoco C., Roca M., García-Valero, A., Murri M., Tinahones F.J, Segundo C., Bartha JL, Aguilar-Diosdado M. Oxidative stress and antioxidant status in patients with late-onset gestational diabetes mellitus. *Acta. Diabetol.*,2011.
- 44- Vegan Sources of Calcium". <http://www.vegansociety.com/food/nutrition/calcium.php>. Retrieved Nov. 01 2009.
- 45- Venderley A.M., Campbell W.W. Vegetarian diets : nutritional considerations for athletes. *Sports Med.*, 2006, **36(4)**, 293-305.
- 46- Glantzounis, G. K.; Tsimoyiannis, E. C.; Kappas, A. M.; Galaris, D. A. (2005). Uric acid and oxidative stress. *Current Pharmaceutical Design*, 2005, **11(32)**, 4145-4151.
- 47- Mattson, M. P. Diet-Brain Connection: Impact on Memory, Mood, Aging and Disease. Kluwer Academic Publishers.2002.
- 48- Ginter E. Vegetarian diets, chronic diseases and longevity. *Bratisl. Lek. Listy*.109(10), 2008, 463-6.
- 49- Guggenbühl N. The source of antioxidants. *Bull. Soc. Belge. Ophtalmol.*, 2006, **(301)**, 41-5.
- 50- Del Rio, D., Stewart, A.J., Pellegrini, N. A review of recent studies on malondialdehyde as toxic molecule and biological marker of oxidative stress" .

- Nutr. Metab. Cardiovasc. Dis.*, 2005, **15** (4), 316–28.
- 51- Jenkins D.J., Kendall C.W., Marchie A., Faulkner D.A., Wong J.M., de Souza R. Effects of a dietary portfolio of cholesterol-lowering foods vs lovastatin on serum lipids and C-reactive protein. *J. Am. Med. Assoc.*, 2003, **290**, 502–510.
- 52- Alexander D., Ball M.J., Mann J. Nutrient intake and hematological status of vegetarians and age-sex matched omnivores. *Eur. J. Clin. Nutr.*, 1994, **48**, 538–46.
- 53- Locong A. Nutritional status and dietary intake of a selected sample of young adult vegetarians. *J. Can. Diet Assoc.*, 1986, **47**, 101–6.
- 54- Rizos E., Mikhailidis D.P. Are high density lipoprotein (HDL) and triglyceride levels relevant in stroke prevention? *Cardiovasc Res.*, 2001, **52**, 199-207.
- 55- Simons M., Keller P., Dichgans J., Jörg B., Schulz J.B. Cholesterol and Alzheimer's disease. Is there a link? *Neurology*, 2001, **57**, 1089-93.
- 56- Gammon C. S., von Hurst P. R., Coad J., Kruger R. Vegetarianism, vitamin B12 status, and insulin resistance in a group of predominantly overweight/obese South Asian women. *Nutrition*, 2012, **28**, 20–24.
- 57- Chiplonkar S.A, Tupe R. Development of a diet quality index with special reference to micronutrient adequacy for adolescent girls consuming a lacto-vegetarian diet. *J. Am. Diet. Assoc.*, 2010, **110**(6), 926-931.
- 58- Davey G.K., Spencer E.A., Appleby P.N., Allen N.E., Knox K.H., Key T.J. EPIC-Oxford: lifestyle characteristics and nutrient intakes in a cohort of 33 883 meat-eaters and 31 546 non meat-eaters in the UK. *Public Health Nutr.*, 2003, **6**(3), 259-269.
- 59- Hubbard R.W., Mejia A., Horning M. The potential of diet to alter disease processes. *Nutr. Res.*, 1994, **14**, 1853-95.
- 60- Haldar S., Rowland I.R., Barnett Y.A., Bradbury I., Robson P.J., Powell J., Fletcher J. Influence of habitual diet on antioxidant status: a study in a population of vegetarians and omnivores. *Eur. J. Clin. Nutr.*, 2007, **61**(8), 1011-1022.
- 61- Davis B., Melina V. The New Becoming Vegetarian. Book Publishing Company., 2003, pp. 57–58.
- 62- Young V.R., Pellett P.L. Plant proteins in relation to human protein and amino acid nutrition. *Am. J. Clinical Nutrition*, 1994, **59** (59), 1203S–1212S.
- 63- Fernandes D. K., de Arruda C. E., Siqueira C. F., Sakugava Sh. N.K. Relation between dietary and circulating lipids in lacto-ovo vegetarians. *Nutr. Hosp.*, 2011, **26**(5), 959.
- 64- Krajcovicova-Kudlackova M., Babinska K., Blazicek P., Valachovicova M., Spustova V. Selected biomarkers of age-related diseases in older subjects with different nutrition. *Bratisl. Lek. Listy.*, 2011, **112**(11), 610-3.
- 65- Toohey M. L., Harris M. A., Williams D., Schmidt W. D. Cardiovascular Disease Risk Factors are Lower in African-American Vegans Compared to Lacto-Ovo-vegetarians. *J. Amer. Coll. Nutr.*, 1998, **17**(5), 425–434.

- 66- Malcolm K. R., Thomas A. Obesity and Cardiovascular Disease. Taylor and Francis Group, LLC. USA., 2006, Pp.309.
- 67- Winckel M. V., Velde S. V., Bruyne R. D., Biervliet S. V. Vegetarian infant and child nutrition. *Eur. J. Pediatr.*, 2011, **170**, 1489–1494.
- 68- Moller P., Loft S. Dietary antioxidants and beneficial effect on oxidatively damaged DNA. *Free Radic. Biol. Med.*, 2006, **41**, 388-415.
- 69- Huang H.Y., Helzlsouer K.J., Appel L.J. The effects of vitamin C and vitamin E on oxidative DNA damage, results from a randomized controlled trial. *Cancer Epidemiol. Biomarkers Prev.*, 2000, **9**, 647-652.
- 70- Key T. J., Appleby P. N., Spencer E. A., Travis R. C, Roddam A. W, Allen.N. E. Mortality in British vegetarians: results from the European Prospective Investigation into Cancer and Nutrition (EPIC-Oxford). *Am. J. Clin Nutr*, 2009, **89(suppl)**,1613S.
- 71- Inoue S., Zimmet P. The Asia-Pacific Perspective: Redefining Obesity and Its Treatment". Published by Health Communications Australia Pty Limited on behalf of the Steering Committee., 2000, pp.17.