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HEMATOLOGICAL STATUS AND FUNCTIONAL ABILITIES OF THE CARDIORESPIRATORY SYSTEM OF PHYSICALLY ACTIVE FEMALE VEGANS

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ABSTRACT

The aim of this study was to examine the influence of vegan diet on hematological parameters, and to compare the functional abilities of the cardiorespiratory system of vegans and counterparts on a standard diet. The sample consisted of 24 females, divided into two groups: 11 vegans and 13 women on a standard diet. Assessment of the level of physical activity of the participants was estimated by the standard IPAQ questionnaire. Hematological analysis included: complete blood count, blood lipid profile, vitamin B12 and iron. Basic anthropometric variables were monitored, and a submaximal Astrand test on a bicycle ergometer was used to estimate maximal oxygen consumption. The RAMP continuous protocol was used to measure the maximum values of heart rate and blood pressure. The results showed that active vegans have a significantly lower body weight and body mass index compared to active women on a standard diet. The number of erythrocytes and platelets, as well as LDL - cholesterol show lower values in vegans compared to the control group ($p<0.01$). Indicators of the functional abilities of the cardiorespiratory system do not differ between active vegans and women on a standard diet.

Keywords: VEGAN DIET / BLOOD TEST / LIPID PROFILE / IRON / VITAMIN B12 / VO_2max

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INTRODUCTION

The vegan diet, except that excludes all animal foods, i.e. meat, fish, dairy products, eggs and even honey, is basically a special philosophy of life that includes an altruistic, humane and ethical attitude towards the environment and animal protection.

Previous studies have shown that plant-based diets have a number of health benefits. Thus, vegetarians and vegans have a lower mortality rate (Kahleova, Levin, & Barnard, 2017; Dinu, Abbate, Gensini, Casini, & Sophie, 2017), are less likely suffer from malignant diseases (Dinu et al., 2017; Key, Appleby, Spencer , Travis, Roddam, & Allen, 2009), have a lower risk for developing obesity (Kahleova, Dort, Holubkov, & Barnard, 2018; Kahleova, Hlozkova, Fleeman, Fletcher, Holubkov, & Barnard; 2019). Also, a diet that excludes animal foods of has been shown to have positive effects on the regulation of metabolic diseases, such as diabetes and insulin resistance (Kahleova et al., 2018; Kahleova et al., 2019; Barnard, Katcher, Jenkins, Cohen, & Turner-McGrievy, 2009). Vegans generally have lower blood pressure (Kahleova et al., 2017; Appleby, Davey, & Key, 2002), lower levels of total and low-density lipoprotein cholesterol–LDL (Yokoyama, Levin, & Barnard, 2017; Fontana, Meyer, Klein, & Holloszy, 2007) and are less likely to develop hypertension (Kahleova et al., 2017; Appleby et al., 2002), coronary heart disease and have a lower risk of cardiovascular disease (Kahleova et al., 2017; Yokoyama et al., 2017; Fontana et al., 2007).

On the other hand, vegan diet can have some negative effects on health. In particular, due to the exclusion of animal food, vegans generally have lower levels of cyanocobalamin (vitamin B12) (Gallego-Narbón, Zapatera, Barrios, & Vaquero, 2019; Clarys, Deriemaeker, & Hebbelinck, 2000). Also, due to lower values of high-density lipoprotein - HDL (Linsel-Nitschke & Tall, 2005) and higher values of serum homocysteine (Gallego-Narbón et al., 2019; Elevated, Case, & Nineteen, 1997), which is an indicator of deficiency of vitamins B12 and folic acid, the risk of atherosclerosis in vegans might be increased. The analysis of the available literature shows that there are a limited number of studies that have studied the effects of a plant-based diet on the functional and motor abilities of physically active subjects. Two studies have shown that physically active vegetarians have higher values of functional abilities of the cardiorespiratory system, i.e. maximal oxygen consumption compared to athletes on a standard diet (Veleba, Matoulek, Hill, Pelikanova, & Kahleova, 2016; Lynch, Wharton, & Johnston, 2016). On the other hand, Nebl et al (2019) did not determine the benefits of a vegan or vegetarian diet on motor and functional abilities.

Given that specific nutritional habits and training are considered the most important factors influencing changes in the morphological and functional abilities of the individuals, the aim of this study was to examine the impact of vegan diet on hematological parameters, as well as to compare the functional abilities of the cardiorespiratory system of vegans and counterparts on a standard diet.

METHODS

Subjects

The inclusive criterion for participation in the study was that the subjects were on a vegan or standard diet for at least a year, as well as to train regularly in moderate and high intensity of at least 3 hours a week in some type of the endurance sports.

The sample consisted of a total of 24 females, divided into two groups: experimental (11 vegan) and control group (13 women on a standard diet). An IPAQ questionnaire was used to assess the level of

physical activity (Vandelanotte, De Bourdeaudhuij, Philippaerts, Sjöström, & Sallis, 2005), while questionnaire was used to assess the average daily intake of macronutrients (Heaney, Connor, Gifford, & Naughton, 2010). The groups of participants were uniform according to gender, age and weekly volume of physical activity.

Experimental protocol and measurement procedures

The experimental protocol was realized in two sessions. The first included blood sampling, which was performed in the morning, and the subjects received the necessary instructions on taking food and drinks. Haematological analysis included:

1. Complete blood count, measured on an automated haematology analyser (CoulterHmX, Beckman Coulter, Miami, Florida);
2. Lipid profile (Hitachi 911, Tokyo, Japan)
3. Vitamin B12 and iron (F2 +) measured by radioimmunoassay method (Diagnostic Products Corp, USA).

The second session included anthropometric measurements and measurements of the functional abilities of the cardiorespiratory system. From the anthropometric variables were measured body height (Martin`s Anthropometer) and body weight (*Portable* scales - Tanita Europe GmbH, Sindelfingen, Germany). The calculation of body mass index (BMI) was performed based on the formula: BMI (kg/ m^2) = TM (kg) / TV (m^2).

A submaximal Astrand test on a bicycle ergometer (*Kettler Ergometer E5 - Kettler, Germany*) was used to estimate the maximum oxygen consumption (VO_2max). To measure the maximal values of heart rate and blood pressure, the RAMP continuous protocol on a bicycle ergometer with an initial load of 50W and increments of 25W per 1 minute was used. Heart rate was measured using the Polar RS400 pulse meter, and blood pressure was measured with a gas manometer (*Gimi, Italy*) by an indirect auscultatory method.

Statistical Analyses

Statistical processing was performed in Microsoft Excel 2010 and SPSS21.0. Central tendency and dispersion were used within the descriptive statistics. T-test for independent samples was used to test the differences between the vegan and the control group for haematological parameters, anthropometric characteristics and functional abilities of the cardiorespiratory system. The level of significance was determined for $p<0.05$.

RESULTS

The physical characteristics of the participants and their weekly volume of physical activity are shown in Table 1. The participants were stratified by age, height and volume of physical activity.

Table 1. The physical characteristics and training volume

Variable	Vegans	Control group
	(Mean ± SD)	(Mean ± SD)
Age	34.18 ± 12.21	30.85 ± 10.95
BH (cm)	166.45 ± 7.65	169.92 ± 5.81
BM (kg)	53.98 ± 5.85 **	62.82 ± 4.73
BMI (kg/m2)	19.55 ± 1.92**	21.62 ± 1.33
MET (mlO2/kg/min)	5612± 3252	5537 ± 2379

Legend: SD – standard deviation; BH– body height; BM – body mass; BMI body mass index; MET – metabolic equivalent; *p<0.05; ** p<0.01

Although the average daily energy intake was slightly lower in the group of vegans (-157.52 Kcal day, r>0.05), the ratio of macronutrients in the diet shows that vegans consume a significantly higher amount of carbohydrates compared to the control group (Table 2).

Table 2. Average daily energy intake

Variable	Vegans		Control group	
	mean ± SD	Relative value	mean ± SD	Relative value
Energy intake (Kcal)	2228.35 ± 88.68	-	2385.77 ± 290.44	-
Carbohydrates (g, g/kg)	361.95 ± 20.46	6,78 ± 0,80**	310 ± 32.93	4.96 ± 0.65
Fats (g, g/kg)	52.39 ± 7.71	0.98 ± 0.19*	79.31 ± 15.70	1.73 ± 0.29
Proteins (g, g/kg)	77.27 ± 10.76	1.45 ± 0.29**	108 ± 14.54	1.28 ± 0.31
Dietary fibers (g, g/kg)	52.45± 10.91	0.97 ± 0.20**	31±9.30	0.50 ± 0.16

*p<0.05; ** p<0.01.

Ratio of the carbohydrates, fats and proteins, observed in the total daily intake in vegans was 65, 21.1 and 13.9%, while in the control group it was 52, 29.9 and 18.1%, respectively (p<0.05).

The observed haematological variables indicate that vegans have lower values of erythrocytes, haemoglobin and haematocrit, while within the lipid profile only LDL showed lower values compared to the control group (Table 3).

Table 3. Hematological parameters

	Variable	Reference value	Vegans (mean ± SD)	Control group (mean ± SD)
Blood test results	WBC (x 10^9/L)	4.0 - 10	5.48 ± 1.40	6.34 ± 1.45
	RBC (x 10^{12}/L)	4.5 - 5.2	4.39 ± 0.43 *	4.73 ± 0.35
	Hb (g/l)	115 - 160	131.36 ± 11.78 *	138.46 ± 7.80
	Hct (l/l)	0.42 - 0.53	0.41 ± 0.04 *	0.43 ± 0.03
	MCV (fl)	83 - 97	92.71 ± 2.85 *	89.66 ± 3.77
	MCH (pg)	27 - 33	30.35 ± 1.29	30.05 ± 1.57
	MCHC (g/l)	320 - 360	327 ± 11 *	337.54 ± 7.28

	PLT (x 10^9/L)	158 - 424	228 ± 38 **	291.25 ± 39.27
	Fe^{2+} (µmol/l)	9 - 30.4	20.45 ± 5.01	21.73 ± 4.88
	B_{12} (pmol/L)	145 - 637	410 ± 225	465.43 ± 150.19
Lipid profile	Hol (mmol/l)	3.1 - 5.2	4.07 ± 0.81	4.75 ± 0.94
	HDL (mmol/l)	1 - 1.6	1.72 ± 0.63	1.40 ± 0.33
	LDL (mmol/l)	0 - 3.4	1.97 ± 0.79 *	2.95 ± 0.79
	Triglycerides (mmol/l)	0 - 1.7	0.75 ± 0.23	0.85 ± 0.32
	Atherosclerosis index	0 - 3	1.38 ± 0.82	2.16 ± 0.60
	Risk factor	0 - 4.5	2.58 ± 0.86	3.45 ± 0.64

Legend: SD – standard deviation; WBC – white blood cells; RBC – red blood cells; Hb – hemoglobin; Hct – haematocrit (proportion of blood cells in total blood volume); MVC – mean erythrocyte volume; MCH – average amount of haemoglobin in erythrocyte; MCHC – mean haemoglobin concentration in erythrocytes; Fe^{2+} - iron; B_{12} – cyanocobalamin (vitamin B12); Hol – cholesterol; HDL - high-density lipoprotein cholesterol; LDL - low-density lipoprotein cholesterol; *p<0,05; ** p<0,01

There are no significant differences in the indicators of functional abilities of the cardiorespiratory system measured at rest and during the all-out test on a bicycle ergometer between the two groups (Table 4). Interestingly, physically active vegans presented slightly higher values of VO_2max compared to women on a standard diet (~ 7 ml/kg/min, r>0.05).

Table 4. Functional abilities of the cardiorespiratory system

Variable	Vegans (mean ± SD)	Control group (mean ± SD)
TA_SIS_{MIR} (mmHg)	118.90 ± 21.35	111.15 ± 8.70
TA_DIJ_{MIR} (mmHg)	76.60 ± 13.99	76.54 ± 5.16
HR_{MAX} (otk/min)	181.73 ± 13.79	175.31 ± 21.06
TA_SIS_{MAX} (mmHg)	146.45 ± 15.40	148.31 ± 14.74
TA_DIJ_{MAX} (mmHg)	76.36 ± 10.85	70.38 ± 4.31
VO_{2APS} (L/min)	3.25 ± 0.62	2.93 ± 0.63
VO_{2REL} (ml/kg/min)	54.70 ± 8.28	47.42 ± 12.21

Legend: SD – standard deviation; TA_SIS_{MIR} – resting systolic pressure; TA_DIJ_{MIR} – resting diastolic pressure; TA_SIS_{MIR} – maximum systolic pressure; TA_DIJ_{MIR} – maximum diastolic pressure; $VO2_{APS}$ – absolute oxygen consumption; $VO2_{REL}$ – relative oxygen consumption; *p<0.05; ** p<0.01.

DISCUSSION

The results of this study showed that vegans have lower body weight and BMI compared to women on a standard diet, and that the vegan diet does not affect the functional abilities of the cardiorespiratory system. Also, the observed haematological variables indicate that a plant based diet can affect the red blood cell parameters, LDL cholesterol, as well as level of vitamin B12.

Optimal body weight and maintenance of BMI within the reference values (De Onis & Habicht, 1996) are associated with a reduced risk of obesity, cardiovascular, metabolic and other diseases (Flegal, Kit, Orpana, & Graubard, 2013). On the other hand, low BMI values (below 18.5 kg/m^2) may indicate

insufficient or unbalanced intake of macro and micronutrients (malnutrition) which results in health disorders and impaired body composition primarily due to decreased muscle and bone mass (Saunders, Smith, & Stroud, 2011). Vegans generally have lower body weight and BMI compared to people on a standard diet (Kahleova et al., 2018, 2019). A meta-analysis by Dino et al. (2017) that included an analysis of scientific papers with a total sample of 8376 vegans showed that this population has a BMI lower by 1.72 kg/m^2 compared to individuals on the standard regimen. These results are consistent with the results obtained in our study, where vegans have a lower BMI of 2.07 kg/m^2 ($p<0.01$). Although vegans have significantly lower body weight and BMI, the observed variables still remain within the reference values, so we cannot claim that a vegan diet can be a risk factor for malnutrition.

A relatively small number of studies followed haematological variables and the influence of different forms of nutrition on their values. Most studies conducted on a sample of vegetarians and vegans have shown that haemoglobin (Waldmann, Koschizke, Leitzmann, Hahn, 2004; Haddad, Berk, Kettering, Hubbard, & Peters, 1999; Tong et al., 2019), erythrocyte (Dong, & Scott, 1982), leukocytes and platelets values (Tong et al., 2019, Haddad et al, 1999) were lower compared to individuals on a standard diet.

Our results are consistent with the findings obtained in the aforementioned studies. Given the relatively small sample and statistical power in most of the mentioned studies, it cannot be stated with certainty that a plant-based diet may be a risk factor for anaemia, immunodeficiency and/or prolonged bleeding time resulting from pancytopenia. Lower erythrocyte values and haemoglobin concentrations in erythrocytes may be associated with erythrocyte destruction due to intense, prolonged physical exertion and mechanical stress (Nieman, 1999; Telford, Sly, Hahn, Cunningham, Bryant, & Smith, 2003).

Optimal serum lipid concentrations have been associated with a reduced risk of cardiovascular and metabolic diseases (Castelli, 1996; Kuklina, Yoon, & Keenan, 2009). Previous study has shown the positive effects of a plant-based diet on the blood lipid profile. A meta-analysis conducted by Yokoyama et al. (2017) showed that compared to people on a standard diet, vegetarians and vegans have lower values of total cholesterol, LDL, HDL, but not triglycerides. Our study revealed that in physically active vegans only LDL is lower than in the control group, and that other lipid profile parameters are within the reference values (Fontana et al., 2007; Yokoyama et al., 2017).

Due to high concentrations of phytate, tannin and phosphate, and due to lower levels of iron in plant foods, vegans may be at greater risk for sideropenic anaemia, which is characterized by low erythrocyte number and volume and low haemoglobin (Hazell, 1988). However, our results show that, although some red blood cell parameters are lower than the mean reference values, they still remain within the reference range for most subjects. One of the reasons may be the high intake of vitamin C, characteristic of the vegan diet, which facilitates the absorption of iron. Therefore, based on the results of our study, we can conclude that adequate amounts of iron can be provided exclusively through plant foods. One of the characteristics of the vegan diet, presented in numerous studies, is vitamin B12 deficiency in the blood (Gilsing et al., 2010; Woo, Gallego-Narbón, 2019; Waldmann, Koschizke, Leitzmann, & Hahn, 2005). In contrast, our results showed normal levels of this vitamin in the bloodstream of vegans. However, analysis of daily nutrient intake data revealed that 7 of 11 vegans have used vitamin B12 supplements. In further analysis, it was found that vegans who did not use supplements had a vitamin B12 level close to the lower limit of the reference interval (195 ± 81.5 pmol/L), while in another two vegans is noted B12 deficiency (values of less than 145 pmol/L). In this regard, this form of diet can be a risk factor for vitamin B12 deficiency, which implies modification of the diet in terms of intake of foods with a higher content of this vitamin or additional supplementation.

The findings of several studies that have addressed the functional abilities of the cardiorespiratory system of vegetarians indicate that they achieve significantly higher values of VO_2max compared to people on a standard diet (Veleba et al., 2016; Lynch et al, 2016). However, Nebl et al. (2019) examining the functional abilities of the cardiorespiratory system of physically active vegans, did not found differences between vegans, vegetarians and people on a standard diet. Our results are in line with the findings of this study. The higher VO_2max in vegans can be explained by factors such as high carbohydrate intake, lower BMI and fact that vegans who participated in the research were mainly engaged in endurance sports.

CONCLUSION

This study showed that female vegans have significantly lower body weight and BMI compared to omnivorous. In female vegans, there is a tendency towards lower values of some blood cell parameters, while from the parameters of the lipid profile, only LDL shows lower values. In term of vitamin B12 deficiency, vegans are recommended to supplement it, since this is the easiest way to provide its adequate amounts. On the other hand, vegans can provide optimal amounts of iron entirely through plant foods, without additional supplementation. Furthermore, although vegans achieve slightly higher values of VO_2max, it cannot be said with certainty that this type of diet is more suitable for the development of cardiorespiratory endurance. Future studies should include a larger number of participants of both sexes and complete body composition analysis.

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