

# Use of Acti-Heart® in Diagnosis, Follow-Up, and Evaluation of Obesity in Childhood and its Relationship to Other Metabolic Parameters

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## Abstract

**Objectives:** In this study we evaluated changes in lifestyle and motivation, obesity diagnosis, diet and exercise treatments in adolescents using Acti-Heart®, as well as effects on body composition, lipid profile, insulin resistance (IR), adipocytokines, basal metabolic rate and daily calorie consumption.

**Materials and Methods:** A total of 14 cases with an age range of 10.1-16.6 years, and puberty stage of III-IV, who were followed up in our department with a diagnosis of exogenous obesity were included. Thirteen children were included as the control group. Body mass index (BMI), waist and hip circumferences, waist/hip ratio, body fat percentage, skin fold thickness measurements were performed, and glucose, insulin, homeostasis model assessment of IR (HOMA-IR), total cholesterol, low-density lipoprotein cholesterol (LDL-C), very-low-density lipoprotein cholesterol (VLDL-C), triglyceride, serum aspartate and alanin aminotransferase (AST and ALT), visfatin, tumor necrosis factor-alpha (TNF-α), interleukin-10 (IL-10) and apolipoprotein A (APO A) levels were determined. Calorie consumption was measured using Acti-Heart®.

**Results:** In our patient group, BMI, ALT, glucose, insulin, HOMA-IR, total cholesterol, LDL-C, VLDL-C, triglyceride, visfatin values were high, but TNF-α, IL-10 and APO A levels were low (p<0.05). BMI, waist and hip circumferences, waist/hip ratio, body fat percentage and skin fold thickness decreased in with diet, exercise, and behavioral therapy. HOMA-IR, AST, ALT, VLDL-C, triglyceride, visfatin and hs-C-reactive protein values were also decreased. However, TNF-α, IL-6, and IL-10 levels increased. Daily calorie consumptions measured by Acti-Heart® significantly increased (p=0.004).

**Conclusion:** This is important as it shows increased calorie consumption, parallel changes in body composition, and improvements in metabolic parameters such as decreased VLDL, triglyceride, and IR using Acti-Heart® in obesity treatment monitoring in adolescents. In conclusion, Acti-Heart® is an objective evaluation method for monitoring obesity treatment. The study is planned to be conducted in larger groups of obese children.

**Keywords:** Acti-Heart®, BMI, diet, HOMA-IR, lipid profile, obesity, visfatin



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## INTRODUCTION

Obesity is a complex, multifactorial disease caused by an imbalance in calorie intake and energy expenditure. The most important cause of obesity is the intake of more energy than consumed. Childhood obesity refers to an unhealthy excess of body fat.<sup>1</sup> The worldwide increase in obesity, which can adversely affect the health of children, appears to be largely influenced by environmental factors, lifestyle and cultural aspects.<sup>2</sup> Childhood obesity has emerged as a global health problem due to its increasing prevalence in both developed and developing countries.<sup>3</sup> Currently, approximately 170 million children worldwide are overweight or obese.<sup>3,4</sup>

Childhood obesity serves as a major contributing factor to the development of several diet-related chronic conditions, including later life conditions such as heart disease, high blood pressure, stroke, type II diabetes, and several types of cancer.<sup>5</sup> Various treatment methods such as appropriate diet preparation, increased physical activity, behavioral modification, pharmacotherapy, and surgical procedures have been employed in obesity treatment.<sup>6-11</sup>

In obesity, the most important source of pro-inflammatory cytokines is macrophages that infiltrate adipose tissue in response to fat cell growth, reduced blood flow, hypoxia, and tissue necrosis. These events collectively create a predisposition to systemic inflammation, a potential triggering factor in the pathogenesis of obesity-related morbidities. Several adipokines including adiponectin, leptin, resistin, visfatin, chemokine, tumor necrosis factor-alpha (TNF- $\alpha$ ), interleukin-1 (IL-1), IL-6, IL-8, IL-10, plasminogen activator inhibitor 1, monocyte chemoattractant protein-1, and retinol binding protein-4 are involved in insulin resistance (IR) regulation. In healthy obese adults, increased transforming growth factor beta 1 and IL-6 inhibit cell differentiation and the function of adiponectin and leptin. Increased IL-6, IL-1 and TNF- $\alpha$  in obese individuals was associated with progression of several disorders including cardiovascular disease, hypertension and IR. Mortality is associated with increased circulating IL-6, IL-1 $\beta$ , TNF- $\alpha$  and IL-8.<sup>12</sup>

The aim of those treatments is to preserve the body weight for a long time after the suitable body weight is reached, and to prevent weight gain. In this study, we used Acti-Heart® as a monitoring tool of daily calorie consumption in a group of obese adolescents.

## MATERIALS AND METHODS

A total of 14 cases with the age range of 10.1-16.6 years and puberty stage of III-IV, who were followed up at the Department of Pediatric Endocrinology and Metabolic Diseases in the Medical School of Çukurova University with the diagnosis of

exogenous obesity were included in this study. These cases were recently diagnosed and had no known systemic, endocrine or neurological diseases. There were 13 children in the control group. Anthropometric measurements and detailed physical examinations were performed in all subjects.

Venous blood samples from participating cases were collected after a 12-hour overnight fast, before, in the middle of, and after the exercise and diet treatments. Fasting blood glucose, fasting insulin, low-density lipoprotein cholesterol (LDL-C), very-low-density lipoprotein cholesterol (VLDL-C), high-density lipoprotein cholesterol (HDL-C), triglyceride, lipoprotein a, serum aspartate and alanin aminotransferase (AST and ALT), free-T4, thyroid stimulating hormone, insulin-like growth factor 1 (IGF-1), follicle-stimulating hormone, luteinizing hormone, estradiol (in females), testosterone (in males and females), C-reactive protein (CRP) (high sensitive), TNF- $\alpha$ , IL-6, IL-10, adiponectin, and visfatin levels were measured in the blood samples in the central laboratory at the Medical School of Çukurova University.

Serum transaminases were measured using the International Federation of Clinical Chemistry method, and the Roche Modular System/Integra 800 device and kit (Germany). Alanine aminotransferase and aspartate aminotransferase levels  $\geq 40$  IU/L were accepted as abnormal serum aminotransferase values. Serum lipid profile, HDL-C, LDL-C and VLDL-C measurements were performed using enzyme calorimetric methods and Roche modular system/integra 800 device and kit (Germany). Triglyceride levels were measured with GPO/PAP method and Roche modular system/integra 800 device and kit (Germany). Lipoprotein A, Apo A and Apo B were measured using immunoturbidimetric method and Roche modular system/integra 800 device and kit (Germany). Serum glucose was measured with hexokinase method and Roche modular system/integra 800 device and kit (Germany). Serum CRP was measured using nephelometric method and II Dade Behring device and kit. Cytokines, TNF- $\alpha$ , IL-6 and IL-10 were measured using micro-enzyme-linked immunosorbent assay (ELISA) automation system (Biosource kit, Belgium), and Triturus (Spain) micro-ELISA automation system. IGF-1 was measured with chemiluminescence method and immutate 2000 and the kit. Visfatin C-Terminal (Human) was measured in the serum using micro-ELISA method Triturus (France) automatic ELISA device (Human). EIA kit (Phoenix pharmaceuticals, USA) was used for the testing. Normal plasma range for visfatin is 0.1-1000 ng/mL. Adiponectin was studied in the serum by using micro-ELISA method, and Triturus (France) automated ELISA device. BioVendor Human Adiponectin ELISA kit was used for testing. IR and insulin sensitivity indices were calculated.

Electrocardiography (ECG), echocardiography, abdominal ultrasonography, were performed, and arterial intimal thicknesses were measured.

A skinfold caliper was used to determine body fat percentage, and measured values were put in the Yuhasz formula. Direct body fat measurement was performed using the bioelectrical impedance method. Resting metabolism rate was calculated using the calorimetric method.

The children we included in the study were given a motivational, dietary and exercise program for two months.

### Regulation of the Diet Program

Patients and families were first informed about the forms to be filled out. Then, diet records were collected, in which the amounts of main and intermediate meals and beverages for one week were written with the criteria determined by the dietician, (such as tea glass, water glass, tablespoon). From these records, average calorie consumption was calculated by a pediatric dietician, taking into account weekdays and weekend days. Carbohydrate, protein, and fat ratios in the diet were also calculated. After this stage, the required daily energy amount was calculated for each patient according to age, gender, and ideal weight. The ideal weight, normal weight, height standard data, and puberty stages of school-age children were taken into consideration.

For each case, the diet was organized in accordance with the age, socioeconomic, and cultural conditions. Nutrients were selected, and the daily energy intake was organized as 50-55% carbohydrate, 15-20% protein, and 30% fat. In this way, the energy intake of the child was limited and food consumption was balanced. Dietary patterns, caloric intake, and energy distribution were obtained from the patients' seven-day nutritional records. Nutritional mistakes and deficiencies were discussed with the patients. The results of this dietary approach were explained in detail. In patients who were not morbidly obese, a balanced diet suitable for the required weight was given. In morbidly obese patients, short-term energy restriction was applied. The fact that the patients had not completed their growth was taken into consideration. Patient's daily energy intake was reduced by 200-500 calories. Patients were checked by a dietitian before, during, and at the end of the study. During the controls, it was detected that some subjects wanted to get faster results, and tried to consume fewer calories than the calories in the given diet. These patients were interviewed again, and their mistakes were corrected.

### Daily Calorie Expenditure

Acti-Heart® is a light and portable device which records data about heart rate and physical activities at minute intervals via two ECG electrodes placed on the chest. The daily caloric expenditure of the patients was measured before and at the end

of the study with the Acti-Heart® version 2,000.10 (Mini Mitter Company, Inc. USA) device (Figure 1) by entering the age, gender, weight and height data of the patients (covering two days of caloric expenditure on weekdays and weekends). The obtained data, including information about age, gender, weight, and height, were recorded on the computer, and then calculations were performed using a pre-existing program.

It was strongly emphasized to patients they should not perform any activity other than their normal activities during the time the device was worn. Daily calorie intake was calculated using a one-week diet recorded at home. Meal plans were prepared considering the age, socioeconomic conditions, and cultural characteristics of each patient.

A two-month home exercise program, which was based on basal heart rate values, was scheduled. A follow-up chart for exercise pulses, which was calculated for each age-group, was prepared. All measurements were performed at the beginning, the middle, and the end of the treatment.

### Ethical Statements

The study was approved by Çukurova University Faculty of Medicine Ethics Committee (approval number: 2008-15, dated: 03.06.2008). The study was performed in accordance with the ethical rules based on the principles of the Helsinki Declaration. Written informed consent forms were obtained (when appropriate) from the parents and the children.

### Statistical Analysis

Data obtained from this study were analyzed using Statistical Package for Social Sciences for Windows, version 10 (IBM Inc., Armonk, NY, USA). Data were expressed as the mean  $\pm$  standard deviation (SD), median (min.-max.). While evaluating the study data, in addition to descriptive statistical methods (mean, SD), analysis of variance table test was used to compare the obese patient group and the control group. The parameters of the

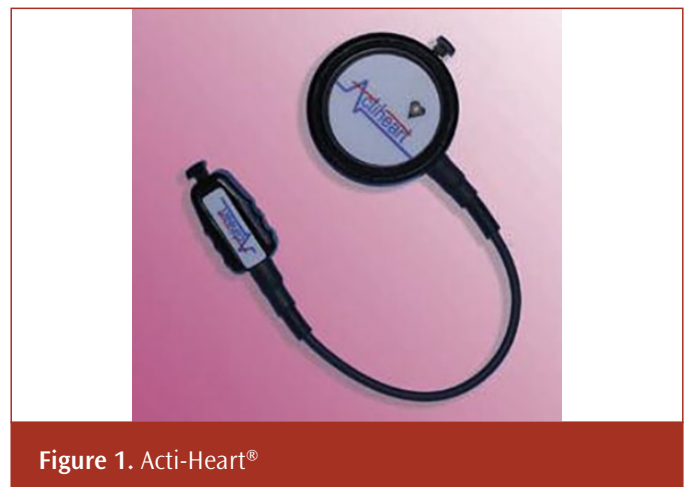


Figure 1. Acti-Heart®

obese patient group that changed with exercise were evaluated with the paired samples t-test. The results were evaluated within the 95% confidence interval and the significance level was  $p < 0.05$ .

## RESULTS

In this study, 27 children (14 were obese, and 13 were healthy children) with the mean age of  $12.92 \pm 1.94$  (range: 10-17) years were included. Of the children, 14 (51.9%) were girls and 13 (48.1%) were boys. Out of the cases, 14 were obese, and 13 were non-obese healthy (control) children. The mean weight of the patient group was  $87.96 \pm 22.31$  kg (min.: 55, max.: 133 kg), and of the control group was  $47.08 \pm 11.54$  kg (min.: 28, max.: 66 kg). There was a statistically significant difference in body weights between patients and controls ( $p=0.001$ ). The mean BMI in obese children was  $34.35 \pm 5.23$  (ranging from 28 to 45)  $\text{kg}/\text{m}^2$ , while it was  $20.02 \pm 2.95$  (ranging from 16 to 27)  $\text{kg}/\text{m}^2$  in the control group; there was a statistically significant difference between the groups ( $p=0.001$ ). Body measurements in the patient group before, in the middle, and at the end of the exercise and diet treatment are displayed in Table 1.

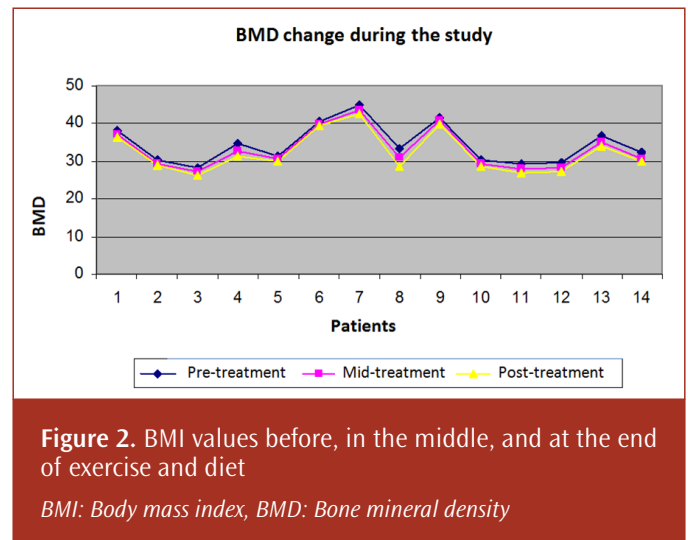
There were statistically significant differences in BMI, skinfold thickness, waist and hip measurement, and waist/hip ratio between pre- and post-treatment measurements. There was a significant difference in the body fat percentage between pre- and post-treatment calculations. The BMI values of the patients before, in the middle, and at the end of exercise and diet are displayed in Figure 2.

There were statistically significant differences in AST, ALT, VLDL-C, triglyceride, and total cholesterol measurements

between pre- and post-treatment measurements (in the same order  $p=0.015$ ,  $p=0.029$ ,  $p=0.015$ ,  $p=0.013$ ,  $p=0.028$ ).

When the correlation between obesity and adipokines was investigated, mean TNF- $\alpha$  level was detected as  $1.66 \pm 2.33$   $\text{pg}/\text{mL}$  in obese children; it was  $3.62 \pm 2.35$   $\text{pg}/\text{mL}$  in the control group. The difference was statistically significant. The mean IL-10 levels were  $0.28 \pm 0.64$   $\text{pg}/\text{mL}$  and  $5.61 \pm 8.84$   $\text{pg}/\text{mL}$  in obese children and the control group, respectively. The difference was statistically significant. Differences in hs-CRP, IL-6, adiponectin and visfatin values between patient and control groups were not considered significant. The comparison of adipokines between patient and control groups is provided in Table 2.

There was a significant difference in homeostasis model assessment of IR (HOMA-IR) measurements between pre- and



**Table 1. Body measurements in the patient group before, in the middle and at the end of exercise and diet treatment**

Variables	Measurements	n	Mean	SD	p-value
BMI ( $\text{kg}/\text{m}^2$ )	Pre-treatment	14	34.35	5.23	0.001
	Mid-treatment	14	33.03	5.32	
	Post-treatment	14	32.10	5.35	
Waist (cm)	Pre-treatment	14	105.18	14.66	0.001
	Post-treatment	14	97.75	12.75	
Hip (cm)	Pre-treatment	14	109.61	10.83	0.003
	Post-treatment	14	106.79	10.80	
Waist/Hip ratio	Pre-treatment	14	0.97	0.09	0.029
	Post-treatment	14	0.91	0.08	
Skinfold thickness	Pre-treatment	14	25.43	2.43	0.003
	Post-treatment	14	23.83	2.06	
Body fat percentage (%)	Pre-treatment	14	33.76	11.77	0.007
	Post-treatment	14	31.98	11.49	

The parameters of the obese patient group that changed with treatment were evaluated by paired samples t-test. The significance level of the tests was set as  $p < 0.05$ . SD: Standard deviation, BMI: Body mass index

post-treatment ( $p=0.001$ ). Changes in IR with exercise and diet treatment in the patient group are shown in Table 3 and Figure 3.

When the calorie consumption of the patient group before and after the study, a statistically significant difference was detected between the two measurements ( $p=0.004$ ). There was no statistically significant difference in the basal metabolism rates of the patient group before and after the study ( $p=0.194$ ).

Evaluation of energy consumption with exercise and diet in the patient group. It is shown in Table 4.

## DISCUSSION

Obesity is a chronic energy metabolism disorder, which may ensue from excessive fat accumulation and cause severe problems physically, mentally. There is no obvious cause in the majority of obesity cases. These are defined as simple obesity or

**Table 2. Comparison of adipokines between patient and control groups**

Variables	Measurements	n	Mean	SD	Median (min.-max.)	p-value
TNF- $\alpha$ (pg/mL)	Obese	14	1.66	2.33	0.50 (0-7)	0.029
	Control	13	3.62	2.03	0 (0-7)	
Hs-CRP (mg/dL)	Obese	14	6.29	3.28	5.82 (3-12)	0.059
	Control	13	9.77	12.75	3.30 (3-13)	
IL-6 (pg/mL)	Obese	14	1.70	1.79	1.25 (0-5)	0.161
	Control	13	1.06	1.08	2 (0-20)	
IL-10 (pg/mL)	Obese	14	0.28	0.64	0 (0-2)	<b>0.033</b>
	Control	13	5.61	8.84	1.80 (0-32)	
Adiponectin ( $\mu$ g/mL)	Obese	14	1.21	1.33	0.60 (0-4)	0.459
	Control	13	1.63	1.59	1.20 (0-7)	
Visfatin (ng/mL)	Obese	14	115.64	19.59	13.15 (3-638)	0.419
	Control	13	64.28	11.83	16.40 (0-403)	

Significance between control and obese group (One-Way ANOVA test). Significance level of the tests was accepted to be  $p<0.05$ . SD: Standard deviation, ANOVA: Analysis of variance, TNF- $\alpha$ : Tumor necrosis factor-alpha, Hs-CRP: High-sensitivity C-reactive protein, IL: Interleukin, min.-max.: Minimum-maximum

**Table 3. Changes in IR in the patient group by exercise and diet treatment**

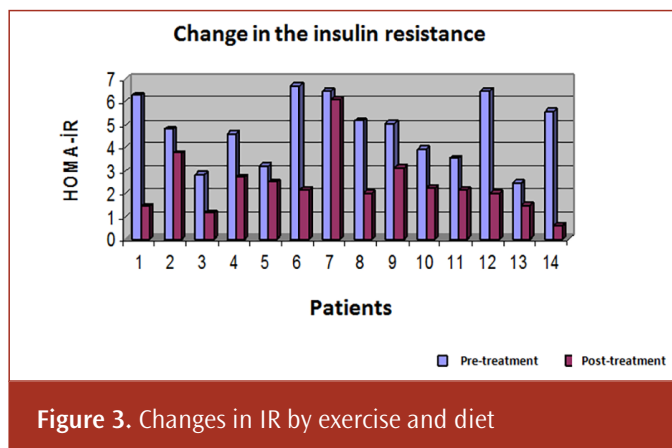
Variables	Measurements	n	Mean	SD	p-value
Glucose (mg/dL)	Pre-treatment	14	84.08	6.66	0.109
	Post-treatment	14	79.38	6.59	
Insulin (mU/L)	Pre-treatment	14	23.25	7.46	0.358
	Post-treatment	14	22.24	7.84	
Glucose/Insulin	Pre-treatment	14	4.22	1.58	0.520
	Post-treatment	14	4.31	1.70	
HOMA-IR	Pre-treatment	14	4.80	1.43	<b>0.001</b>
	Post-treatment	14	2.38	1.33	

The parameters of the obese patient group that changed with treatment were evaluated by paired samples t-test. The significance level of the tests was accepted as  $p<0.05$ . SD: Standard deviation, HOMA-IR: Homeostasis model assessment of insulin resistance

**Table 4. Evaluation of energy consumption in the patient group by exercise and diet**

Variables	Measurements	n	Mean	SD	p-value
Basal metabolism rate	Pre-treatment	14	1675.69	438.21	0.194
	Post-treatment	14	1782.29	508.66	
Daily calorie consumption	Pre-treatment	14	1254.36	491.80	<b>0.004</b>
	Post-treatment	14	1524.71	546.93	

The parameters of the obese patient group that changed with treatment were evaluated by paired samples t-test. The significance level of the tests was accepted as  $p<0.05$ . SD: Standard deviation



**Figure 3.** Changes in IR by exercise and diet

exogenous obesity. Obesity has become a public health problem all over the world with its increasing prevalence in the last two decades, especially in the Western populations.<sup>13,14</sup> The increase in obesity frequency in children is parallel with that in adults. This is caused by fats, carbohydrates, and fast-foods becoming more prominent among modern dietary habits, and children preferring to watch television and play computer games, rather than performing physical activities.<sup>15,16</sup> Obesity and the severe complications that this pathological condition may lead to have increased the interest in and need for effective, easily applicable, and simpler methods to prevent and treat obesity. Diet, exercise, and behavioral motivation are highly important and effective approaches.<sup>17,18</sup> If exercise treatment is accompanied by diet and behavioral modifications in obese patients, it causes more weight loss than diet alone. Hensrud et al.<sup>19</sup> conducted a two-year study by dividing people into diet only, exercise only, and diet plus exercise groups. In the diet plus exercise group, a mean, 13 kg weight loss was observed among 24 obese women after 1 year. While there was a 6 kg loss in the exercise only group, a 13 kg weight gain was observed in the diet only group.<sup>19</sup>

When the changes in lipid profile in the obese patient group before and after exercise were compared, significant decreases were found in total cholesterol, triglyceride, and VLDL-C levels. There was no statistically significant difference between HDL-C, LDL-C, APO A, APO B, and lipoprotein measurements. In our study, as in the study of Kim et al.,<sup>20</sup> no significant difference was found between the values of HDL-C and LDL-C after diet and exercise treatment. We think that the small number of patients included in the study and the short duration of exercise may have caused this difference. Similar to the study of Kang et al.,<sup>21</sup> no significant changes were found in HDL-C, LDL-C, Apo A, Apo B and lipoprotein A levels with diet and exercise in our study.

When the relationship between obesity and liver function tests was investigated in our study, a statistically significant difference was found in ALT levels in obese children. Similar

to the study of Li et al.,<sup>22</sup> no significant difference was found in AST measurements in our study. When AST and ALT values before and after exercise were compared in the obese patient group, a statistically significant decrease was found between the two measurements. This finding supports the view that exercise improves liver function by regulating energy and lipid metabolism. Jung et al.<sup>23</sup> found significant decreases in TNF- $\alpha$  and IL-6 levels and significant increases in IL-10 and adiponectin levels in obese individuals after a 12 week exercise program.

In the study by Kim et al.,<sup>20</sup> IL-6, TNF- $\alpha$  and hs-CRP levels were found to be significantly elevated in obese individuals, whereas adiponectin levels were found to be significantly lower. While adiponectin levels were found to be low in the same individuals after exercise, no significant difference was found in IL-6, TNF- $\alpha$ , and hs-CRP levels.<sup>20</sup> In our study, TNF- $\alpha$  and IL-6, which are inflammation markers, were found to increase after diet and exercise, contrary to expectations. However, the level of IL-10, an anti-inflammatory cytokine, was found to be lower in obese children compared to the control group, as expected. It increased with exercise. In our study, we attributed the increase in IL-6 and TNF- $\alpha$  to the effect of exercise on muscles, noting that IL-6 and TNF- $\alpha$  are myokines released from muscles during exercise, which demonstrates the effectiveness of exercise by an increase in their levels.<sup>24,25</sup>

In our study, when serum adiponectin levels were examined, no significant difference was found in obese children compared to the control group, nor was there a significant difference in obese children after exercise compared to before exercise. We think that the discordance in adiponectin levels in this study may be related to the insufficient number of patients included in the study, and the short duration of exercise. Furthermore, we suggest that longitudinal studies should be performed in large patient groups in the future to obtain clearer data.

In our study, we did not find a significant difference in serum visfatin levels in obese patients compared to the control group; however, we found a significant decrease in visfatin levels with exercise in obese children. This finding supports the view that obesity reduction may be due to the positive effect of exercise, which has been previously emphasized in the literature.

Crouter et al.<sup>26</sup> performed a study on energy consumption measurement and the reliability of Acti-Heart® in adults. The study was performed on 48 patients (24 males and 24 females; mean age: 35 years). Patients were divided into three groups according to their lifestyles and exercising habits (sedentary time, time at home, and exercising), and concomitant oxygen consumption of patients was measured. Six routine activity programs were scheduled for patients as 10 minutes of physical activity and 1-2 minutes of rest. Heart rate and energy consumption were

measured by Acti-Heart® by estimating heart activity, and the recorded data during all activities were transferred to the computer. In the meantime, energy consumption was measured during each routine activity by using the portable indirect calorimetric method (Cosmed K4b2, Italy). The study is valuable as it has indicated the reliability of measurement by Acti-Heart®.<sup>26</sup> In another study performed by Barreira et al.,<sup>27</sup> energy consumption during short-term physical activity was measured in 34 healthy subjects with the mean age of 21.8 months using Acti-Heart®, and it was reported that there was an increase in daily calorie consumption after the exercise program, which was measured by Acti-Heart®. Changes in daily calorie consumption were measured in obese adolescents before and after a triple treatment program of behavioral motivation, diet, and exercise for one day per week and two days on the weekend. A significant difference was determined in daily calorie consumption of obese patients, before and after treatment ( $p=0.004$ ). Thus, we believe that by using Acti-Heart® we have objectively assessed energy expenditure in the diagnosis, treatment and follow-up of pediatric obesity. Moreover, we think that since the study results showed significant correlations between energy consumption measured before and after treatment by Acti-Heart® and BMI, skin fold thickness, body fat percentage, waist/hip ratio, hepatosteatosis, HOMA-IR, plasma lipid, IL-6, IL-10, TNF- $\alpha$ , hs-CRP, and visfatin levels, further research should explore these correlations in greater detail.

## CONCLUSION

In our study, we showed that energy consumption increases during obesity treatment in adolescence by changing the lifestyle through behavior, diet, and exercise approaches. We used Acti-Heart® to assess calorie consumption in the diagnosis, treatment and follow-up of adolescent obesity. We also showed that Acti-Heart® was an easily applicable, non-invasive, cost-effective (no additional cost other than electrodes) device, and it could gather information about the energy consumption in every part of daily life, including sleeping and sports. Moreover, it was well-correlated with other parameters in obesity treatment follow-up; therefore, it is also an important and practical evaluation method. However, we believe that larger studies should be performed to support our conclusions.

## Ethics

**Ethics Committee Approval:** The study was approved by Çukurova University Faculty of Medicine Ethics Committee (approval number: 2008-15, dated: 03.06.2008).

**Informed Consent:** Written informed consent forms were obtained (when appropriate) from the parents and the children.

## Footnotes

### Authorship Contributions

Surgical and Medical Practices: İ.Ö., S.E., A.K.T., B.Y., N.Ö.M., Concept: İ.Ö., E.M., S.E., G.D., A.K.T., B.Y., N.Ö.M., Design: İ.Ö., E.M., S.K., G.K., G.D., N.Ö.M., Data Collection or Processing: İ.Ö., S.K., G.K., K.A., G.D., A.K.T., B.Y., N.Ö.M., Analysis or Interpretation: İ.Ö., E.M., S.K., G.K., S.E., K.A., F.T., F.G., G.D., A.K.T., B.Y., N.Ö.M., Literature Search: İ.Ö., E.M., F.T., F.G., G.D., A.K.T., B.Y., N.Ö.M., Writing: İ.Ö., E.M., N.Ö.M.

**Conflict of Interest:** The authors have no conflicts of interest to declare.

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