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Effect of exercise training on energy expenditure, muscle volume, and maximal oxygen uptake in female adolescents

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Objectives: **American female adolescents are at high risk of a physically inactive lifestyle that likely leads to health problems later in life. We hypothesized that a brief program of endurance exercise training in female adolescents would result in increased energy expenditure and quantifiable structural and functional adaptations.**

Study design: **Forty-four high school girls (aged 15 to 17 years, none were elite athletes) enrolled in a 5-day per week anatomy class for 5 weeks and were randomly assigned to control (n = 22) and training groups. All subjects participated in a 2-hour daily teaching program. During the remaining time (2 hours), the training group members underwent endurance-type training and control group subjects participated in a computer workshop. The intervention was assessed by (1) comparison of total energy expenditure between groups with the doubly labeled water technique, (2) determination of changes in thigh muscle volume by magnetic resonance imaging, and (3) determination of changes in maximal oxygen uptake by use of respiratory gas exchange responses.**

Results: **Total energy expenditure was significantly greater (15.3%) in the training group compared with the control subjects (*p* <0.003). Five weeks of training led to a 4.3% ± 1% increase in thigh muscle volume (*p* <0.0002) and a 12.1% ± 3.7% increase in maximal oxygen uptake (*p* <0.004); there were no changes in the control group. The training effect was most pronounced in the least fit subjects.**

Conclusions: **Exercise training programs for female adolescents can be successfully integrated into a high school summer curriculum. Quantifiable, substantial structural and functional responses occur with relatively short periods of training. Approximately 60% of the training response was related to factors independent of muscle size per se. These data may serve to better design physical activity programs for female adolescents. (J Pediatr 1996;129:537-43)**

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It is becoming increasingly apparent that patterns of physical activity established during childhood and adolescence can improve health later in life.[1-3] Despite these insights, American adolescents, in particular girls, continue to be at high risk of a physically inactive lifestyle.[4] Although there is a deepening understanding of the cellular mechanisms responsible for the health-promoting effects of exercise,[5] it is not yet clear precisely to what extent training programs change the structural and functional components of fitness in a population of healthy female adolescents.

A major obstacle in gaining these insights has been the difficulty in obtaining precise measurements of the following variables: (1) the relative magnitude of the exercise "input" to the system, (2) the extent of the anatomic response (e.g., in peripheral muscles), and (3) the degree of functional adaptation (e.g., change in maximal oxygen uptake). In studies involving children and adolescents, these problems are compounded because interventions must be minimally invasive, ethically acceptable, and agreeable to both the subjects and their guardians.

$\dot{V}O_2max$	Maximal oxygen uptake
DLW	Double-labeled water
TEE	Total energy expenditure
MRI	Magnetic resonance imaging
BMI	Body mass index

In this study we hypothesized that even a brief (5-week) program of endurance exercise training would result in quantifiable structural and functional adaptations in female adolescents. The intervention was assessed by (1) comparison of total energy expenditure between groups with the doubly labeled water technique, (2) determination of changes in thigh muscle volume by magnetic resonance imaging, and (3) determination of changes in $\dot{V}O_2max$ by means of respiratory gas exchange responses.

METHODS

Protocol. Forty-four healthy adolescent girls volunteered for the study. The participants were all students at Torrance High School (Torrance, Calif.) and enrolled in an anatomy class during the summer of 1995 (July to August) with class hours from 8 AM to 12:30 PM. The ethnic configuration of the group was as follows: 68.4% Asian subjects, 20.4% white subjects, and 11.4% Hispanic subjects. No attempt was made to recruit subjects who participated in competitive extramural athletic programs. The study was designed to examine late pubertal subjects with an age range of 15 to 17 years. Measurements of height, weight, and body mass index (weight/height^2, an indirect estimate of lean body mass[6]) were obtained according to standard techniques. At each time point the mean of three separate measurements was used for height and weight. Assessment of pubertal status was determined by history, examination, or both in all subjects; all subjects were found to be at Tanner stage V.

Subjects were randomly assigned to control (n = 22) and training groups (n = 22). All subjects participated in the 2-hour daily teaching program. During the remaining time 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 activities, about 50% involved running, 25% team sports, and 25% aerobic dance. Training was directed by a member of the Torrance High School faculty. 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 in either the control or training groups. Staff members involved in taking anthropometric measurements (e.g., height, weight and BMI), performing total energy expenditure and magnetic resonance imaging studies, and $\dot{V}O_2$ max tests were unaware of the subjects' group status. The study was approved by the institutional human subject review board, and informed consent was obtained from the subjects and their parents or guardians.

Measurement of total energy expenditure. The DLW technique was used to measure TEE for a 10-day period beginning on week 3 of the protocol. Ideally, preintervention and postintervention measurements of TEE with the DLW would have been performed; however, this was prohibited by the high cost of $H_2{}^{18}O$. Subjects from the control and training groups (n = 9 each) were randomly selected for TEE measurements on the basis of a power analysis.

After a baseline urine sample was obtained, each subject was given a standard oral dose of DLW. A standard dose of 25 ml of a 1:1 mixture of ^2H_2O and $H_2{}^{18}O$ (99% enriched from Isotec Inc., Williamsburg, Ohio) was given to minimize calculation error. The dose is calculated to provide an average of 0.22 gm/kg of ^2H_2O or $H_2{}^{18}O$ with a range of 0.15 to 0.29 gm/kg. A urine sample was obtained 2 hours later, and then daily for the next 10 days. Oxygen and hydrogen isotopic ratios were measured by standard techniques with a model Delta-S gas isotope–ratio mass spectrometer (Finnigan, San Jose, Calif.). The isotope ratio data were analyzed by linear regression analysis after log transformation. Converting carbon dioxide production rate to TEE was done according to methods recommended by the International Dietary Energy Consultancy Group in 1990.[7, 8]

Magnetic resonance imaging of thigh musculature.

Table. Height, weight, and BMI in control and training group subjects

	Height (cm)	Weight (kg)	BMI (kg/m²)
Control group			
Before	159.5 ± 1.0 (151-170)	56.4 ± 2.6 (42.3-87.3)	22.1 ± 0.9 (15.3-33.3)
After	160.1 ± 1.0 (151-170)	56.3 ± 2.7 (41.3-89.1)	21.9 ± 1.0 (15.5-34.0)
Training group			
Before	161.9 ± 1.0 (154-172)	60.3 ± 2.1 (43.6-83.6)	23.0 ± 0.7 (17.1-29.0)
After	162.1 ± 1.0 (155-173)	60.7 ± 2.1 (44.1-84.1)	23.1 ± 0.7 (17.0-29.3)

Data presented are mean ± SEM and range.

Studies of thigh muscle volume were done before and immediately after the 5-week protocol in all 44 subjects. We chose to examine the musculature of the right thigh because these muscles would be largely involved in the endurance-type training program as described above. Magnetic resonance imaging has been used previously to assess the effects of training on muscle groups.[9, 10]

A 1.5 tesla whole-body MRI system was used (General Electric Medical Systems, Waukesha, Wis.). A body coil was used for both signal detection and for radiofrequency transmission for imaging. The subject was positioned with the lower extremities at the isocenter of the magnet bore. Pilot image coronal slices of the right thigh were obtained to select an image that included the distal femur. Twelve axial slices from above the knee to below the femoral neck were obtained. These axial slices were 20 mm thick with no gap and were obtained with a T1-weighted sequence with a time to echo of 12 msec and repetition time of 400 msec. The matrix was 192 × 256 with two acquisitions at each phase encode step.

The thigh muscle cross-sectional areas of consecutive 2 cm slices were easily recognizable and measured with computerized planimetry. The volume (in cubic centimeters) of each slice was estimated as the cross-sectional area (in centimeters squared) · 2 cm. These calculations then were added to calculate the muscle volume. Roman et al.[10] recently validated a similar approach with cadaver limbs that showed that MRI estimates of muscle volume were consistently only 3.7% greater than volumes obtained by water displacement.

Measurement of maximal oxygen uptake. Studies of $\dot{V}O_2max$ were done before and immediately after the 5-week protocol in all 44 subjects. Each subject performed a ramp-type progressive exercise test on a cycle ergometer so that the subject exercised to the limit of her tolerance. Gas exchange was measured breath by breath,[11] and the $\dot{V}O_2max$ was determined as previously described in children and adolescents.[12]

Comparison of maximal oxygen uptake with previous studies. We compared the $\dot{V}O_2max$ data obtained from this study population with the following data: (1) previous values obtained in our laboratory in 1984[12] of children and adolescents from the same geographic area who were predominantly white subjects (the different ethnicity largely reflects demographic changes that have occurred in this community during the past 10 years), and (2) previous values obtained by Åstrand[13] for female Scandinavians in 1952.

Relationship between maximal oxygen uptake and muscle volume. The relationship between $\dot{V}O_2max$ and muscle volume was assessed in two ways: first, by a cross-sectional analysis that used the values obtained from all 44 girls at the beginning of the study. Linear regression was used to calculate the correlation coefficient and the regression slope. Second, at the end of the study, we assessed the effect of training on $\dot{V}O_2max$ and muscle volume in each individual by calculating the ratio of the training-induced change in $\dot{V}O_2max$ to the training-induced change in muscle volume (i.e., $\Delta\dot{V}O_2max/\Delta$muscle volume).

Assessment of relative fitness. We estimated the fitness of each subject relative to the group as a whole. This was done by first calculating the linear regression between $\dot{V}O_2max$ and body weight from all 44 subjects at the beginning of the study. The resulting equation was: Predicted $\dot{V}O_2max$ (L/min) = 0.023 · Weight (kg) + 0.455 ($r = 0.68$, $p < 0.0005$). We then calculated the predicted $\dot{V}O_2max$ (by weight) for each subject and determined the percent of predicted (i.e., [observed $\dot{V}O_2max$/predicted] · 100) in each individual. To gauge the effect of the fitness level at the beginning of the study on the subsequent response to exercise training, we used linear regression to calculate the correlation between pretraining fitness and the percent change in $\dot{V}O_2max$ after the program. In addition, we determined the best-fit exponential decay between these variables with the statistical software package BMDP.[14]

Statistical analysis. An unpaired t test was used to compare (1) height, weight, and BMI between the control and training groups, and (2) TEE between control and training group subjects. Paired t tests were used to compare (1) preprotocol and postprotocol measurements of height, weight, BMI, thigh muscle volume, and $\dot{V}O_2max$; (2) the percent increase in $\dot{V}O_2max$ with the percent increase in muscle volume in each subject of the training group; and (3) the observed $\dot{V}O_2max$ with predicted values on the basis of Ås-

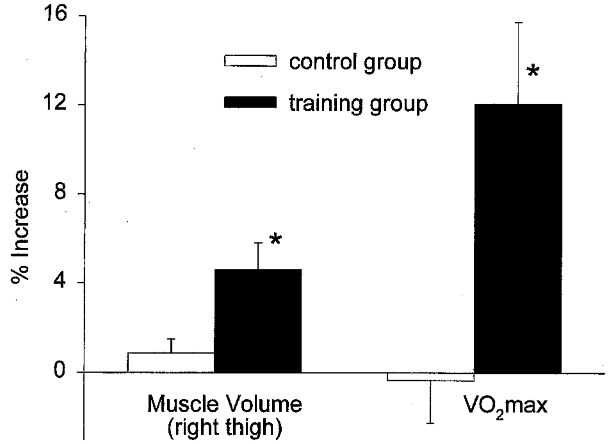

Fig. 1. Percent changes in $\dot{V}O_2max$ and right thigh muscle volume in the control and training group subjects. Training led to significant increases in these variables, but there was no change in the control group. The relative increase in $\dot{V}O_2max$ was significantly greater than was the increase in muscle volume (*asterisk, p* <0.05).

trand's data[13] and previous data collected in our laboratory.[12] As noted, standard linear regression techniques were used to correlate $\dot{V}O_2max$ with body weight, $\dot{V}O_2max$ with thigh muscle volume, and pretraining fitness with the percent increase in $\dot{V}O_2max$. In the training group, a single variable *t* test was used to compare the $\Delta\dot{V}O_2max/\Delta$muscle volume with the pretraining regression slope of $\dot{V}O_2max$ and muscle volume. Data are presented as the mean ± SE.

RESULTS

Height, weight, and body mass index. There were no significant differences in height, weight, and BMI between the groups before the course. There were no significant changes in height, weight, and BMI after the intervention in either group (Table).

Total energy expenditure. There was no significant difference in weight between the control and training subjects who participated in the DLW measurements. Total energy expenditure was 15.3% less in the control group (1815 ± 95 kcal/day) compared with the training group (2092 ± 124 kcal/day, p <0.0002).

Thigh muscle volume. There were no significant differences in thigh muscle volumes between the groups before the program (970 ± 19 cm^3 vs 987 ± 30 cm^3 in control and training groups, respectively). There were no significant changes in thigh muscle volume in the control group after the course (974 ± 39 cm^3). In contrast, there was a remarkable increase in thigh muscle volume in the training group subjects (21 of 22 participants, 1028 ± 31 cm^3, p <0.0002).

Maximal oxygen uptake. Before random assignment to the study groups, the study subjects were heterogeneous in fitness with $\dot{V}O_2max$ per kilogram of body weight ranging from fit (43 ml O_2/min per kilogram) to sedentary (21 ml O_2/min per kilogram). There were no significant differences in $\dot{V}O_2max$ between training group and control subjects before the course (1480 ± 60 ml/min vs 1570 ± 70 ml/min, respectively). There was no significant increase in $\dot{V}O_2max$ in the control subjects after the course, whereas $\dot{V}O_2max$ increased significantly in the training group subjects to 1630 ± 50 ml/min, p <0.004. The percent increase in $\dot{V}O_2max$ was about three times as great as the percent increase in muscle mass (p <0.05, Fig. 1).

There were no apparent differences in $\dot{V}O_2max$ between the 44 female adolescents in this study and results obtained in our laboratory more than 10 years ago in a geographically similar, but ethnically distinct, sample population. By contrast, the adolescents in this study appeared to have substantially lower $\dot{V}O_2max$ than did the subjects studied by Åstrand[13] in 1952 ($\dot{V}O_2max$ was 61% ± 9.5% predicted based on Åstrand's data, p <0.001; Fig. 2).

Linear regression analysis revealed a strong correlation between $\dot{V}O_2max$ and thigh muscle volume among the 44 subjects before the intervention (Fig. 3). The regression slope was 1.42 ± 0.20 ml O_2/min per cubic centimeter muscle volume. In contrast, as a consequence of the exercise intervention in the 22 members of the training group, we found in each individual a much greater increase in $\dot{V}O_2max$ per change in muscle volume (mean 4.78 ± 1.46 ml O_2/min per cubic centimeter increase in muscle volume, p <0.03). Finally, there was a significant negative correlation between fitness before the intervention and the training-induced increase in $\dot{V}O_2max$ ($r = -0.68$, p <0.0003; Fig. 4).

DISCUSSION

We designed a prospective, exercise training study for female adolescents (ranging in fitness from well-trained to sedentary) that was integrated into the summer curriculum of a local high school. All 44 subjects completed the program, and there were no injuries. The TEE was significantly greater (15.3%) in the training group compared with the control subjects. No structural or functional adaptations were observed in the control group. In contrast, training led to a 4.3% increase in thigh muscle mass and a 12.1% increase in $\dot{V}O_2max$. The training effect was most pronounced in the subjects who were least fit at the start of the program.

Interpretation of the few previous studies on endurance training in female adolescents has been confounded by a number of factors including imprecise assessments of energy expenditure and of the structural and cardiorespiratory components of the adaptive response.[15] These were addressed in our study by use of the DLW technique to assess TEE, MRI for muscle volume, and breath-to-breath measurements of respiratory gas exchange.

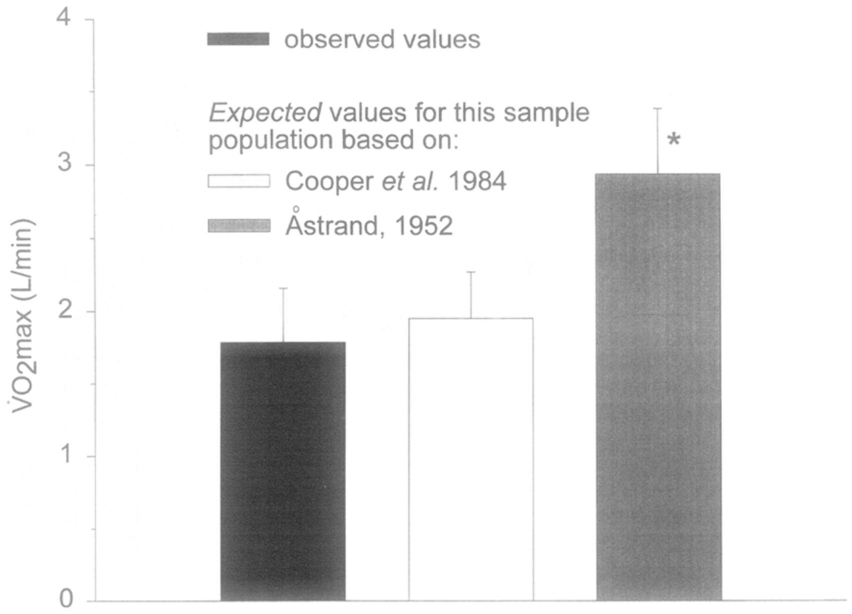

Fig. 2. Comparison of $\dot{V}O_2max$ values obtained in this study with (1) values predicted from the data of Cooper et al.[12] in this laboratory about 10 years ago, and (2) values predicted from the data of Åstrand[13] from female Scandinavians in the early 1950s. Although there were no differences between the study population and those obtained earlier in this laboratory, fitness was substantially reduced compared with Åstrand's data (*asterisk, p* <0.05).

The decline in physical fitness that occurs among high school–aged girls in this country is well documented[4] and is probably related to reduced physical activity known to occur in this group between the ninth and twelfth grades. Our data (Fig. 2) suggest that levels of fitness in adolescent girls have remained low in our own community over more than a 10-year interval and despite a major shift in ethnic background. Why the Scandinavian girls studied by Åstrand in 1952 seemed to have better fitness compared with Southern California girls is not known. Although issues of subject selection and other methodologic factors could have been factors in the discrepancy, it is noteworthy that we found no difference in $\dot{V}O_2max$ between the male subjects studied previously in our laboratory and the male subjects studied by Åstrand.[13] One can speculate that activities of daily living in Scandinavia in the 1950s likely required more physical effort than does the automobile-oriented culture of Southern California in the 1990s.

Physical inactivity is worrisome in any population, but the finding is of particular concern in high school girls for a number of reasons. First, it is precisely during the peripubertal stage that the level of physical activity profoundly influences peak bone mass in women.[16] Reduced peak bone mass (reached in women in their mid- to late-20s) is associated with increased incidence of osteoporosis later in life.[17] Second, although $\dot{V}O_2max$ per body weight remains relatively constant in male subjects throughout adolescence, it

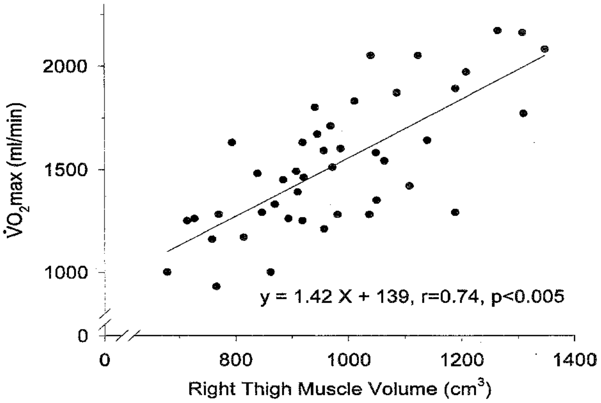

Fig. 3. The relationship between $\dot{V}O_2max$ and right thigh muscle volume in 44 healthy female adolescents before the subjects were randomly assigned to control and training groups. There was a strong correlation between these variables. As shown by the slope of the linear regression, each cubic centimeter increase in muscle volume was accompanied by a 1.42 ml O_2/min increase in $\dot{V}O_2max$.

continuously declines among female adolescents,[18] reflecting both an increase in body fat and reduced levels of physical activity. This likely contributes to the increased prevalence of obesity among American women that is now known to be associated with higher morbidity and mortality rates.[19] Finally, the effects of physically active lifestyles are not limited only to clinical or physiologic outcomes. Recent data

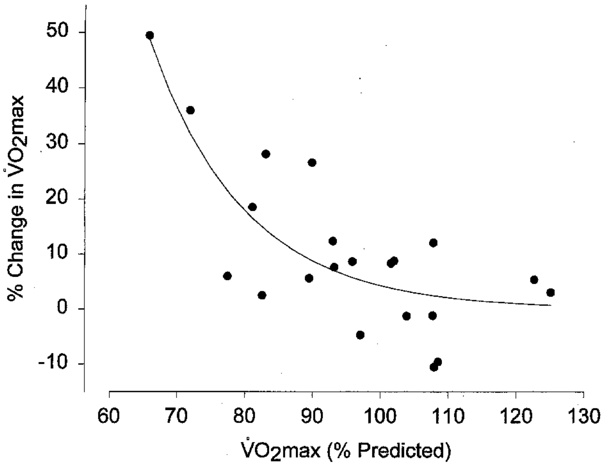

Fig. 4. Relationship between pretraining fitness (as % predicted $\dot{V}O_2$max) and the change in $\dot{V}O_2$max associated with the training intervention. Linear regression demonstrated a significant inverse correlation (see text) as did a first-order exponential decay *(solid line)*. The least fit subjects benefited most from the training.

suggest that physically active teenage girls are less likely than their less physically active cohorts to engage in high risk health behaviors such as cigarette smoking.[20]

There are very few studies in female adolescents in which both TEE by DLW and $\dot{V}O_2$max were measured in the same subjects. The TEE values in our study were lower than values also obtained by DLW from a sample of nine somewhat younger girls (age range 10 to 16 years) studied by Wong[21] (mean TEE 2322 ± 281 kcal/day) in whom $\dot{V}O_2$max was also measured. The higher energy expenditure was accompanied by substantially greater levels of fitness than we observed (mean $\dot{V}O_2$max in Wong's study, 43.9 ± 3.7 ml O_2/min per kilogram). These observations, combined with our own findings, suggest that daily energy expenditure may be a correlate of fitness in female adolescents.

Although TEE, functional fitness, and thigh muscle mass increased in the training subjects, body weight did not change. In fact, weight loss is not necessarily observed with successful training interventions even in obese subjects. For example, Blaak et al.[22] noted a 12% increase in TEE (also measured by DLW) in obese boys (age range 10 to 11 years) during a 4-week endurance training program that resulted in increased fitness but no change in body weight. Weight reduction was not a goal of our intervention, and we did not instruct the participants to alter their dietary patterns in any specific manner. We speculate that the likely beneficial effect of a physically active lifestyle established during childhood or adolescence is not necessarily immediate weight loss, but rather the prevention of weight gain known to occur in women throughout adolescence and adulthood.

Our data show that the effect of training could not be explained solely by the increase in thigh muscle mass. The training-induced percent increase in $\dot{V}O_2$max was 2.8 times the percent increase in muscle mass (Fig. 1). In addition, although the cross-sectional analysis showed that $\dot{V}O_2$max increased by 1.42 ml O_2/min per cubic centimeter of thigh muscle, training led to increases of more than 4.7 ml O_2/min per cubic centimeter of thigh muscle. Indeed, in addition to muscle hypertrophy the "training effect" is composed of a variety of size-*independent* factors including mitochondrial oxidative activity, muscle capillary density, and improved cardiorespiratory function.[23] These factors presumably accounted for the majority of the increase in our functional fitness (i.e., the $\dot{V}O_2$max) in this population of adolescent girls.

As recently outlined in *Great Transitions*, the concluding report on adolescent development sponsored by the Carnegie Corporation,[24] there are many obstacles for American adolescents to establish healthy lifestyles. The report emphasizes the role that schools ought to play as "health promoting environments" and that adolescents be provided, "... opportunities for exercise for all in the school community, not just varsity competition between different schools." This is of particular relevance to our study in girls, a population that until recently has been generally excluded even in the area of competitive sports. Moreover, many of the participants in our study suggested that despite the "required" status of physical education courses during the regular school year, it was quite easy to avoid any vigorous participation by using a variety of "legal," readily available excuses.

Therapeutic exercise training interventions have met with varying degrees of success, and it is clear that in any individual both fitness and levels of habitual physical activity have behavioral (possibly mutable) as well as genetic (immutable) determinants.[25] In our study, the largest relative increase in $\dot{V}O_2$max occurred in the least fit subjects, suggesting that the behavioral intervention (i.e., the exercise training) could significantly influence physiologic responses even in individuals not predisposed to physical fitness or participation in athletic activities for whatever reasons.

In addition, prospective studies of exercise training in children and adolescents have been hampered by lack of adequate control groups and poor motivation on the part of the subjects.[15] By working with local schools, we took advantage of existing facilities and an environment that was familiar to and relatively comfortable for high school girls. The integration of a summer course work in anatomy and physiology with a prospective study of physiologic responses to exercise generated interest and enthusiasm among all the participants. As a result, we could successfully implement a fairly rigorous training program with good compliance leading to substantial increases in TEE. Whether this degree of intervention is optimal is not known; however, the approach might serve as a model for further investigations or programs designed to increase levels of physical activity among children and adolescents.

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