ECRONICON OPEN ACCESS

EC NUTRITION
Research Article

The Effect of a Six-Month Physical Activity and Nutrition Education Program on Biochemical Health Parameters, Physical Activity and Body Composition in Healthy Young Women

Roberto Ramírez-Domínguez[1]*, Teresa Mingo[2], M Ángeles Ferrer Pascual[3], Isabel Bayona[4], Francisco Navas[1] and Lucía L Pérez- Gallardo[1]

[1]Department of Biochemistry, Molecular Biology and Physiology, University of Valladolid, University School of Physiotherapy, Spain

[2]Department of Surgery, Ophthalmology, Otorhinolaryngology and Physiotherapy, University of Valladolid, University School of Physiotherapy, Spain

[3]Nursing Department, University of Valladolid, University School of Nursing, Spain

[4]Department of Pathological Anatomy, Microbiology, Preventive Medicine and Public Health, Forensic and Legal Medicine, University of Valladolid, University School of Physiotherapy, Spain

***Corresponding Author**: Roberto Ramírez-Domínguez, Department of Biochemistry, Molecular Biology and Physiology, University of Valladolid, University School of Physiotherapy, Spain.

Received: August 23, 2017; **Published:** September 20, 2017

Abstract

It was designed an educational intervention program on nutrition and physical activity (six months) in order to observe its effect on general and specific biochemical parameters, anthropometric parameters, body composition, physical condition (VO2max) and level of physical activity in college women. 28 healthy women were included, aged between 18 - 28. We have measured, the serum glucose, high density lipoprotein, low density lipoprotein, total cholesterol, triglycerides, prealbumin, albumin, leptin and production of malondialdehyde. Before and after the intervention program we have also valuated, body mass index, body composition, VO2max and exercise performance (hours/week). After the intervention program, the albumin concentration increased ($p < 0.05$) and in 25 participants (89.3%) leptin also increased ($p < 0.001$). Both, time spent walking and time spent doing vigorous physical activity decreased ($p < 0.05$), conversely time spent doing moderate intensity activity increased ($p < 0.001$). Before the intervention, a negative correlation of leptin with the % water ($r = -0.664$; $p < 0.001$) and positive correlations of leptin with body mass index ($r = 0.582$; $p < 0.01$) and % fat mass ($r = 0.659$; $p < 0.001$) have been observed. These correlations remained after the intervention. A negative correlations of albumin with triglycerides ($r = -0,404$; $p < 0,05$) and malondialdehyde ($r = -0.419$; $p < 0.05$) or leptin with VO_2max ($r = - 0.498$; $p < 0.05$) have also been observed. With this intervention program, we have managed to slightly modify the physical activity habits in adult healthy women and have also seen an increase in albumin levels and leptin without modification in body composition.

Keywords: *Physical Activity; Nutrition; Body Composition; Health Promotion; Maximum Aerobic Capacity*

Abbreviations

BMI: Body Mass Index; FM: Fat Mass; HDL: High Density Lipoprotein; IPAQ: International Physical Activity Questionnaire; LDL: Low Density Lipoprotein; LH Lutein Hormone; MDA: Malondialdehyde; PA: Physical activity; SD: Standard Deviation; TBF: Total Body Fat; TBW: Total Body Water; VO_2max: Maximum Oxygen Consumption; WHO: World Health Organization

Introduction

Programs of health education promoted by public administrations and private institutions have achieved high levels of success in a relatively short period of time [1]. In a previous study, we found that these programs had contributed positively to improve the eating

habits of schoolchildren [2]. The University stage is a particularly sensitive phase in which lifestyle modifications refers [3]. There are plenty of studies that have described eating habits and lifestyle of students suggesting the necessity to coordinate efforts to reduce the tendency to a sedentary lifestyle and promote adherence to the Mediterranean Diet patterns [4,5]. In addition, the last Health Behaviour in School-aged Children study report for Spain showed that only 24.4% of adolescents reached the recommended physical activity levels for health (according to the WHO recommendations), being boys significantly more active than girls (31.7% and 17.3%, respectively) [6]. The aim of this study was to study biochemical parameters, body composition, cardiopulmonary fitness and physical activity in healthy women before and after six months program of nutrition and exercise intervention.

Materials and Methods

Subjects

28 healthy women between the ages of 18 - 28 were recruited for the present research. We based on the project to promote health among University students, conducted during the academic year 2011 - 2012. Details of the project have been reported previously [7], approval was obtained on 19/10/2011 from the Committee for Clinical Research of the Valladolid University and all patients provided written informed consent. The study was in accordance with the Helsinki Declaration 1964. Informed consent was obtained from all individual participants included in the study.

Anthropometric and body composition assessment

Stature was measured with a sensitive stadiometer; all participants were in anatomical posture and without shoes. Bioimpedance analysis (Tanita BC®418MA Segmental) was used for estimating body composition before and after 6-month physical activity and nutrition education program. Body composition was comprised of, fat mass (FM), total body fat percent (% TBF), total body water percent (% TBW) and body weight. Body mass index (BMI, in kilograms per square meter) was calculated according to the Quetelet formula.

Blood samples and Analysis

Blood samples were collected twice; before and after six months intervention programs in nutrition and physical activity. 5 ml blood samples were taken from antecubital vein, in sitting position, by an experienced staff, in the morning after 12-hour night fasting. Blood samples were centrifuged and transferred to the Laboratory under cold chain conditions. They were stored at −20°C until analysis.

Parameters such as glucose, LDL- and HDL-cholesterol, triglycerides, prealbumin and albumin were examined using an automated analyzer (Cobas 6000). We took as serum reference values of glucose 70 - 100 mg/dl; of triglycerides 50 - 150 mg/dl; of cholesterol 100 - 200 mg/dl; of prealbúmina 20 - 40 mg/dl; of albumin 3.50 - 5.20 g/dl; desirable level for HDL-cholesterol: low risk > 60 mg/dl and high risk < 40 mg/dl and discriminant values for LDL-cholesterol: optimal risk < 100 mg/dl and near optimal risk: 101 - 130 mg/dl.

Serum leptin concentrations were determined in duplicate with an enzyme-linked immunosorbent assay (FLUOstar plate reader BMG Labtech Omega) using the Millipore Reagent Kit (Catalog # EZHL-80SK) with a sensitivity for each assay of 0.195 ng/ml - 25 ng/ml for total leptin.

Malondialdehyde (MDA) levels, for assessing lipid peroxidation, were measured spectrophotometrically by the method of Yagi and Prasad [8].

Physical activity assessment

The short version of the International Physical Activity Questionnaire (available at: http://www.ipaq.ki.se) was used in the survey. This questionnaire, which has been developed by the World Health Organization, asks three specific types of physical activity (PA): walk (going from one place to another for recreation or pleasure); moderate-intensity (carrying light loads, cycling bicycling at a regular pace, or play doubles tennis) and vigorous-intensity (heavy lifting, digging, aerobics, or pedal fast bike). The survey results were analyzed under the guidance for processing IPAQ data [9].

Physical condition assessment

The assessment of the maximum aerobic capacity was conducted through the Forest Service Test [10]. This test consists of up and down a 33 cm step for women for a period of 5 minutes and a rate of 22.5 cycles per minute controlled by a metronome (Digital Metronome Model DM-10. SEIKO). At the end of five minutes of effort pulse is recorded in the sitting position. The maximum oxygen consumption (mlO2/kg/min) it is estimated based on body weight and heart rate recorded in beats per minute (ppm) in a nomogram created for this effect.

Statistical Analyses

All analyses were performed using SPSS version 22.0 (Statistical Package for Social Science Inc., Chicago, IL, USA) for Windows. Results are given as mean ± SD. To verify that the population sample is distributed normally Kolmogorov Smirnov test was used and to compare means the nonparametric Mann-Whitney U test was used. The relationship between the results of leptin and the rest of variables were analyzed by Pearson correlation with a statistical significance level of $p < 0.05$.

Exercise intervention

Subjects participated in a program taught by physical educator teachers. It involved self-monitored endurance, strength, postural and flexibility training. The program was divided into supervised sessions and home-based exercise. During the intervention program, participants learnt exercise techniques in individual or small group sessions one time every three weeks, supervised by a physical educator. They also performed an independent exercise program prescribed by physical educator during 6 months.

After learning basic exercise techniques, participants met regularly with the physical educator in small groups (8 - 10 participants), where they continued to learn techniques for maintaining exercise in their daily lives. There were four purposes for the group sessions: first, to model appropriate exercise and stretching behaviors, therefore assisting participants with learning these exercise-related skills and reducing injury potential; second, to facilitate mechanisms of self-assessment of their fitness level; third, to identify the main individual needs in order to develop personal training to be done at home as well as in the following supervised session; and fourth, to provide social support and reinforcement to participants as they changed exercise behaviors and they were encouraged to exercise either at home or at the study exercise facility during the intervention.

The supervised exercises began with 8 - 10 minutes of warm-up continued by 30 - 40 minutes of endurance and strength training and 10 minutes of stretching and end with 5 minutes of cooldown. Participants were also encouraged to engage in physical activity at home between supervised sessions.

The participants, between supervised sessions, received by email the detailed planning of the physical activity to be performed individually. The scheduled activity included aerobic, strength-endurance, posture and flexibility, moderate to severe intensity and duration of sessions.

Adherence to the exercise intervention was controlled in the supervised sessions with the use of heart rate monitors; daily self-monitoring exercise logs and physical activity questionnaires.

Nutrition Education

Through a series of three sessions, the participants received education in composition of foods, recommended energy intakes, frequency of food consumption recommended and self-monitoring using food diaries to achieve consumption of a Mediterranean-style diet [11]. The dietary advice was tailored to each participant on the basis of 7-day food records. Basic recommendations for macronutrient composition of the diet were as follows: carbohydrates, 50% to 60%; proteins, 13% to 15%; total fat, less than 30%; saturated fat, less than 10%; and cholesterol consumption, less than 300 mg per day. In addition, participants were advised to consume at least between 250 and 300 g of fruits, 125 and 150g of vegetables, they were also encouraged to consume 400g of whole grains (legumes, rice, maize,

and wheat) daily and to increase their consumption of olive oil. Compliance with the program was assessed by attendance at the meetings and completion of the diet diaries.

Results and Discussion

There were no significant differences ($p > 0.05$) between before and after the subjects participation in a six month program of nutrition and exercise in terms of weight averages, BMI averages, % TBF and %TBW (Table 1).

	Pre-intervention Mean ± SD	Post-intervention Mean ± SD	p
BMI (kg/m^2)	22.38 ± 2.75	22.26 ± 2.85	0.286
Weight (kg)	59.49 ± 7.77	59.14 ± 7.82	0.240
% TBW	53.42 ±3.80	53.13 ± 4.02	0.202
% TBF	27.00 ± 5.35	27.38 ± 5.57	0.226

Table 1: *Defining features of participants (n = 28) and statistical significance between pre-intervention and post- intervention program (Mann Whitney U test).*

BMI: Body Mass Index; % TBW: Total Body Water Percent; % TBF: Total Body Fat Percent

After having participated for 6 months in this educative program, although there were no significant differences in body weight, the subjects showed an increase in serum leptin concentrations ($p > 0.001$) and albumin ($p = 0.015$). However, there were no statistically significant differences in biochemical parameters such as: glucose, cholesterol, triglycerides, HDL, LDL and pre-albumin (Table 2).

Biochemical parameters	Pre-educative intervention	Post-educative intervention	p
Glucose (mg/dl)	84.61 ± 6,19	84.93 ± 5,68	0.721
Cholesterol (mg/dl)	183.32 ± 27,87	177.11 ± 23,74	0.110
HDL (mg/dl)	68.18 ± 13,99	65.64 ± 13,03	0.132
LDL (mg/dl)	100.86 ± 23,14	96.25 ± 18,37	0.108
Triglycerides (mg/dl)	71.82 ± 24,81	76.36 ± 24,91	0.139
Prealbumin (mg/dl)	27.65 ± 3,86	27.13 ± 4,46	0.413
Albumin (g/dl)	4.63 ± 0,24	4.76 ± 0,27	0.015
Leptin (ng/ml)	7.21 ± 3,41	11.77 ± 4,84	< 0.001
MDA (µM)	3.87 ± 1,72	4.69 ± 1,54	0.074

Table 2: *Comparison of pre-educative intervention and post-educative intervention (6 months) biochemical variables in the subjects (Mann Whitney U test). Values are mean ± SD.*

HDL: High Density Lipoprotein; LDL: Low Density Lipoprotein; MDA: Malondialdehyde

We also observed significant differences between pre and post intervention in terms of time spent in the 3 categories of activities based on the IPAQ analysis framework [9] (Figure 1). We recorded an increase in the duration of moderate activity and a reduction in time spent in vigorous physical activity and walking.

After excluding participants with missing data (n = 10) analyses of the VO_2max. were performed in the remaining 19 individuals. There were no changes when compared the mlO_2/Kg/min before and after the subject participation in a six month program of nutrition and exercise (38.2 ± 5.9 vs 37.2 ± 3.7; $p = 0.291$).

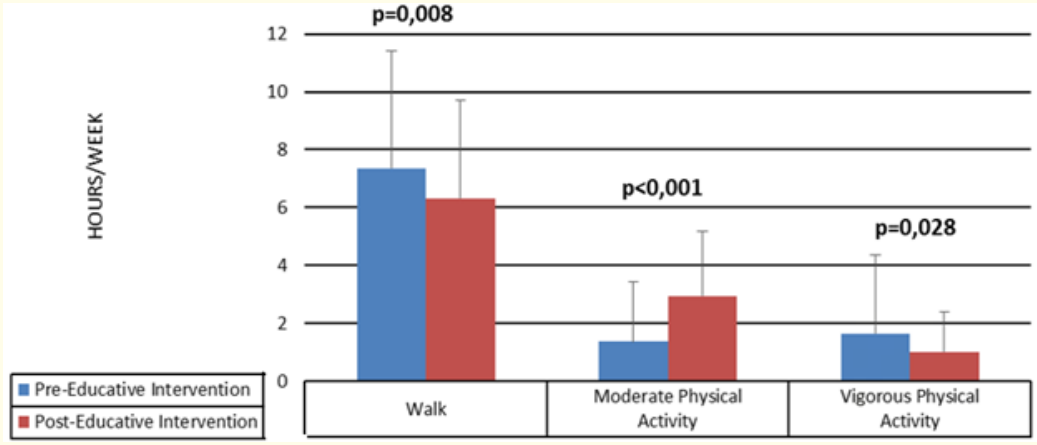

Figure 1: *Change in total time spent (hour/week) at different physical activity intensities pre and post educative intervention in healthy women (n = 28) (Mann Whitney U test). Values are mean ± SD.*

In both stages of the study leptin concentrations were positively correlated with BMI ($p > 0.01$) and % TBF ($p < 0.001$) and negatively correlated with % TBW ($p < 0.001$). After the subject participation in the intervention educative program, a moderately negative correlation between leptin and VO2max was observed ($p < 0.05$). No significant relationship was found between leptin and the general biochemical nutrition parameters or oxidative stress marker concentrations (MDA), neither before nor after six months of intervention (Table 3).

	Pre-educative intervention		**Post-educative intervention**	
	r	**p**	**r**	**p**
Glucose (mg/dl)	-0.35	0.859	0.194	0.321
Cholesterol (mg/dl)	0.184	0.348	0.075	0.706
HDL (mg/dl)	0.037	0.850	0.221	0.257
LDL (mg/dl)	-0.042	0.832	-0.285	0.141
Triglycerides (mg/dl)	0.235	0.228	0.240	0.219
Prealbumin (mg/dl)	0.017	0.930	0.263	0.176
Albumin (g/dl)	-0.216	0.269	-0.246	0.207
MDA (μM)	-0.081	0.683	0.158	0.440
BMI (kg/m^2)	0.582	0.001	0.516	0.006
% TBF	0.659	0.000	0.870	0.000
% TBW	-0.664	0.000	-0.871	0.000
TMB (Kcal)	0.122	0.536	0.390	0.055
TBF (kg)	0.555	0.002	0.788	0.000
VO_2max ($mlO_2/kg/min$)	-0.262	0.295	-0.498	0.042

Table 3: *Relationship and statistical significance between serum leptin levels and body composition, biochemical parameters and physical condition pre and post educative intervention in healthy women. (Pearson correlation was used).*

BMI: Body Mass Index; % TBW: Total Body Water Percent; % TBF: Total Body Fat Percent; TBF: Total Body Fat

Albumin concentrations showed a significant negatively correlation with triglycerides (r = -0.404; p = 0.028) and MDA (r = - 0.419; p = 0.033) after the subjects participation in the educative intervention. No significant correlation was found between albumin and the rest of biochemical nutrition parameters either between albumin and body composition parameters at any time of the study (Table 4).

	Pre-educative intervention		Post-educative intervention	
	r	p	r	p
Glucose(mg/dl)	0.227	0.246	-0.004	0.983
Cholesterol (mg/dl)	-0.207	0.291	-0.198	0.303
Triglycerides (mg/dl)	-0.84	0.671	-0.404*	0.028
HDL (mg/dl)	-0.240	0.218	-0.211	0.280
LDL (mg/dl)	-0.023	0.909	0.006	0.974
Prealbúmin (mg/dl)	0.142	0.471	0.136	0.489
Leptin (ng/ml)	-0.216	0.269	-0.246	0.207
MDA (μM)	-0.064	0.747	-0.419*	0.033
BMI (kg/m^2)	0.281	0.147	0.030	0.879
% TBF	-0.207	0.291	-0.295	0.127
% TBW	0.208	0.094	0.181	0.356
TMB (Kcal)	-0.195	0.319	-0.099	0.618
TBF (kg)	-0.198	0.313	-0.174	0.376

Table 4: *Relationship and statistical significance between serum albumin levels and body composition or biochemical parameters pre and post educative intervention in healthy women. (Pearson correlation was used).*

BMI: Body Mass Index; % TBW: Total Body Water Percent; % TBF: Total Body Fat Percent; TBF: Total Body Fat

We assessed the effects of a six months combined program of nutrition and exercise intervention in fasting serum leptin concentration and body composition in 28 healthy women. The study showed that fasting leptin concentrations were higher after intervention program. However, the leptin levels, pre and post intervention, were within normal limits as it has been reported in similar investigations in normal-weight women [12]. We do not have any clear interpretation of these results [13] found that in healthy women during the spontaneous menstrual cycles, peak leptin concentrations were achieved during the luteal phase and show greater than 1.5 fold variation with minimal changes in BMI. In this study has not taken into account the ovulation phase in which participants were, however all subjects reported having regular menstrual cycles. We could suppose that after program intervention some of the participants were in the luteal phase when leptin concentrations are highest. Which partly explain the difference in the concentration of leptin observed before and after program intervention.

The increase of leptin after intervention program was observed without accompanying modification in BMI or %TBF. Leptin is known that circulates primarily at levels proportional to the amount of adipose tissue and secondarily under the influence of other acute modulators including sympathetic nervous activity, caloric intake or physical activity [14]. The large fluctuations in serum leptin concentrations before and after intervention in the presence of relatively small changes in body weight suggest that leptin secretion could be regulated by other factors in addition to the size of the adipose-tissue depot. Previously, we had observed that six months program of intervention reduced the total energy ingested [15] and increased the time spent in moderate activity and reduced the time dedicated in high and low activities. Probably the participants were in negative energy balance which led them to reduce the time dedicated to intense activity in order to reduce energy expenditure and restore caloric balance and thus allowing serum leptin concentrations to increase again. The results regarding the effects of exercise on plasma leptin concentrations, independent of fat mass, are conflicting, there are authors that conclude

that physical activity in women is a determinant factor for circulating leptin levels after adjusting for confounding factors [16,17]; some others researches found that long-term diet and exercise interventions may have direct effects on the plasma leptin concentrations beyond the effect expected due to changes in fat mass [18] and some others argues that physical exercise don´t have direct effects on plasma leptin concentration, when body composition is unaltered [19].

We found that circulating leptin levels were positively associated with BMI and percentage of body fat (%TBF). The values were most closely correlated with the % TBF and the strength of this correlation ($r = 0.87$, $p < 0.001$) is much greater after program intervention. Our results agree with some previous studies [12,14,20].

The results of the present study indicate that cardiopulmonary fitness (VO_2max) is negatively associated with leptin after an intervention program. We are aware of only a few studies on the relationship between VO2max and leptin levels. A cross-sectional study conducted in European adolescents [21], in which the authors observed a negative association, and suggest that the association of both physical activity and fitness with leptin, in adolescents, are independent of each other [22] also pointed that in normal weight individuals peak VO_2 expressed as mlO_2/kg/min was more closely associated with leptin than body fat.

The mean of human serum albumin concentrations increased slightly after six months program intervention, although concentrations before and after the intervention were maintained in the normal range. Various factors may influence the Increased of blood albumin including the high protein diet intake observed after six months of intervention [15], increased time spent doing aerobic activity or increase albumin synthesis after upright exercise posture [23].

On the other hand, a negative correlation between albumin and MDA has been found [24] similar results in chronic renal failure patients. MDA level is commonly known as a marker of oxidative stress which can result either from low levels of antioxidants and/or from an increased production of reactive species [25]. In this study there was no statistically significant difference before and after intervention in the MDA parameter. However, the average concentration of MDA at the end of intervention increased by 21% compared to the initial concentration (3.87± 1.72 μM and 4.69 ± 1.54 μM) respectively. In this study participants increased the time spent doing moderate activity, which is considered an aerobic activity accompanied by increased free radical. Although, it is not clear whether the different modes of exercise can affect the oxidative stress response, some studies report that regular aerobic exercise reinforces antioxidant defense [26]. In this regard, it is important to point out that albumin is considered an important extracellular antioxidant molecule that can counteract free radical generation [27].

The present article is not without limitations. First, there was no control group without any intervention, therefore, we cannot exclude the possibility that might have similar effects on results after six months without educational intervention. Second, measurement error from the use of self-reported physical activity is likely to have occurred. Third, accurate methods to estimate ovulatory status, such as the presence of urinary LH peak, were not feasible in the present study. However, is an study with specific focus on the effect of an educative intervention program on physical activity and eating habits realized using valid international physical activity questionnaire, which provides the possibility for comparison of our results with other populations. Furthermore we used reliable methods to assessed biochemical health parameters when evaluating the association between them.

Conclusion

The results of this study demonstrate that six months physical activity and nutrition program modify the physical activity habits in adult healthy women and have also seen despite the fact that there were no changes in body weight or body composition increases in leptin and albumin levels. Our findings suggest that through this educative program can achieve changes in lifestyle and improve the antioxidant blood capacity in adult healthy women.

Acknowledgements

All authors approved the final version of the article. We thank Dr. Moro for the technical analysis. Also we thank the collaboration of the clinical analysis service of the Institutional Hospital of Soria and the center of high performance sports for having left their facilities.

Conflict of Interest

We declare that have no conflict of interest exists.

Bibliography

1. AESAN (Spanish Agency for Consumer Affairs, Food Safety and Nutrition of the Ministry of Health, Social Services and Equality). Spanish strategy for nutrition, physical activity and prevention of obesity (2005).
2. Pérez Gallardo L., *et al*. "Performance of nutritional education programmes to prevent obesity in children through a pilot study in Soria. [Utilidad de los programas de educación nutricional para prevenir la obesidad infantil a través de un estudio piloto en Soria]". *Nutricion Hospitalaria* 26.5 (2011): 1161-1167.
3. Montero A., *et al*. "Evaluation of dietary habits of a population of university students in relation with their nutritional knowledge. [Evaluación de los hábitos alimentarios de una población de estudiantes universitarios en relación con sus conocimientos nutricionales]". *Nutricion Hospitalaria* 21.4 (2006): 466-473.
4. Barrios-Vicedo R., *et al*. "A lower adherence to mediterranean diet is associated with a poorer self-rated health in university population. [Una menor adherencia a la dieta Mediterranea se asocia a una peor salud auto-percibida en población universitaria]". *Nutricion Hospitalaria* 31.2 (2014): 785-792.
5. Pérez-Gallardo L., *et al*. "Diet quality in college students with different academic profile". *Nutricion Hospitalaria* 31.5 (2015): 2230-2239.
6. Kelly C., *et al*. "Health behaviours". In Jo Inchley, Dorothy Currie, Taryn Young, Oddrun Samdal, Torbjørn Torsheim, Lise Augustson, Frida Mathison, Aixa Aleman-Diaz, Michal Molcho, Martin Weber and Vivian Barnekow (Ed.), Growing up unequal: Gender and socioeconomic differences in young people's health and well-being. Copenhagen, Denmark: WHO Regional Office for Europe (2016): 107-143.
7. Pérez-Gallardo L., *et al*. "Health promotion plan in college students". *Annals of Nutrition and Metabolism* 62.2 (2013): 49-50.
8. Kakkar R., *et al*. "Lipid peroxidation and activity of antioxidant enzymes in diabetic rats". *Molecular and Cellular Biochemistry* 151.2 (1995): 113-119.
9. WHO. "Global physical activity questionnaire (GPAQ)" (2011).
10. Sharkey BJ. "Physiology of fitness (Third edition)". Champaign Illinois: Human Kinetics Books (1990).
11. Bach A., *et al*. "The use of indexes evaluating the adherence to the mediterranean diet in epidemiological studies: A review". *Public Health Nutrition* 9.1A (2006): 132-146.
12. Ruhl CE and Everhart JE. "Leptin concentrations in the united states: Relations with demographic and anthropometric measures". *American Journal of Clinical Nutrition* 74.3 (2001): 295-301.
13. Hardie L., *et al*. "Circulating leptin in women: A longitudinal study in the menstrual cycle and during pregnancy". *Clinical Endocrinology* 47.1 (1997): 101-106.

14. Corr M., *et al*. "Circulating leptin concentrations do not distinguish menstrual status in exercising women". *Human Reproduction* 26.3 (2011): 685-694.

15. Navas J., *et al*. "Modifying dietary habits in university students after an educational intervention program". *Annals of Nutrition and Metabolism* 62 (2013): 46-46.

16. Esteghamati A., *et al*. "Physical activity is correlated with serum leptin independent of obesity results of the national surveillance of risk factors of noncommunicable diseases in Iran (SuRFNCD-2007) RID F-5769-2011". *Metabolism-Clinical and Experimental* 59.12 (2010): 1730-1735.

17. Miyatake N., *et al*. "Circulating leptin levels are associated with physical activity or physical fitness in Japanese". *Environmental Health and Preventive Medicine* 19.5 (2014): 362-366.

18. Reseland JE., *et al*. "Effect of long-term changes in diet and exercise on plasma leptin concentrations". *American Journal of Clinical Nutrition* 73.2 (2001): 240-245.

19. Dirlewanger M., *et al*. "Effect of moderate physical activity on plasma leptin concentration in humans". *European Journal of Applied Physiology* 79.4 (1999): 331-335.

20. Considine RV., *et al*. "Serum immunoreactive-leptin concentrations in normal-weight and obese humans". *New England Journal of Medicine* 334.5 (1996): 292-295.

21. Jiménez-Pavón D., *et al*. "Physical activity, fitness, and serum leptin concentrations in adolescents". *Journal of Pediatrics* 160.4 (2012): 598-603.e2.

22. Hosick PA., *et al*. "The relationships between leptin and measures of fitness and fatness are dependent upon obesity status in youth". *Pediatric Exercise Science* 22.2 (2010): 195-204.

23. Nagashima K., *et al*. "Intense exercise stimulates albumin synthesis in the upright posture". *Journal of Applied Physiology (1985)* 88.1 (2000): 41-46.

24. De Castro LL., *et al*. "Hypoalbuminemia and oxidative stress in patients on renal hemodialysis program". *Nutricion Hospitalaria* 30.4 (2014): 952-959.

25. Halliwell B and Whiteman M. "Measuring reactive species and oxidative damage in vivo and in cell culture: How should you do it and what do the results mean?" *British Journal of Pharmacology* 142.2 (2004): 231-255.

26. Bianchi VE and Ribisl PM. "Reactive oxygen species response to exercise training and weight loss in sedentary overweight and obese female adults". *Journal of Cardiopulmonary Rehabilitation and Prevention* 35.4 (2015): 263-267.

27. Roche M., *et al*. "The antioxidant properties of serum albumin". *FEBS Letters* 582.13 (2008): 1783-1787.

Volume 11 Issue 1 September 2017