ORIGINAL ARTICLE ÖZGÜN ARAŞTIRMA

The Correlation Between Adherence to Mediterranean Diet and HOMA-IR in Children and Adolescents

Çocuk ve Adölesanlarda Akdeniz Diyetine Uyum ile HOMA-IR Arasındaki İlişki

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Keywords
Dietary habits, Mediterranean diet, insulin resistance, pediatric obesity

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Abstract

Introduction: Nowadays, children's exposure to the obesogenic environment can lead to obesity and insulin resistance in the early period. However, it is suggested that the risk of developing health problems such as obesity, insulin resistance, and type 2 diabetes may decrease with increasing adherence to the Mediterranean diet. This study was aimed to evaluate the relationship between insulin resistance and the Mediterranean Diet Quality Index (KIDMED) in children and adolescents.
Materials and Methods: A total of 174 children and adolescents (35.6% boys, 64.4% girls) with a mean age of 11.6±3.17 years were included in the study. The data were collected by a questionnaire containing individuals' socio-demographic information, anthropometric measurements, KIDMED, and 24-hour food consumption records. According to World Health Organization criteria, the body mass index z-scores of the participants were evaluated, and participants were divided into three groups as normal weight (29.9%), overweight (23.6%), and obesity (46.5%).
Results: This study's findings were: mean KIDMED score was 5.7±2.62 points in the normal weight group, 5.4±2.39 points in the overweight group, and 4.6±2.70 points in the obesity group which was significantly lower than the normal weight group ($p<0.05$). Additionally, the mean KIDMED score of children with insulin resistance (4.3±2.72) was lower than those without insulin resistance (5.4±2.57) ($p<0.05$). A negative, moderate, and significant correlation was found between the participants' KIDMED scores and the homeostatic model assessment of insulin resistance value ($r=-0.338$, $p<0.001$).
Conclusion: The low level of adherence to the Mediterranean diet was associated with the risk of developing insulin resistance.

Öz

Giriş: Günümüzde, çocukların obezojenik ortama maruz kalması erken dönemde obezite ve insülin direncine yol açabilmektedir. Ancak, Akdeniz diyetine uyumun artmasıyla obezite, insülin direnci ve tip 2 diyabet gibi sağlık sorunlarına yakalanma riskinin azalabileceği ileri sürülmektedir. Bu çalışma, çocuk ve adolesanlarda insülin direnci ile Akdeniz Diyet Kalite İndeksi (KIDMED) arasındaki ilişkiyi değerlendirmeyi amaçlanmıştır.
Gereç ve Yöntem: Yaş ortalamaları 11,6±3,17 yıl olan 174 çocuk ve adolesan (%35,6 erkek, %64,4 kız) çalışmaya dahil edilmiştir. Veriler, bireylerin sosyo-demografik bilgilerini, antropometrik ölçümlerini, KIDMED ve 24 saatlik besin tüketim kayıtlarını içeren anket formu ile toplanmıştır. Dünya Sağlık Örgütü kriterlerine göre katılımcıların beden kütle indeksi z-skorları değerlendirilmiş ve

katılımcılar normal ağırlıklı (%29,9), fazla kilolu (%23,6) ve obez (%46,5) olarak üç gruba ayrılmıştır.
Bulgular: Normal ağırlıklı bireylerin KIDMED puanı ortalaması 5,7±2,62 puan, fazla kilolular 5,4±2,39 puan, obezite grubunda 4,6±2,70 puandır ve obezite grubunun normal ağırlıklılara kıyasla KIDMED puan ortalaması anlamlı olarak daha düşüktür (p<0,05). Ayrıca, insülin direnci olanların ortalama KIDMED puanı (4,3±2,72), insülin direnci olmayanlardan (5,4±2,57) daha düşüktür (p<0,05). Katılımcıların KIDMED puanları ile insülin direncinin homeostatik modeli değerlendirmesi değeri arasında negatif yönlü, orta düzeyde, anlamlı ilişki saptanmıştır (r:-0,338; p<0,001).
Sonuç: Akdeniz diyetine uyum düzeyinin düşük olması insülin direnci gelişme riski ile ilişkilidir.

Introduction

The Mediterranean diet, a diet model, based on the consumption of traditional foods and beverages of the countries surrounding the Mediterranean, plays a significant role in protecting and improving health (1). The use of olive oil as the primary source of fat, increasing the consumption of whole grains, nuts, legumes, vegetables, and fruits are among the Mediterranean diet principles. Also, it is recommended to consume moderate amounts of dairy products, fish, chicken and eggs, and low amounts of red meat and meat products (1-3).

The studies show that the Mediterranean diet with healthy nutritional recommendations and its components have beneficial effects on weight loss, reducing the risk of developing insulin resistance, cardiovascular diseases, and type 2 diabetes (4-6). It has been reported that the prevalence of obesity is low in children who follow the Mediterranean diet (7). Additionally, a study in children with obesity was found that those with poor compliance with the Mediterranean diet had higher fasting insulin levels and homeostatic model assessment of insulin resistance (HOMA-IR) values, which is an indicator of insulin resistance (8). In children and adolescents with obesity, compliance with the Mediterranean diet for 16 weeks has been found to reduce body mass index (BMI), fat mass, fasting blood glucose, triglyceride, total cholesterol, and low-density lipoprotein cholesterol levels (9). A study in children with high cholesterol levels was found that the Mediterranean diet intervention for 12 months improved lipid profile and decreased glucose levels and HOMA-IR values (10). The Mediterranean diet, rich in vitamins, minerals, polyphenols, and unsaturated fatty acids [mainly monounsaturated fatty acids (MUFAs)], results in anti-inflammatory, antioxidant, antimutagenic effects; therefore, it has a protective effect against many diseases. In particular, antioxidant compounds reduce oxidative stress associated with pancreatic β-cell dysfunction and insulin resistance. These findings can explain the low risk of developing type 2 diabetes with the Mediterranean diet (11).

Today, obesity, which develops from unhealthy eating habits and low physical activity levels, has become a significant public health problem. Besides, obesity causes behavioral and emotional difficulties in children; moreover, it is also associated with insulin resistance which is related to developing complex processes such as type 2 diabetes, hypertension, dyslipidemia, and inflammation. For these reasons, it is crucial to identify the factors that play a role in preventing the increase in childhood obesity and insulin resistance. In this context, this study was aimed to evaluate the relationship between insulin resistance and the Mediterranean Diet Quality Index (KIDMED), which is an indicator of compliance with the Mediterranean diet in children and adolescents. Also, it was aimed to determine the relationship between the nutrient intake in the daily diets of children and HOMA-IR values.

Materials and Methods

Participants

A total of 174 children and adolescents [62 boys (35.6%), 112 girls (64.4%)] aged 6-17 who applied to the pediatric endocrine department participated in the study. The study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects/patients were approved by the Ethics Committee. Before the data collection, the participants and their parents were informed about the research. Informed consent was obtained from the children and their parents. The study did not include those who regularly used any nutritional supplements, non-volunteers, with thinness, growth retardation, and chronic diseases.

Design

Data collection took place from August 2019 to December 2019. The data were collected by the researchers using a face-to-face interview technique with the questionnaire prepared for this cross-sectional study.

Questionnaire

The questionnaire consists of individuals' socio-demographic information, anthropometric measurements, KIDMED, and 24-hour food consumption records.

The researchers took the height and body weight measurements of the participants by the procedure, and BMI was calculated using these values. BMI z-scores of the participants were evaluated using the World Health Organization AnthroPlus program. Accordingly, those with BMI z-score, -1SD≥BMI≤+1SD were categorized in the normal-weighted group, those with +1SD>BMI≤2SD were categorized in the overweight group, and those with BMI>2SD were categorized in the obesity group (12).

The biochemical parameters [fasting insulin (uIU/mL), fasting glucose (mg/dL)] were obtained from the patient folders. The insulin resistance status of the participants was determined by pediatric endocrinologists using the formula HOMA-IR=[fasting insulin (uIU/mL) × fasting glucose (mg/dL)/405] (13). HOMA-IR reference values for Turkish children and adolescents (5-18 years) were used: insulin resistance was defined as in the prepubertal period, ≥2.67 in boys and ≥2.22 in girls; and in the pubertal period, ≥5.22 in boys and ≥3.82 in girls (14).

KIDMED, which was developed by Serra-Majem et al. (15) (2004), consists of 16 questions and determines some principles related to the Mediterranean diet and evaluates the nutritional habits of children. Twelve questions of this index are positive, and four questions are negative. Those who answer "Yes" to positive questions get +1 points, and those who answer "Yes" to negative questions score -1. As a result of the sum of these points, scores ranging from 0-12 are obtained. A total score of ≥8 is considered to be the optimal Mediterranean diet adherence (good), between 4-7 scores are regarded as an improvement needs to Mediterranean diet adherence (average), and ≤3 points are also considered as poor Mediterranean diet adherence (low).

The participants' food consumption data were obtained with a 24-hour recall method using and "Photo Catalog of Foods and Nutrients: Measures and Amounts" (16) was used to determine portion sizes of consumed foods. The average intake of energy and nutrients in the daily diet was determined by the "Nutritional Information Systems Package Program" (17).

Statistical Analysis

SPSS 26.0 (statistics program for social sciences) package program for Windows was used to analyze the data. Descriptive statistics were shown as mean (M) ± standard deviation (SD) and minimum-maximum values. Nominal variables were expressed as the number of cases (n) and percentage (%). The chi-square test was used to evaluate the relationship between categorical variables. The chi-square test was used to assess the comparison of qualitative data. Whether the quantitative data were normally distributed was examined using the Kolmogorov-Smirnov test or the Shapiro-Wilk test. The independent samples t-test was used to compare the means of two independent groups. When the number of groups was more than two, the one-way analysis of variance was used to evaluate whether there was a statistically significant difference between the quantitative variables. The Pearson correlation test was used to assess the correlation between two variables under the condition that normal distribution assumptions. However, when at least one of the variables did not meet the normal distribution conditions, the Spearman correlation test was used. Statistical significance was accepted at $p<0.05$.

Results

A total of 174 children and adolescents, 62 boys (35.6%) and 112 girls (64.4%), between the ages of 6-17 years, with a mean age of 11.6±3.17 years, participated in the study. Participants were divided into three groups according to their BMI z-scores; 29.9% were categorized in the normal weight group, 23.6% were categorized in the overweight group, and 46.5% were categorized in the obesity group.

No statistically significant difference was found between the groups according to the KIDMED classification ($p>0.05$). The mean KIDMED score was found 5.7±2.62 points in the normal weight group, 5.4±2.39 points in the overweight group, 4.6±2.70 points in the obesity group. The obesity group had a significantly lower mean KIDMED score compared to the normal weight group ($p<0.05$). However, KIDMED score of BMI groups in the ANOVA test, partial eta squared was found as 0.036. A significant difference with a small effect size was found in the KIDMED score according to BMI groups (Table 1).

No statistically significant difference was found between children and adolescents with and without insulin resistance according to KIDMED classification (p>0.05). However, the mean KIDMED score of those with insulin resistance (4.3±2.72) was lower than those without insulin resistance (5.4±2.57) (p<0.05) (Table 2).

A negative, moderate, significant relationship was found between the HOMA-IR value and KIDMED scores of the children and adolescents (r:-0.338, p<0.001). A positive, weak, and significant relationship was found between the HOMA-IR and dietary intake of energy and protein (respectively r:0.216, p:0.004; r:0.164, p:0.030). A positive, moderate, significant relationship was found between the daily intake of carbohydrates and HOMA-IR values (r:0.264, p<0.001). There was no significant relationship between the HOMA-IR values of the participants and their intake of saturated, monounsaturated, and polyunsaturated fatty acids (PUFAs), dietary fiber, soluble and insoluble dietary fiber (p>0.05). Additionally, no significant relationship was found between the vitamin and mineral (except iron) intakes and HOMA-IR values (p>0.05). A positive, weak, and significant relationship was found between dietary iron intake and HOMA-IR (r=0.157, p=0.039) (Table 3).

Discussion

This study was conducted to evaluate the relationship between KIDMED and insulin resistance in children and adolescents with normal-weight, overweight, and obesity. The participants with obesity had lower KIDMED scores than individuals with normal weight (p<0.05). However, in line with other studies of children and adolescents in Turkey (18-20), this study was determined that more than half of the participants (52.9%) should improve Mediterranean diet adherence (average) (Table 1). Also, similar

Table 1. Distribution of children and adolescents according to KIDMED scores

KIDMED classification	Normal Weight (n=52)		Overweight (n=41)		Obesity (n=81)		Total (n=174)		χ^2/F	p
	n	%	n	%	n	%	n	%		
Low	9	17.3	10	24.4	28	34.6	47	27.0	7.418	0.115[a]
Average	28	53.9	22	53.6	42	51.8	92	52.9		
Optimal	15	28.8	9	22.0	11	13.6	35	20.1		
KIDMED score										
Mean ± SD	5.7±2.62		5.4±2.39		4.6±2.70		5.1±2.64		3.169	0.045[b*1]
minimum-maximum	0-11		0-10		0-10		0-11			

[a]Pearson chi-square test [b]One-way analysis of variance *p<0.05 [1]The significance arises from the difference between obesity and normal-weight groups, KIDMED: Mediterranean diet quality index, SD: Standard deviation

Table 2. KIDMED scores according to insulin resistance (IR) status of children and adolescents

KIDMED classification	IR + (n=37)		IR - (n=137)		Total (n=174)		χ^2/t	p
	n	%	n	%	n	%		
Low	14	37.8	33	24.1	47	27.0	4.081	0.130[a]
Average	19	51.4	73	53.3	92	52.9		
Optimal	4	10.8	31	22.6	35	20.1		
KIDMED score								
Mean ± SD	4.3±2.72		5.4±2.57		5.1±2.64		-2.324	0.021[b*]
minimum-maximum	0-9		0-11		0-11			

[a]Pearson chi-square test [b]Student's t test *p<0.05, KIDMED: Mediterranean diet quality index, SD: Standard deviation

Table 3. Correlation of HOMA-IR Value and KIDMED scores, energy and nutrient intake of children and adolescents								
Energy, some nutrients, KIDMED Score	Normal weight (n=52)		Overweight (n=41)		Obesity (n=81)		Total (n=174)	
	r	p	r	p	r	p	r	P
KIDMED score	-0.273	0.050[b]	-0.415	0.007 [b]*	-0.283	0.011 [b]*	-0.338	<0.001[b]*
Energy (kcal)	0.095	0.505[a]	0.274	0.083[a]	0.288	0.009[b]*	0.216	0.004[a]*
Carbohydrates (g)	0.019	0.891[a]	0.260	0.100[b]	0.356	0.001[a]*	0.264	<0.001[b]*
Dietary fiber (g)	0.106	0.455[b]	-0.021	0.897[b]	0.139	0.215[a]	0.129	0.089[b]
Soluble fiber (g)	0.031	0.827[b]	0.031	0.848[a]	0.225	0.043[b]*	0.128	0.092[b]
Insoluble fiber (g)	0.152	0.281[a]	-0.062	0.701[a]	0.240	0.031[b]*	0.148	0.052[b]
Protein (g)	0.013	0.928[b]	0.088	0.584[a]	0.167	0.137[b]	0.164	0.030[b]*
Fat (g)	0.093	0.513[a]	0.084	0.604[a]	0.001	0.993[b]	0.033	0.663[a]
SFAs (g)	-0.135	0.341[b]	0.008	0.960[a]	0.029	0.799[a]	0.010	0.898[a]
MUFAs (g)	0.200	0.156[a]	0.073	0.652[a]	-0.014	0.898[a]	0.080	0.292[b]
PUFAs (g)	0.051	0.722[a]	0.075	0.639[a]	-0.113	0.317[b]	0.047	0.541[b]
Vitamin A (mcg)	0.024	0.867[b]	-0.275	0.081[a]	-0.115	0.308[b]	-0.096	0.207[b]
Vitamin E (mg)	0.164	0.245[a]	0.039	0.806[a]	-0.060	0.594[b]	0.092	0.228[b]
Thiamine (mg)	-0.032	0,820[a]	0.046	0.773[a]	0.086	0.445[b]	0.039	0.611[b]
Riboflavin (mg)	-0.105	0.458[b]	-0.203	0.202[a]	-0.006	0.957[a]	-0.023	0.768[b]
Niacin (mg)	-0.102	0.473[b]	0.078	0.626[a]	0.170	0.128[b]	0.147	0.053[b]
Vitamin B_6 (mg)	0.062	0.661[a]	0.191	0.232[a]	-0.102	0.363[a]	0.103	0.175[b]
Folate (mcg)	0.236	0.092[a]	-0.116	0.469[a]	0.058	0.604[b]	0.072	0.344[b]
Vitamin B_{12} (mcg)	0.009	0.950[b]	-0.203	0.204[a]	-0.074	0.510[b]	-0.111	0.145[b]
Vitamin C (mg)	-0.005	0.974[b]	0.070	0.665[a]	-0.088	0.432[b]	-0.016	0.832[b]
Sodium (mg)	0.029	0.838[a]	-0.030	0.852[a]	0.207	0.064[a]	0.129	0.089[a]
Potassium (mg)	-0.081	0.570[b]	0.060	0.709[a]	-0.129	0.251[a]	0.030	0.695[b]
Calcium (mg)	-0.214	0.127[a]	-0.161	0.315[a]	0.007	0.947[a]	-0.077	0.310[a]
Phosphorus (mg)	-0.050	0.641[a]	-0.084	0.600[a]	0.044	0.698[a]	0.049	0.518[a]
Magnesium (mg)	-0.066	0.727[a]	-0.033	0.840[a]	-0.037	0.746[a]	0.083	0.278[b]
Iron (mg)	0.241	0.085[b]	0.007	0.963[a]	0.066	0.560[b]	0.157	0.039[b]*
Zinc (mg)	0.183	0.194[a]	-0.007	0.965[a]	0.035	0.757[a]	0.051	0.501[b]

[a]Pearson correlation [b]Spearman correlation *p<0.05, SFAs: Saturated fatty acids, MUFAs: Monounsaturated fatty acids, PUFAs: Polyunsaturated fatty acids, KIDMED: Mediterranean diet quality index, HOMA-IR: Homeostatic model assessment of insulin resistance

results were seen in other countries (Turkish Republic of Northern Cyprus, Greece, Italy) with a coast to the Mediterranean (21-23). In Sicily, the Mediterranean Adequacy Index scores of normal weight children were higher than those of overweight (24). Although the KIDMED score varies according to countries and age groups, it has been determined that adherence to the Mediterranean diet should be improved, and this result is supported by a systematic review published by Garcia Cabrera et al. (25). Improving compliance with the Mediterranean diet will be beneficial for health because the Mediterranean diet's effects on the prevention of obesity and other nutrition-related chronic diseases are well known (26,27).

Increasing compliance with the Mediterranean diet is an effective method in reducing the risk of developing chronic diseases (28,29). Additionally, the Mediterranean diet components positively affect body composition and metabolic health (30,31). It has been reported that individuals who comply with the Mediterranean diet in different populations have a low level of insulin resistance (8,10,32,33).

In this study, the mean KIDMED score of children and adolescents with insulin resistance (4.3±2.72) was found to be lower than those without insulin resistance (5.4±2.57) ($p<0.05$) (Table 2). Also, while the Mediterranean diet quality decreased, the HOMA-IR value was found to be increased ($p<0.05$) (Table 3). The Mediterranean diet, which is rich in components such as polyphenols, unsaturated fatty acids, and dietary fiber, has positive effects on insulin sensitivity (34-36). Among the foods recommended for daily consumption in the Mediterranean diet, vegetables, fruits, whole grains, legumes, nuts, olive oil, and tea are important sources of polyphenols (35,37). Polyphenols, which have antioxidant and anti-inflammatory properties, contribute to insulin resistance prevention through various mechanisms. These mechanisms include slowing carbohydrate digestion and glucose absorption, stimulation of insulin secretion, activation of insulin receptors, and glucose uptake in insulin-sensitive tissues (38). It has also been reported that polyphenols can activate and/or inhibit transcription factors, therefore affect gene expression, and contribute to glucose homeostasis by regulating different signaling pathways in muscle, liver, pancreatic β-cells, hypothalamus, and adipose tissue (35). MUFAs and PUFAs from olive oil and nuts can also have beneficial effects on insulin sensitivity by improving the inflammatory responses of adipose tissue (39,40). It was stated that MUFA, essentially oleic fatty acid, can prevent insulin resistance in the myotubes by the activation of PI3K and a mechanism dependent on amp-activated protein kinase (41). Proinflammatory cytokines and chemokines such as tumor necrosis factor-alpha, interleukin-6, and resistin, which are overproduced by dysfunctional adipose tissue in obesity, can trigger insulin resistance by activating intracellular pathways in insulin-target tissues. However, the anti-inflammatory potential of PUFAs may indirectly improve peripheral insulin response and reduce the risk of glyco-metabolic changes in patients with insulin resistance. Additionally, PUFAs can increase the secretion of glucagon-like peptide 1 (GLP-1) hormone from enteroendocrine L cells. With the consequences of increased glucose uptake from skeletal muscles by stimulating insulin release from pancreatic cells, increased GLP-1 levels can prevent postprandial hyperglycemia. Besides, GLP-1 can affect satiety at the central nervous system level, reducing the feeling of appetite (39). As compliance with the Mediterranean diet can lead to positive health outcomes such as preventing obesity, increasing insulin sensitivity, and reducing cardiovascular disease risk, healthy nutrition education should be made widespread to cover all age groups starting from childhood.

This study was found that the HOMA-IR value increased significantly as the energy, carbohydrate, protein, and iron intake of the participants increased ($p<0.05$) (Table 3). No significant relationship was found between the HOMA-IR value of the participants and their intake of dietary fiber, water-soluble fiber, water-insoluble fiber, fat, saturated fatty acids, monounsaturated fatty acids, polyunsaturated fatty acids, vitamins, and minerals (except iron) ($p>0.05$) (Table 3). The types and amounts of carbohydrates, which are the body's primary energy source, have substantial effects on metabolic health (42). Excessive consumption of carbohydrates triggers insulin resistance by increasing insulin secretion (43). Increased energy intake with a diet high in added sugar and fructose is associated with adverse health outcomes such as obesity, insulin resistance, high blood pressure, and dyslipidemia (44). On the other hand, dietary fiber affects the glycemic index of the food, delaying gastric emptying time and decreasing glucose absorption (45). For these reasons, in the prevention and treatment of insulin resistance, adequate energy intake should be provided with a diet plan containing healthy carbohydrate types by the individual's requirements. Additionally, the carbohydrate content of the diet should be composed of foods with a low glycemic index and load, such as whole grains, legumes, vegetables, and fruits, instead of foods with high glycemic index and load, such as sucrose and refined grains. Also, dietary protein and amino acids may affect insulin sensitivity and glucose metabolism. However, it is reported that the results of human studies are contradictory (46). Additionally, the results of studies on the effect of high protein diets on insulin resistance are also conflicting (47-50). For this reason, it is a critical necessity to evaluate the daily protein intake individually, taking into account the protein amounts of animal and plant origin in the diet. In order to discuss the effect of dietary fat on insulin resistance, the fat content and fatty acid pattern of the diet should be analyzed well. The excess amount of total fat intake with the diet, trans fatty acids, and saturated fatty acids may be associated with insulin resistance. In contrast, the MUFAs and PUFAs may have positive effects on insulin sensitivity (51).

One of the findings of this study was the positive correlation between dietary iron and HOMA-IR (Table 3). It has been reported that excessive iron overload in the body is associated with insulin resistance in the liver and adipose tissue by increasing gluconeogenesis, pancreatic β-cell mass (increase in insulin release), and decreasing adiponectin levels (52). Considering that excessive iron increases may potentially play a role in the pathogenesis of insulin resistance, it may be beneficial to control serum iron levels and dietary iron intake of individuals with insulin resistance. Besides, dietary iron intake should be maintained at an optimal level to prevent both deficiency and excess. Iron supplements should only be used in a controlled manner when deemed necessary by the physician.

Study Limitations

A limitation of our study is that Tanner stage of puberty and sex hormones were not assessed in our subjects. In the study, the age range was 6-17 years. The overweight and normal weight sample could not be reached as much as the obesity sample during the research period. In future studies, it will be beneficial to increase the number of samples and reach the BMI groups in more similar numbers by children in the same age range.

Conclusion

This study determined that compliance with the Mediterranean diet should be improved in children and adolescents. Also, low compliance with the Mediterranean diet was associated with an increased risk of insulin resistance. Insulin resistance and obesity are among the preventable health problems with a healthy diet and lifestyle changes. For this reason, it is essential to acquire the correct nutritional habits in childhood. Gaining adequate and balanced dietary habits in the early period contributes to preventing nutrition-related health problems such as obesity and insulin resistance in childhood and adulthood.

Ethics

Ethics Committee Approval: The study was conducted according to the guidelines laid down in the Declaration of Helsinki, and all procedures involving human subjects/patients were approved by the Ankara University Faculty of Medicine Human Research Ethics Committee (Decision no: İ2-32-19, date:).

Conflict of Interest: The authors declare that they have no conflict of interest.

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