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Research Article | Araştırma Makalesi

THE RELATIONSHIP BETWEEN METAL ELEMENT CONTENT OF DRINKING WATER AND BODY COMPOSITIONS OF PEOPLE IN BATMAN (TÜRKİYE)

İÇME SUYU METAL ELEMENT İÇERİĞİ İLE BATMAN'DA (TÜRKİYE) YAŞAYAN BİREYLERİN VÜCUT KOMPOZİSYONLARI ARASINDAKİ İLİŞKİ

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ABSTRACT

Objective: Previous studies showed that the levels of micro elements may have a contribution to weight loss. Nevertheless, it is not possible to find many comprehensive studies analysing the potential relation between micro elements found in water used for drinking purposes and body composition. The present study aims to assess the relation between micro element levels of drinking water and body composition in normal, fat and obese subjects.

Methods: The study consisted of 345 female participants, who were divided into 3 groups of 115, according to body mass index (BMI) for leanness, overweight, and obesity; and who applied to Diet Polyclinic of State Hospital. Iron (Fe), copper (Cu), cobalt (Co), zinc (Zn), manganese (Mn), molybdenum (Mo), selenium (Se), chromium (Cr) and bromine (Br) levels in water samples were analyzed with inductively coupled plasma mass spectrometry (ICP-MS); and body composition measurements were made by bioelectrical impedance analysis.

Results: The obesity percentage in females demonstrated statistically positive correlations with Co and Se in water for drinking. Also, it was found that the BMI values of females significantly positively correlated with Cr.

Conclusions: It may be suggested that women who consume drinking water containing high levels of Co, Se and Cr might be at a greater risk of developing obesity. On the other hand, the lack of knowledge about influence of levels of micro elements in drinking water on blood parameters associated with obesity or tissue remains unclear and deserves more investigation.

Keywords: Obesity, body mass index, cobalt, selenium, chromium

ÖZ

Amaç: Önceki çalışmalar mikro element düzeylerinin kilo kaybına katkı sağlayabileceğini göstermiştir. Ancak, içme suyu mikro element düzeyleri ile vücut kompozisyonları arasındaki potansiyel ilişkiyi analiz eden kapsamlı bir çalışma bulunmamaktadır. Bu çalışmada, içme suyunda bulunan mikro element düzeyleri ile normal, kilolu ve obez kişilere ait vücut kompozisyonları arasındaki ilişkinin incelenmesi amaçlanmıştır.

Yöntem: Bu çalışmaya, Devlet Hastanesi Diyet Polikliniğine başvuran ve vücut kütle indeksi (VKİ) referans değerlerine göre normal, kilolu ve obez olarak üç gruba ayrılan, 55-70 yaş aralığında toplam 345 kadın dâhil edilmiştir. Su örneklerindeki demir (Fe), bakır (Cu), kobalt (Co), çinko (Zn), manganez (Mn), molibden (Mo), selenyum (Se), krom (Cr) ve brom (Br) düzeyleri indüktif eşleşmiş plazma-kütle spektrometresi (ICP-MS) ile analiz edildi ve vücut kompozisyonu ölçümleri biyoelektrik impedans analizi ile yapıldı.

Bulgular: Kadınlara ait obezite yüzdesi, içme suyu Co ve Se düzeyleri ile pozitif korelasyon gösterdi. Ayrıca, kadınlara ait VKİ değerlerinin içme suyu Cr düzeyleri ile pozitif korele olduğu bulundu.

Sonuç: Co, Se ve Cr yüksek düzeyde içeren içme suyu tüketen kadınların obezitenin gelişmesi açısından daha büyük bir risk altında olabileceği öne sürülebilir. Diğer taraftan, içme suyu mikro element seviyelerinin obezite ile ilişkili kan parametrelerine etkisi belirsizdir ve daha fazla araştırmayı hak etmektedir.

Anahtar Kelimeler: Obezite, beden kitle indeksi, kobalt, selenyum, krom

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Introduction

Excess body weight is accepted as a crucial risk factor aiding the overall burden of disease worldwide. Obesity is a process where fat is piled up in the body abnormally, generally 20% or more than the body weight of an individual.[1] It is found out that a great number of mortalities stem from various diseases related to obesity, for instance; diabetes, chronic kidney disease, gastro intestinal disease, and cardiovascular disease. Sustaining weight loss is frequently proved to be challenging or unsuccessful. Therefore, prevention and treatment of obesity are relevant to health promotion.[2]

It has been reported that deficient intake of trace elements might be relevant to obesity development.[3] In contrast, in several examinations, obese and overweight children seem to be at a greater risk of advancing an instability [chiefly deficiency] of trace elements compared to those with a normal body weight; furthermore, resembling results have been found in adults.[4-6] Some studies state that raised body mass index (BMI) is related to increased levels of serum ferritin and low transferrin saturation.[7,8] A number of various epidemiological investigations have witnessed negative correlations between cellular Zn levels and the prevalence of obesity; while others have pronounced that serum Cu levels share common properties in obesity. In obese patients, abnormality in cellular Fe, Se, Cr and other element concentrations has also been seen.[9,10]

Drinking water embodies a number of microelements, and their types and concentrations depend on the geochemical properties of the earth layers. The pollution of waters by industrial
plants has, with great probability, had an impact on the content of microelements.[11] It was suggested that element content of water used for drinking could offer negative health influences; and the carbohydrate metabolism and fats in human organisms are linked to several microelements.[12,13] Yet, the studies conducted on elements in water have usually concentrated on whether or not minerals taken with water contribute to the suggested Daily intake amount of elements. Besides, the emphasis has been put on how elements that exist in hard water and are generally found missing in the general population (calcium and magnesium, especially) decrease the risk of disease, mainly cardiovascular diseases, and thus contribute to health. In 2008, the World Health Organization (WHO) published a guide on the quality of drinking water, and, due to their effect on human health, water hardness and nutrients in drinking water.[14,15]

Considering the literature knowledge we have, the former studies examining the relationship of obesity or fat tissues and micro elements have been conducted mainly to examine the influence of their benefits as a supplement on obesity-related molecule levels, fat tissues and weight.[5-9,13] However, elements scarcely go into the body in pure form or on their own, which means that to correctly calculate the influence of trace elements on adipose tissue or the existence of fat tissue, the influence of water—the most significant reservoir of minerals—on the occurrence of fat tissues needs to be studied. Thereby, the objectives of this study were to assess the effect of Zn, Mn, Se, Cu, Co, Fe, Cr, Br and Mo levels in public drinking water wells on the body composition of people who consume this water in Batman, where most residents opt for tap water drawn from groundwater sources for economic and cultural reasons.

Methods

Study Design and Population

The subjects of the study consisted of 345 women, divided into 3 groups having 115 applicants each, who appeared at Batman Regional State Hospital, Diet Polyclinic from December 2015 till the end of February 2016. We received ethics committee approval from ethics committee of Batman University (06.07.2015/2015/1-1).

All subjects were told about the aim of the study. In addition, each was required to hand in a written informed consent document before they were accepted into the study, complying with Helsinki Declaration (World Medical Association). All subjects participating in the study were female, from the age of 18 to 50, and were living in the province of Batman. The subjects were neither have any systemic diseases such as coronary disease, diabetes, cancer, or other fatal diseases except for obesity nor taking any regular medication. Some subjects were let out of the study since they drank drinking water which is bottled or water from different water resources.

The subjects were weighed while they wore thin clothing without shoes. Before each measurement, a pre-calibrated electronic balance was utilized to find out the weight of each subject within 0.1 kg. The median (25^{th}-75^{th} percentile) weight of obese, overweight and control group subjects was 86.1 (79.8-92.1), 73.3 (68.5-78.3) and 61.9 (58.2-67.7) kg, respectively.

A stadiometer mounted on the wall was employed to compute how tall they are to the closest 0.1 cm taking Frankfort horizontal plane into consideration. The medians (25^{th}-75^{th} percentile) height of obese, overweight and control group subjects were 162.0 (157.0-168.0), 163.0 (158.0-167.0) and 164.0 (159.0-170.0) cm, respectively.

Body mass index (BMI) values were calculated using weight and height (kg/m^2). Three groups were made up depending on the clinical cut-off values used by the general health community. Those who had a BMI between 18.50 and 24.99 kg/m^2 were grouped as lean, the control group; those who had a BMI between 25.00 and 29.99 kg/m^2 were placed in the overweight group; and those who had a BMI over 30.00 kg/m^2 were placed in the obese group. The median (25^{th} -75^{th} percentile) BMI of obese, overweight and control subjects was 32.4 (31.5-33.6), 27.8 (26.35-28.90) and 23.2 (22.0-24.2) kg/m^2, respectively.

We got in touch with the municipality water authorities in connection with 16 wells in Batman to attain knowledge and consent for studying the samples of water obtained from the reservoirs of water providing the water requirements of the city and settlements nearby. The participants were matched with water sources according to their residence addresses so as to find the connection between the drinking water element levels and their body compositions. In accordance with international standards, samples of water were picked up from the wells providing water to every district of Batman.

Bioelectrical Impedance

In State Hospital Diet Polyclinic, measurements of the participants' body composition were fulfilled with the help of a body composition analyser (Tanita BC 418 MA). Before measurements were carried out for the terms of bioelectrical impedance analysis, knowledge about the gender, age and height of subjects' were loaded to the device. Once the body compositions were assessed with the help of balance, the subjects were wanted to hold their hands alongside their body during the measurement of impedance (Hand to foot BIA). A visceral index (from 1 to 55) was also suggested to predict visceral fat.[16] To figure out statistical process, total fat mass, lean body mass, and the visceral adiposity index were taken down by the investigator on an excel sheet. This method works with body water conductivity principle changing in different parts of body. While it passes through the body's water pool, BIA calculates the impedance of an applied small electric current.[17]

Water Analysis and Analytical Procedures

The water samples with the number 1, 2, 3, 4, 5, 6, 7, 8, 9, 10, 11, 12, 13, 14, 15, or 16 were obtained from wells in the Selmo Formation, and the rest was picked up from wells located in the old alluvium area. Paying no attention to the sources, each sample was obtained after the pump was run for nearly 1 h taking out water from the well; afterwards it was filled into sterilised containers into which 10% hydrochloric acid was added in order to acquire pH levels of <2. On 16 samples, we carried out laboratory analyses according to the standard methods of the American Public Health Association (APHA). They were then preserved in a portable fridge at +4 °C. Additionally, we filled the water samples in 1 L sterilized plastic bottles double capped. Then, anion and cation analyses of the water were conducted in conformity with the standard methods of American Public Health Association rules.[18] Nitrate analyses and phenol quantities were carried out in Diyarbakir Metropolitan Municipality laboratory. With the 2C Full Suite (ACME Analytical Laboratories, Vancouver, Canada), the samples were analysed chemically employing ICP-MS in order to specify all significant and heavy metal contents.[19]

Statistical Analysis

Using statistics packages with SPSS software version 15.0 and Sigma Stat 3.5, statistical analyses were conducted. The data distribution normality was assessed via Kolmogorov- Smirnov test. A one-way ANOVA test was employed so as to study the average differences between groups to fit the normal distribution; nevertheless, the Kruskal Wallis test did not match within the normal distribution. The changes in the distribution of categorical variables were assessed by using the Chi-square test. The Pearson and Spearman Correlation test was carried out so as to look into the connection between element levels and body composition. Categorical variables were expressed with numerical digits, and continuous variables were stated as median (25^{th}-75^{th} percentile) or mean±SD where possible.

Results

Table 1 shows the demographic characteristics and body compositions of the participants. The median (25^{th} -75^{th} percentile) age of obese, overweight, and control group subjects was 31.0 (26.0-37.0), 31.0 (26.0-37.0), and 31.0 (26.0-35.0) year, respectively while the mean age was approximately 30 years in the patient groups, in the control group it was about 28 years. The median (25^{th} -75^{th} percentile) BMR of obese, overweight, and control group subjects was 1576.0 (1490.5-1701.0), 1487.0 (1396.0-1576.5), and 1400.0 (1326.2-1515.0) kcal, respectively. As was anticipated, abdominal adiposity, fat mass and fat percentage coefficient were reported to be peak in the obese subjects. It was determined that there were important statistical variations among all groups with the exception of height and age ($p<0.001$; Table 1).

Table 2 displays the distribution of the water wells from which patients consumed water with reference to their addresses. The average mineral levels of the wells were determined as 0.067± 0.129 mg/L for Fe, 0.003±0.002 mg/L for Cu, 0.0009±0.003 mg/L for Co, 0.034±0.905 mg/L for Zn, 0.071±0.2690 mg/L for Mn, 0.001±0.0009 mg/L for Mo, 0.001±0.001 mg/L for Se, 0.003±0.002 mg/L for Cr and 0.078±0.049 mg/L for Br. When reviewing the distribution of patients to wells according to their address in Batman, it was observed that the individuals in control group mainly consumed drinking and domestic water from wells numbered as 1, 2, 3, 4, 5 and 7 in Batman. In contrast, it was observed that overweight individuals consumed drinking and domestic water from wells numbered as 1, 3, 4, 11, 12 and 14 in Batman. Finally, it was observed that obese and overweight individuals used up drinking and domestic water from wells numbered as 7, 8, 9, 11, 12 and 14 in Batman (Table 2).

Table 3 shows the correlation values between elements in drinking water and the anthropometric measurements and BIA computations of individuals. The BMI values demonstrated statistically significant moderate positive correlation with Cr in all subjects ($p<0.05$). The percentage of obesity showed statistically significant moderate positive correlation with Co and Se in drinking water in all subjects ($p<0.01$). In addition, statistically insignificant mild correlations were found between individuals' BIA values and other elements present in the drinking water (Table 3).

Table 1. Demographic characteristics and body compositions of the participants

Parameters	Control (n=115)	Patients		Comparisons		
		Overweight (n=115)	Obese (n=115)	a	b	c
Age (year)	31.0(26.0-35.0)	31.0(26.0-37.0)	31.0(26.0-37.0)	p=0.375	p=0.123	p=0.473
Height (cm)	164.0(159.0-170.0)	163.0(158.0-167.0)	162.0(157.0-168.0)	p=0.056	p=0.952	p=0.600
Body weight (kg)	61.9(58.2-67.7)	73.3(68.5-78.3)	86.1(79.8-92.1)	p<0.001	p<0.001	p<0.001
BMI (kg/m^2)	23.2(22.0-24.2)	27.8(26.35-28.90)	32.4(31.5-33.6)	p<0.001	p<0.001	p<0.001
FFM (kg)	45.3(43.0-50.5)	47.80(45.50-51.25)	50.8(48.2-54.6)	p<0.001	p<0.001	p<0.001
Bone mass	43.3(41.0-48.2)	2.40(2.30-2.60)	2.60(2.50-2.80)	p<0.001	p<0.001	p<0.001
Obesity %	2.58±7.29	21.7±5.38	41.0±8.4	P<0.001	P<0.001	P<0.001
FM (kg)	24.4±6.9	32.2±5.6	38.5±5.2	p<0.001	p<0.001	p<0.001
Fat %	25.6(19.8-28.8)	33.10(30.25-35.75)	40.1(37.3-41.7)	p<0.001	p<0.001	p<0.001
Amount of minerals	3.22(2.94-3.59)	3.48(3.24-3.70)	3.66(3.40-3.94)	P<0.001	P<0.001	P<0.001
Amount of protein	9.09(8.44-10.11)	9.46(8.88-10.23)	9.96(9.41-10.82)	P=0.013	P<0.001	P<0.001
Abdominal adiposity	3.00(1.25-4.00)	5.0(4.0-6.0)	8.00(7.00-9.00)	p<0.001	p<0.001	p<0.001
Body density	1.038(1.031-1.052)	1.021(1.015-1.028)	1.005(1.002-1.012)	p<0.001	p<0.001	p<0.001
BMR (kcal)	1400.0(1326.2-1515.0)	1487.0(1396.0-1576.5)	1576.0(1490.5-1701.0)	p<0.001	p<0.001	p<0.001
Calorie of activity	195.25(185.50-214.75)	175.7(166.0-184.2)	156.2(146.50-156.25)	P<0.001	P<0.001	P<0.001
Total activity	1602.9(1507.3-1721.9)	1654.0(1567.3-1748.1)	1735.2(1648.8-1855.1)	P=0.008	P<0.001	P<0.001
TBW (kg)	33.3(31.6-36.9)	35.0(33.3-37.5)	37.2(35.3-39.9)	p<0.001	p<0.001	p<0.001
TBW %	54.25(52.07-58.64)	48.7(47.0-51.1)	43.9(42.6-45.9)	P<0.001	P<0.001	P<0.001
Bone mineral weight (kg)	2.25(2.10-2.50)	2.40(2.30-2.60)	2.60(2.60-2.80)	P<0.001	P<0.001	P<0.001
Skeletal muscle mass(kg)	25.86(24.51-28.64)	27.34(25.75-29.06)	28.7(27.2-30.9)	P=0.002	P<0.001	P<0.001
Intracellular fluid (kg)	19.30(18.29-21.37)	20.40(19.22-21.69)	21.4(20.25-23.10)	P=0.002	p<0.001	p<0.001
Extracellular fluid (kg)	14.14(13.14-15.63)	14.94(14.07-15.88)	15.70(14.94-16.90)	P=0.002	p<0.001	p<0.001

Data are expressed as mean±SD or median (25th-75th percentile) for continuous variables. Comparisons of study groups; a: Obese-Overweight, b: Overweight-Control, c: Obese-Control. BMI: Body Mass Index, BMR: Basal metabolic rate, FFM: Fat free mass, FM: Fatt mass, TBW: Total Body Water, TBW %: Total Body Water Percentage.

Table 2. The drinking water well distributions according to address of populations in Batman

Wells	Obese (n=115)	Overweight (n=115)	Controls (n=115)	Total participants
1. Well	2	10	26	38
2. Well	1	7	16	24
3. Well	7	19	23	49
4. Well	2	15	13	30
5. Well	5	-	9	14
6. Well	-	1	4	5
7. Well	9	4	1	14
8. Well	8	9	7	24
9. Well	12	3	2	17
10. Well	6	3	3	12
11. Well	23	15	1	39
12. Well	18	9	2	29
13. Well	1	2	2	5
14. Well	9	9	3	21
15. Well	6	2	2	10
16. Well	6	7	1	14
Total participants	115	115	115	345

Table 3. The correlation values between the BIA values of individuals and elements in drinking water

Variables/Minerals	Fe	Cu	Co	Zn	Mn	Mo	Se	Cr	Br
Height	-0.209	-0.156	-0.076	-0.072	-0.086	0.034	-0.086	-0.259	-0.073
Body weight	0.005	-0.021	0.065	0.094	-0.047	0.021	0.096	0.045	-0.137
BMI	0.316	0.144	0.238	0.278	0.101	-0.018	0.204	**0.328***	-0.069
FFM	0.042	-0.055	0.066	-0.060	-0.082	-0.063	0.140	-0.018	-0.097
Bone mass	0.024	-0.045	0.049	0.037	-0.019	-0.075	0.066	-0.001	-0.127
Obesity %	0.241	0.296	**0.386***	0.115	0.277	-0.080	**0.446****	0.282	0.223
FM	0.038	-0.019	0.165	0.234	0.084	0.091	0.017	0.054	-0.054
Fat %	0.048	-0.063	0.099	0.143	0.081	0.106	-0.118	0.012	-0.109
Amount of minerals	0.139	0.102	-0.032	-0.064	-0.144	-0.059	0.092	-0.095	-0.004
Amount of protein	-0.122	-0.016	-0.044	0.138	-0.032	0.196	-0.282	-0.008	-0.289
Abdominal adiposity	0.178	-0.233	-0.101	-0.143	-0.079	-0.122	0.122	0.121	-0.107
Body density	-0.045	0.072	0.017	-0.060	-0.129	-0.151	0.210	-0.084	0.061
BMR	0.070	0.017	0.019	-0.126	-0.069	-0.184	0.232	-0.024	0.215
Activity calorie	-0.218	0.253	0.112	-0.081	0.073	-0.129	0.292	0.121	0.136
Total activity calorie	-0.060	0.161	0.155	-0.004	0.027	-0.121	**0.337***	0.062	0.206
TBW	-0.044	0.136	0.002	0.056	-0.035	-0.180	0.222	0.055	-0.049
TBW %	-0.034	0.064	-0.089	-0.143	-0.069	-0.113	0.115	-0.013	0.106
Bone mineral weight	0.061	-0.022	0.090	0.045	0.045	-0.070	0.144	-0.001	-0.036
Skeletal muscle	0.004	0.072	0.054	-0.007	-0.009	-0.124	0.219	-0.024	-0.012
Extracellular fluid	0.011	0.071	0.064	-0.002	0.000	-0.124	0.220	-0.019	-0.010
Intracellular fluid	0.004	0.072	0.054	-0.007	-0.009	-0.124	0.219	-0.024	-0.012

BMI: Body Mass Index, BMR: Basal metabolic rate, FFM: Fat free mass, LM: Lean mass, FM: Fatt mass, TBW: Total Body Water, TBW %: Total Body Water Percentage. *P <0.05 **P < 0.01

Discussion

Our outcomes provide evidence of obesity percentage showing moderate positive correlation with Co in drinking water. Despite the fact that the direct action of Co has not been displayed in adipose tissue, one of the studies demonstrates that cobalt chloride treatment repairs high-fat diet-induced hypertrophic adipocytes to their normal size.[20,21] It was also contemplated that Co ions have a preventive role in dysregulated lipid metabolism. Additionally, Co ions have useful influences on lipid metabolism; for instance, a rise in blood levels of HDL-cholesterol level and a drop in LDL-cholesterol, free fatty acid, and triglyceride in the mice fed with a high-fat diet.[21] Blood glucose levels and visceral fat content reduces, while adiponectin level increases with the help of Co protoporphyrin supplementation in obese/diabetic animal models.[20] It was also suggested that although the mechanism of the increased adiponectin levels in Co-treated mice is still unknown, the alterations could be due to the indirect influences of the metals. As restricted knowledge exists about the influence of Co in drinking water on weight loss or gain, further investigations ought to search for determining the link between obesity and Co levels found in drinking water.

In our study, we also found that the Se content in drinking water showed moderate positive correlations with the obesity percentage. It has been found that high Se levels give rise to increased adiposity and may also boost the risk of diseases such as hypertension, diabetes, and dyslipidaemia.[22] Se is necessary for glutathione peroxidase, which has a significant role in the organism protection against oxidative stress. Oxidative stress leads to increasing adiposity in the organism, and the requirement of antioxidants such as Se increases.[23] High-Se diets might trigger the discharge of glucagon, promoting hyperglycaemia, giving rise to insulin resistance and obesity.[24] Adverse health effects of Se are known and might occur owing to supra-nutritional Se intake even below the levels necessary for intoxication.[25] Considering these aspects, it might be speculated that Se content of drinking water might possess a number of significant regulatory influences on percentage of obesity.

Perhaps, another most important finding of our investigation was that the demonstration of the substantial influence of element levels in drinking water on the development of fat tissues, especially in the Cr levels of drinking water, which correlates positively with BMI.

Elements and elemental species are used in various biological functions, such as energy homeostasis in glucose metabolisms and lipid.[11,13,21] The magnitude and function of adipocytes was found to possibly be influenced by elements. The literature has had a great many studies about the influences of necessary and unnecessary trace elements on adipocytes.[26] However, there are contradictory findings on this issue. For example, it is shown that Cr supplementation lessens body weight, arranges hunger, and also reduces body fat. In parallel with influencing insulin-mediated pathways, Cr decreases inflammation which is known to play an important role in insulin resistance and obesity advancement .[27,28] In contrast, some studies reported that regular Cr supplementation may have anabolic effects on individuals who do regular physical exercise. Adipose tissue and Cr have also been shown to significantly correlate with serum adiponectin, leptin, and insulin levels values.[29]

Consistent with regular Cr supplementation study,[27-29] in our study results showed positive correlations between the Cr content of drinking water and BMI values. This knowledge may be interpreted as the Cr content of drinking water having possible positive effects on BMI values of subjects who consume this water. In addition, metal element levels measured in drinking water are below toxic levels.[30,31]

Study limitations: The population world of the study was small and it was conducted with a single gender. Blood micro element levels were absent. So, influences of micro element levels in drinking water on blood parameters were not assessed.

In conclusion, the findings of our study showed associations between obesity-related parameters and Co, Se and Cr levels of drinking water. It may be suggested that women who consume drinking water that contains high levels of Co, Se and Cr may be at a greater risk of developing obesity. More comprehensive research regarding Co, Se and Cr and their relationships to weight status are suggested.

Compliance with Ethical Standards

The study protocol was approved by the Batman University Ethics Committee (06.07.2015/2015/1-1).

Conflict of Interest

The author declares no conflicts of interest.

Author Contribution

This study was carried out in collaboration among all authors. İÇ: Designed the study; İÇ, SA, KT: Obtained data; İÇ, SA, MTN, KT: Analyzed and interpreted the data; İÇ, SA, NK: Develop the draft manuscript; MTN: Performed the statistical analysis. All authors read and approve the final manuscript.

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