## Title
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**Authors**
Eliakim, A
Makowski, GS
Brasel, JA
et al.

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# Adiposity, Lipid Levels, and Brief Endurance Training in Nonobese Adolescent Males

A. Eliakim[1], G. S. Makowski[2], J. A. Brasel[3], D. M. Cooper[1]

[1] Department of Research, Connecticut Children's Medical Center, University of Connecticut Health Center, Hartford, CT, USA
[2] Department of Laboratory Medicine, University of Connecticut Health Center, Farmington, CT, USA
[3] Division of Pediatric Endocrinology, Harbor-UCLA Medical Center, Torrance, CA, USA

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**Physical activity during childhood and adolescence may influence the development of childhood obesity and cardiovascular disease later in life. Research focused prospectively on the effects of training on lipid levels in nonobese subjects, and studies using noninvasive measurements of subcutaneous and intraabdominal fat are lacking. It was hypothesized in nonobese sedentary adolescent males that a brief endurance-type exercise training intervention would reduce body fat and improve lipid profiles. Thirty-eight healthy, nonobese sedentary adolescent males (mean age 16 ± 0.7 years old; 18 controls, 20 trained) completed a 5-week prospective, randomized, controlled study. Adiposity was measured using magnetic resonance images of the thigh and abdomen (subcutaneous abdominal adipose tissue [SAAT] and intraabdominal adipose tissue [IAAT]). Lipid measurements included serum triglycerides (TG), total cholesterol (TC), HDL and LDL cholesterol. There was no change in body weight in either control or training groups. Training led to small but significant reductions in thigh fat (-4.6 ± 1.5%, $p < 0.03$) and SAAT% ( 1.7 ± 0.8%, $p < 0.02$). There was no change in IAAT%. Unexpectedly in the control group there were significant increases in thigh fat (5.2 ± 1.7%, $p < 0.01$), SAAT% (1.8 ± 0.6, $p < 0.007$) and IAAT% (4.5 ± 1.1, $p < 0.0007$). Training-induced changes in adiposity were not accompanied by changes in circulating lipids. In nonobese adolescent males a brief period of endurance training led to reductions in body fat depots without weight change while body fat increased rapidly in the control group. Exercise training did not change lipid levels, the latter may require more sustained alterations in patterns of physical activity.**

**Key words: Fitness, cardiovascular risk, growth, development.**

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## Introduction

Childhood obesity is the most common pediatric chronic illness in western countries. The prevalence of childhood obesity in North America ranges from 10 – 25% and increased markedly in the last two decades [3]. There is growing evidence that body adiposity and its distribution are related to levels of physical activity and to coronary artery disease risk factors (e.g. circulating cholesterol and lipoprotein levels) in both children and adults [8,19,24,26]. The interaction of physical activity and cardiovascular risk factors has been examined in sample populations of obese children and adolescents [17,22,23]. However, research focused prospectively on the effects of training on subcutaneous and intraabdominal fat, and on lipid levels in nonobese subjects are lacking. This is important because understanding the "normal" adaptation of the adipose tissue to the stress of exercise in nonobese children is necessary to better understand the epidemic of childhood obesity and the response of obese children to exercise training. Understanding these relationships is particularly important in children and adolescents since it is becoming clear that increasing risk of heart disease during adulthood may be related to nutritional and exercise patterns established earlier in life [27].

In this study we hypothesized that 5 weeks of a controlled, prospective, randomized endurance type training would lead to reduced body fat and to an improved lipid profile. Magnetic resonance imaging (MRI) of the thigh and abdomen was used to measure site specific changes in thigh fat, subcutaneous abdominal adipose tissue (SAAT), and intraabdominal adipose tissue (IAAT). These measurements were accompanied by anthropometric evaluation of skinfold thickness and percent body fat.

## Methods

### *Protocol*

Forty-four healthy, sedentary adolescent boys volunteered for the study. Other aspects of this study, which focused on bone and other growth factors, have already been published [10, 12,14,15]. The participants were all students at Torrance High School (Torrance, CA) and enrolled in an honors anatomy class during the summer of 1996 (July – August) with class hours from 8.00 a.m. to 12.30 p.m. The ethnic configuration of the group was 71% Asian, 20% Caucasian, and 9% Hispanic. The study was designed to examine late pubertal subjects aged between 15 – 17 yr. Weight was measured using standard techniques. Assessment of pubertal status using Tanner staging of pubic hair [29] was performed by physical examination in all of the subjects. Seventy percent of the subjects were found to be at Tanner level V, 28 % at Tanner level IV, and 2 % at Tanner level III.

External physicians unrelated to the study prescreened subjects for participation in high school level athletics. Individuals using chronic medications (other than bronchodilators) or with a history of chronic lung or heart disease or a history of smoking, drug or alcohol abuse were excluded.

Subjects were randomized to control (n = 22) and training groups (n = 22). All subjects participated in the 2-hour daily teaching program. During the remaining time (2 – 2.5 hours/ day, 5 days/week, 5 weeks) the training group members underwent endurance-type training consisting of running, aerobic dance, competitive sports (e.g. basketball), and occasional weight-lifting. These activities were varied in duration and intensity throughout the week primarily to encourage maximal participation of the subjects. On average "aerobic" or endurance type activities accounted for about 90 % of the time spent in training. Of these about 50 % involved running, 40 % team sports, and 10 % aerobic dance. Training was directed by a member of the Torrance High School faculty. The duration of the study was five weeks.

Control group subjects participated in a computer workshop designed to improve their computer skills and used this time to analyze some of the data collected from the study. No attempt was made to influence extracurricular levels of physical activity or dietary habits of either the control or training group subjects. Staff members responsible for the anthropometric measurements, MRI studies, and serum lipid analysis were blinded to the subjects' group.

The study was approved by the Institutional Human Subject Review Board, and informed consent was obtained from the subjects and their parents or guardians.

*MRI of thigh fat*

Studies were done before and within 24 hours after the 5 week protocol in all subjects. MRI has been used previously to assess fat distribution [1]. MRI was performed on a General Electric 1.5 Tesla whole body MRI System. Briefly, 13 axial sections beginning at the knee to a level of 2 – 3 cm below the femoral neck were obtained. These axial sections were 2 cm thick with no gap and obtained with a TI weighted sequence with a time-to-echo of 12 msec and repetition time of 400 msec. The matrix was 192 × 256 with 2 acquisitions at each phase encode step.

The fat was easily identified in the serial MRI sections and measured using computerized planimetry to determine the cross-sectional area (CSA). The total thigh fat volume (in $cm^3$) was estimated by summing the volume of fat in each serial section (i.e. fat CSA [in $cm^2$] × 2 cm). The intraobserver variability was about 2 %.

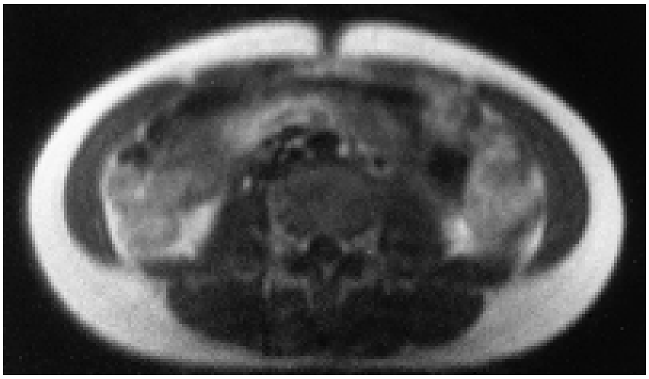

**Fig. 1** Abdominal MRI at the level of the umbilicus. Note that subcutaneous abdominal adipose tissue (SAAT) was clearly demarcated in the MR images and therefore easily measured. In contrast, intraabdominal adipose tissue (IAAT) was not as well outlined in this population of nonobese adolescent males.

*Abdominal MRI*

Abdominal MRI was performed at the level of the umbilicus (Fig. **1**). Subcutaneous abdominal adipose tissue (SAAT) was clearly demarcated in the MR images and therefore easily measured by trained observer. Intraobserver variability was about 2 %. Intraabdominal adipose tissue (IAAT) was not as well outlined in this population, and this was reflected in an intraobserver variability of about 15 %. The percent of SAAT and IAAT were calculated as the fat CSA divided by the whole abdomen CSA at the level of the umbilicus (i.e. SAAT % and IAAT %). The values are expressed as percentages because we are comparing the relative proportion of two different fat depots within the abdomen. Studies were done before and within 24 hours after the training intervention.

*Skinfold measurements*

Triceps, biceps, subscapular and supra iliac skinfolds (SF) were measured to the nearest 0.1 mm, using standard techniques [4] with Lange skinfolds calipers (Cambridge Scientific Industries, Inc. Cambridge, Maryland). Measurements were made on the right side of the body. All measurements, pre- and postintervention, were performed by the same individual. Calculation of body fat percent was based on standard equations [4]. Body density was calculated from the regression equation of Durnin and Rahaman [9] for adolescent boys. The intraobserver variability in these measurements was about 5 %.

*Blood sampling*

Fasting blood samples for lipid profile analysis were collected in the General Clinical Research Center before and within 24 hours after the training intervention. Analytical measurements were performed on an automated instrument (Model Synchron CX-7, Beckman Instruments Inc., Brea, CA). Briefly serum TG concentration was determined by timed end point method using coupled enzymatic assay with colorimetry [5]. TG intra- and inter-assay coefficient of variation (CV) were 1.4 % and 4.7 %, respectively. TG assay sensitivity was 10 mg/dl. Total cholesterol (TC) was determined by coupled cholesterol esterase/oxidase timed end-point assay with colorimetry [2]. TC intra- and inter-assay CV were 1.1 % and 5.1 %, respectively.

TC assay sensitivity was 5 mg/dl. HDL cholesterol was determined as described above following dextran sulfate-magnesium precipitation [32]. HDL cholesterol intra- and inter-assay CV were 4.4% and 5.4%, respectively. HDL cholesterol assay sensitivity was 5 mg/dl. LDL cholesterol level was calculated for samples containing TG < 400 mg/dl by the equation: LDL = Total Cholesterol – HDL – (TG/5).

*Statistical analysis*

Unpaired t-tests were used to compare data of subjects randomized to either the control or training group prior to the training intervention. Analysis of variance (ANOVA) for repeated-measures was used to determine the effect of the training intervention on thigh fat volume, SAAT%, IAAT%, anthropometric measurements, and circulating lipids with time serving as the within-group factor and training as the between-group factor. When ANOVA was found to be significant, intergroup comparisons were made using modified t-tests by the method of Duncan. Data are presented as mean ± SE. Statistical significance was taken at $p < 0.05$.

## Results

*Sample population*

Forty-four adolescent males participated in the study. One control group subject did not agree to blood sampling. During the intervention 5 subjects (3 controls and 2 training group subjects) dropped out but none for reasons of inability to perform the exercise training. Therefore, 18 subjects from the control group and 20 subjects from the training group completed the study protocol. There were no significant differences in weight, height, (Table **1**) and fitness level between the subjects randomized to the control and training groups prior to the training intervention.

**Table 1** The effects of a brief endurance-type training intervention on anthropometric measurements and lipid levels in healthy adolescent males

| | Control Group | | Training Group | |
|---|---|---|---|---|
| | Pre | Post | Pre | Post |
| Height (cm) | 170.3 ± 1.6 | 170.5 ± 1.8 | 169.2 ± 1.6 | 169.3 ± 1.8 |
| Weight (kg) | 66.2 ± 3.5 | 66.8 ± 3.3 | 61.0 ± 1.8 | 61.8 ± 2.0 |
| Triceps SF (mm) | 12.3 ± 1.2 | 12.0 ± 1.3 | 11.8 ± 1.0 | 11.0 ± 0.9 |
| Biceps SF (mm) | 5.4 ± 0.5 | 5.0 ± 0.7 | 5.1 ± 0.5 | 5.1 ± 0.5 |
| Sub Scapular SF (mm) | 11.8 ± 0.8 | 12.4 ± 1.1 | 10.7 ± 0.8 | 10.1 ± 0.7** |
| Supra Iliac SF (mm) | 10.4 ± 1.4 | 11.9 ± 1.5 | 9.1 ± 1.2 | 9.5 ± 1.2 |
| Body Fat SF (%) | 15.5 ± 1.1 | 15.9 ± 1.2 | 14.7 ± 1.4 | 14.4 ± 0.9 |
| Triglycerides (mg/dl) | 122.1 ± 23 | 129.1 ± 21 | 120.8 ± 26 | 98.8 ± 15 |
| Cholesterol (mg/dl) | 137.4 ± 5 | 139.7 ± 8 | 133.1 ± 9 | 135.3 ± 9 |
| HDL (mg/dl) | 32.6 ± 1.4 | 36.8 ± 1.6* | 37.7 ± 1.7 | 39.5 ± 1.8* |
| LDL (mg/dl) | 71 ± 6 | 72 ± 7 | 61 ± 7 | 57 ± 6 |

Results are shown as mean ± SE. *$p < 0.05$, pre vs. post the training intervention. **$p < 0.05$, for between group differences

*Prospective training intervention*

As previously reported [12,14,15], both cardiorespiratory fitness and thigh muscle volume improved in the training group (4.6% increase in peak $\dot{V}O_2$ normalized to body weight, 7.6% increase in peak $O_2$ pulse [peak $\dot{V}O_2$/peak HR], and 3.8% increase in thigh muscle volume) but was not changed in the control group. Moreover, energy expenditure measured by the doubly labelled water technique was 15.5% higher in the training group subjects [12]. This finding indicates that the training intervention was successful. There were no significant changes in body weight and height and in total caloric intake in either group during the study period.

*SAAT%*

There were no statistically significant differences in SAAT% between controls and training group subjects before the training intervention. Unexpectedly there was a significant increase in SAAT% in the control group subjects (from 27.0 ± 2.5% to 28.8 ± 2.8%, $p < 0.02$). In contrast there was a significant decrease in SAAT% in the training group subjects following the intervention (from 23.7 ± 2.1% to 21.9 ± 1.9%, $p < 0.02$). These changes were accompanied by significant between group differences ($p < 0.001$, Fig. **2**).

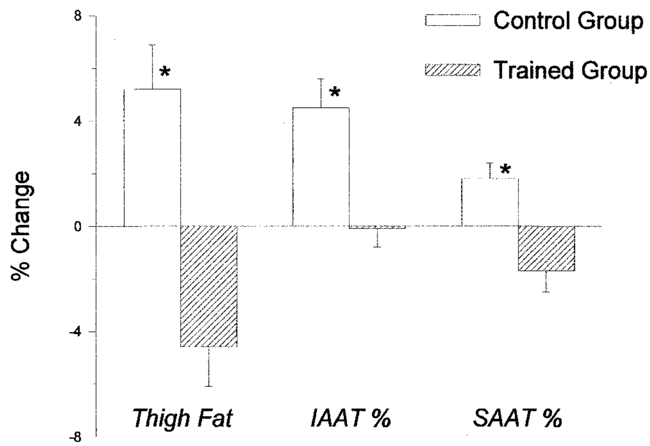


**Fig. 2** Effect of the training intervention on thigh fat volume, SAAT%, and IAAT%. Thigh fat volume, SAAT%, and IAAT% increased in the control subjects while thigh fat volume and SAAT% decreased in the trained subjects following the intervention (*$p < 0.05$ for between group differences).

*IAAT%*

There were no statistically significant differences in IAAT% between control and training group subjects before the training intervention. Similar to SAAT%, IAAT% significantly increased in the control group subjects (from 12.9 ± 1.1% to 17.4 ± 1.7%) but did not change in the training group (from 11.4 ± 1.0% to 11.2 ± 1.1%). These changes were accompanied by significant between group differences ($p < 0.001$, Fig. **2**).

### *MRI of thigh fat*

There was no significant difference in thigh fat volume between the control and training group subjects before the training program. Similar to our findings for IAAT and SAAT, there was a significant increase in thigh fat volume in the control subjects by the end of the study period (from $700 \pm 91$ $cm^3$ to $724 \pm 88$ $cm^3$, $p < 0.03$). In contrast, there was a significant decrease in thigh fat volume in the training group subjects following the intervention (from $603 \pm 61$ $cm^3$ to $574 \pm 60$ $cm^3$, $p < 0.03$). These changes were accompanied by significant between group differences ($p < 0.0002$, Fig. **2**).

### *Anthropometric measurements*

The effect of the training intervention on the skinfold measurements and percent body fat by skinfolds is summarized in Table **1**. There were no significant differences in these parameters between the control and training group subjects before the training intervention. Subscapular SF was significantly increased in the control subjects and decreased in the training group subjects. No significant change in triceps SF, biceps SF, supra iliac SF, and percent body fat by SF were observed in either the control or training group subjects.

### *Lipid profile*

The effect of the training intervention on TG, TC, HDL, and LDL is summarized in Table **1**. There were no significant differences in these parameters between the control and training group subjects before the training intervention. There was a significant within group increase in HDL levels in both the control and training group subjects. No change in serum TG, TC, or LDL levels were observed in either the control or training group subjects.

## Discussion

The prospective training intervention, although brief, led to small but significant decreases in body fat. Unexpectedly we found opposite changes in the control subjects, namely that there was a small but significant increase in adiposity over the same 5-week period. The effects on body composition in both the control and training groups were not accompanied by changes in body weight or by alterations in circulating lipids. Finally our study points toward the importance of MRI as a tool to relate biochemical and anatomic alterations in response to inputs like exercise training.

The effects of exercise training on body fat in adolescents have not been well studied, and most investigations were cross-sectional in design [8,19]. Of the few prospective studies in children and adolescents, most have demonstrated decreases in percent body fat following exercise training interventions [6,16,20] but none examined the effects over as brief a period as in the present study. In addition, the use of MRI in the present study allowed us to examine local anatomic changes in adiposity without ionizing radiation. Our study demonstrates that only 5 weeks of exercise training specifically decreases thigh fat and SAAT without measurable changes in IAAT.

The decreases in body fat noted here occurred over the same time period in which increases in fitness, muscle mass and bone formation markers were observed as reported previously [12,14]. Despite these anabolic adaptations in muscles and bones, data from the male subjects of the present study [12] and results from female adolescents studied earlier [11] suggest that the endocrine response to 5 weeks of training was best characterized as catabolic in nature: both growth hormone binding protein and insulin-like growth factor-1 were reduced, and insulin-like growth factor binding protein-2 was elevated. These changes are commonly seen in conditions associated with reduced caloric intake and marked weight loss [25,27,31] neither of which was observed in the present study [15]. Our finding of decreased fat in the trained subjects supports the hypothesis that the early responses to the imposition of an exercise training program simultaneously include both catabolic (primarily in fat stores) and anabolic (primarily muscle and bone) adaptations.

Of potentially equal importance were the unexpected changes that occurred in the control group subjects. Both thigh fat and SAAT increased, suggesting that the finding was more than just a spurious increase in adiposity occurring only in a single location. Moreover, IAAT (currently felt to be the fat depot that is most highly correlated with biochemical markers of cardiovascular risk factors [7]) also increased. We speculate therefore that in nonobese adolescent males the exercise training prevented the increases in IAAT that were observed in the control subjects.

The increase in adiposity in the male adolescents was somewhat similar to observations made in our previous study [13] in females. The control group females increased fat in the distal thigh over the five-week period of intervention. The mechanism for the increases in abdominal and thigh adiposity in the control group subjects over such a brief period is not readily apparent. It is possible however that the increase in adiposity resulted from a generally reduced daily physical activity in the summer months compared to school days.

The changes in subscapular SF among the control and training group subjects were consistent with the MRI data. However, there was no change in the other skinfold thickness measurements and in the percent total body fat by SF. This indicates that training associated changes in body fat might be site specific and/or that, compared to MRI, skinfold measurements are probably not sensitive enough to monitor small changes in adiposity over a brief period of endurance training.

Despite the significant and consistent changes in thigh and abdominal fat in the control and training subjects we did not find accompanying changes in TG, TC, and lipoproteins. To our knowledge there have been few prospective studies of the effects of brief training on TGs and lipoproteins in adolescents. Harrell et al. [20] studied the effect of 8 weeks of exercise training on elementary school children and found a significant reduction in total cholesterol level. Fardy et al. [16] also found a reduction in cholesterol after a 10 week exercise and nutrition intervention in male and female adolescents. Most studies in adults suggest that reductions in body weight with exercise training are accompanied by reductions in TGs and increases in HDL (e.g. [21]). Thompson et al. [30] found increases in HDL and decreases in TGs even when exercise training was not associated with weight loss in adults. It is possible that a training program of 5 weeks was too short to reduce lipid level, and that the differences in the training intensity might be responsible for the inconsistency of our study compared with previous results. In addition, it may be that individuals with favourable lipid concentrations such as our subjects do not alter lipid level even if fat distribution changes, particularly at young ages.

The mechanisms that determine patterns of fat distribution are not known. However, centrally located fat seems to be a fairly strong indicator of metabolic abnormalities such as hyperlipidemia and insulin resistance [18,32]. Our data suggest that exercise training may play a key role in the attenuation and/or decrease of central fat accumulation and therefore in the prevention of its related metabolic abnormalities and increased risk for coronary heart disease later in life. Moreover, the rapidity with which central and peripheral (thigh) adiposity changed in both the control and training adolescent males emphasize the need to initiate these preventive interventions in early adolescence.

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Corresponding Author:

Alon Eliakim, MD

Child Health and Sports Center
Department of Pediatrics
Meir General Hospital
Kfar-Saba
Israel

Phone: + 972 (9) 7472134
Fax. + 972 (9) 7425967
E-mail: eliakim@internet-zahav.net