

# Fitness is a Stronger Predictor of Fasting Insulin Levels than Fatness in Overweight Male Middle-School Children

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**Objective** We studied the relationship between % body fat (%BF), cardiovascular fitness (CVF), and insulin resistance (IR) in overweight middle-school children.

**Study design** Middle school children (n = 106, body mass index [BMI] > 95<sup>th</sup> percentile for age) underwent evaluation of body composition, maximal volume of oxygen utilization (Vo<sub>2</sub>) uptake/kg lean body mass (Vo<sub>2</sub>max/kgLBM), and fasting glucose and insulin (FI) concentrations and derived homeostasis model assessment index (HOMA<sub>IR</sub>).

**Results** Both %BF (r = .33, P < .001) and Vo<sub>2</sub>max/kgLBM (r = -0.42, P < .0001) were significantly correlated with FI. Bivariate regression analysis revealed %BF (P = .008 vs FI, P = .035 vs HOMA<sub>IR</sub>) and Vo<sub>2</sub>max/kgLBM (P < .001 vs FI, P = .009 vs HOMA<sub>IR</sub>) to be independent predictors of insulin sensitivity. In males, Vo<sub>2</sub>max/kgLBM was a better predictor of FI and HOMA<sub>IR</sub> than %BF.

**Conclusions** In obese middle-school children, both %BF and Vo<sub>2</sub>max/kgLBM are independent predictors of FI levels. The relationship between CVF and FI levels was significant in both sexes but was particularly profound and stronger than %BF in males. Efforts to reduce risk of type 2 diabetes mellitus in an increasingly obese child population should include exercise intervention sustained enough to improve CVF. (*J Pediatr* 2007;150:383-7)

An increasingly pervasive environment of reduced physical activity coupled with easy access to calories is spawning an epidemic of poor cardiovascular fitness (CVF), obesity, insulin resistance (IR), type 2 diabetes mellitus, blood lipid abnormalities, and hypertension in our youth.<sup>1,2</sup> Studies in adults have shown IR to be an independent predictor for the development of hypertension, coronary heart disease, stroke, cancer, and type 2 diabetes, and greater insulin sensitivity a protective factor against all of these clinical events.<sup>3</sup> These data serve as a strong stimulus to devise effective public health strategies to improve insulin sensitivity in children and adolescents. The degree to which such a strategy should emphasize improving fitness or reducing fatness in children remains unresolved.

Although obesity clearly increases the risk of insulin resistance, type 2 diabetes mellitus, and other cardiovascular morbidities,<sup>4,5</sup> it has been shown in adults that level of fitness is a more accurate predictor of cardiovascular and all-cause death than weight status.<sup>5,6</sup> It is presumed that the beneficial effect of improved CVF on insulin sensitivity reflect combined effects of increased lean mass and reduced fat mass<sup>7</sup>; however, physical activity alters insulin sensitivity independent of changes in weight and body composition in adults<sup>8</sup> and in children.<sup>9,10</sup> If CVF is shown to have a more profound effect on insulin sensitivity than percent body fat, efforts to improve insulin sensitivity in children may be best focused on increasing physical activity rather than simply restricting calories to achieve weight control.<sup>11</sup> In this study, we report the relationships between insulin sensitivity (assessed by fasting insulin [FI] levels and HOMA<sub>IR</sub>), CVF (measured by maximal oxygen uptake/kilogram lean body mass [LBM]), and percent body fat (measured by dual-energy x-ray absorptiometry [DXA]) in a cohort of overweight middle school children.

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%BF	Percent body fat	HOMA <sub>IR</sub>	Homeostasis model assessment index
BMI	Body mass index	IR	Insulin resistance
CVF	Cardiovascular fitness	LBM	Lean body mass
DXA	Dual-energy x-ray absorptiometry	Vo <sub>2</sub> max	Maximal oxygen consumption
FI	Fasting insulin		

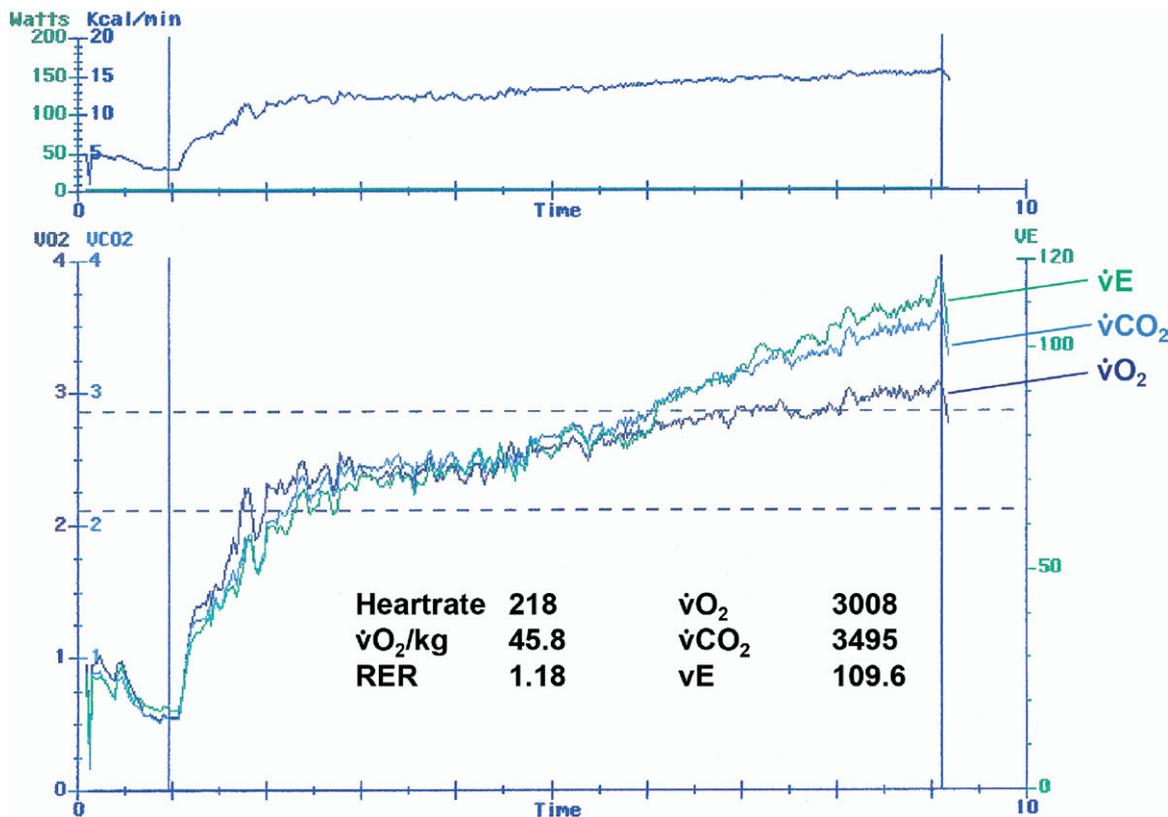


Figure 1. Graphical representation of maximal exercise testing in child demonstrates successful fulfillment of criteria for  $VO_{2max}$  measurement.

## METHODS

Children ( $n = 106$ ) at a single middle school with a body mass index above the 95th percentile for age participated in this study. Over a 24-month period, each participant underwent testing at the University of Wisconsin Exercise Science Laboratory before and at the conclusion of the 9-month school year during a single visit after an overnight fast, supervised by the same investigators. The procedures were approved by the Human Subjects Committee, and informed written consent was obtained before initiating the testing protocol. Testing included a physical examination, blood work to determine fasting glucose and FI levels, baseline body composition, and CVF assessment before beginning the program. Subjects with evidence for glucose intolerance or overt type 2 diabetes were excluded. Height was measured on a wall-mounted stadiometer to the nearest 0.5 cm. Weight was measured on a calibrated beam balance platform scale to the nearest 0.1 kg.

Percent body fat and LBM were measured by DXA. Whole body scans were performed with the Norland XR-36 whole body bone densitometer (Norland Corporation, Ft. Atkinson, WI) and tissue masses were analyzed with software version 3.7.4/2.1.0. Each scan session was preceded by a calibration routine with multiple quality control phantoms that simulate soft tissue and bone. Based on 18 scans of 6 subjects with the XR-36 whole body procedures, the total body coefficients of variation are as follow: soft tissue mass

0.2%, total body mass 0.2%, lean body mass 1.0%, fat mass 2.5%, percent fat 2.4%, and total BMC 0.9%.<sup>12</sup>

Children underwent a 4-minute submaximal treadmill walk test at 5% grade and subsequent measurement of maximal oxygen consumption ( $VO_{2max}$ ) performed by open circuit spirometry with a progressive treadmill walking protocol to volitional fatigue with a Medical Graphics CPX-D (St. Paul, MN). Specifically, the speed of the treadmill is set initially per the subject's comfort, starting at 0% grade and increasing 2% every minute. Requirements to strictly define whether subjects reached their maximal oxygen consumption by this protocol included at least 2 of the following 4 criteria: (1) visible evidence of exhaustion; (2) maximal heart rate  $>200$  beats/min; (3) respiratory exchange ratio ( $VCO_2/VO_2$ )  $>1.0$ ; and (4) a plateau in oxygen consumption (Fig 1). The plateau in oxygen consumption was defined as a change of  $< 2$  mL/kg/min in  $O_2$  consumptions over the last 60 seconds of the test. All subjects tested to exhaustion, 98.5% had respiratory exchange ratio (RER)  $> 1.0$  and fulfilled 2 criteria, and 78% of participants achieved 3 or all 4 of the criteria. Analysis of LBM by DXA was used to calculate  $VO_{2max}/\text{kgLBM}$ . Since the major influence of body weight on  $VO_{2max}$  is explained by fat free mass (FFM) and fat mass itself has minimal effect on  $VO_{2max}$ , CVF was expressed as  $VO_{2max}/\text{kgLBM}$ .

A 10-mL fasting blood sample for insulin and glucose was obtained from an antecubital vein. Samples were separated by low-speed centrifugation 4000g for 10 minutes.

**Table. Study subject characteristics (n = 106 subjects)**

	Total	Males	Females	P value
Sex	106	48 (45%)	58 (55%)	.943*
Age (years)	12.8 ± 1.4	12.7 ± 0.9	12.9 ± 1.7	.731†
BMI	30.8 ± 5.9	31.7 ± 6.9	30.1 ± 5.0	.237†
VO <sub>2max</sub> /kgLBM (mL/kg/min)	64.8 ± 21.8	68.3 ± 16.4	62.0 ± 16.4	.251†
%Body fat	35.8 ± 5.5	36.0 ± 6.3	35.6 ± 4.9	.462†
LBM (kg)	51.33 ± 11.29	53.35 ± 19	49.66 ± 8.26	.336†
Fasting insulin (μIU/mL)	24.3 ± 16.1	23.9 ± 17.1	24.6 ± 15.4	.665†
Fasting glucose (mg/dL)	86.0 ± 6.6	85.9 ± 6.5	86.0 ± 6.8	.947†
HOMA <sub>IR</sub>	5.0 ± 2.5	4.9 ± 2.7	5.1 ± 2.3	.872†

\*Chi-square test.

†Wilcoxon rank sum test.

Fasting insulin concentration was determined with the chemiluminescent assay (Esoterix, Callabaras Hills, CA), and glucose concentration was determined by an enzymatic method (Beckman Diagnostics, Fullerton, CA). HOMA<sub>IR</sub> was calculated with the formula: Fasting insulin (uU/mL) × Fasting glucose (mmol/L)/22.5, assuming that normal young subjects have an insulin resistance of 1.0.<sup>13</sup>

During the 9-month school periods within the 24 month testing period, children participated in physical education classes 5 times every 2 weeks for a 45-minute class period. Three subjects dropped out from this study; 2 moved away during the school year, and one student stopped coming to school because of non-medical reasons. None of their data were included in these analyses, and none of these data were significantly different from the rest of the cohort at baseline. Tests were repeated in participating children at the conclusion of and beginning of each school year during the 24-month period, resulting in 222 observations in 106 different children.

### Statistical Methods

Body composition, cardiovascular fitness, and insulin sensitivity measurements were summarized by standard descriptive statistics in terms of means and standard deviations. The bivariate associations between body composition, cardiovascular fitness and fasting insulin concentration were examined with the Spearman rank correlation analysis. Univariate and multiple regression models with subject specific random effects were used to predict fasting insulin concentration from body composition and cardiovascular fitness measurements. Fasting insulin concentration was log-transformed in the regression analyses to meet the assumption of normality. Since the study design involved repeated tests in subjects, an autoregressive correlation structure was used to account for correlations between repeated measurements within a subject. Statistical inference was performed with maximum likelihood estimation. For the cross-validation studies, univariate regression models, with %BF or VO<sub>2max</sub>/kgLBM as predictor variables, were fitted using data from a subset of k (k = 50 and 100) randomly selected subjects.<sup>14,15</sup> The fasting insulin concentrations of the subjects/observations not selected for the regression analysis were compared to the predicted fasting

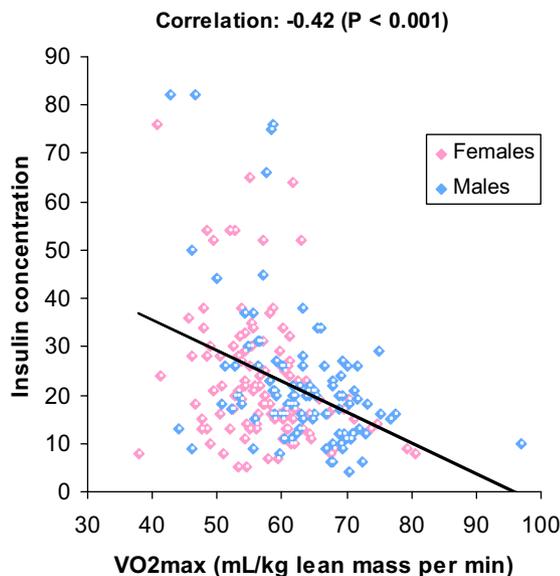
insulin concentration from the regression equations. This process was repeated 100 times, and the mean squared errors of the predicted versus observed fasting insulin concentrations were computed. A smaller mean squared error represents a better fit. All statistical analyses were performed with SAS software (version 8.2; SAS Institute, Cary, NC). All P values were 2-sided, and P values ≤ .05 were considered statistically significant.

### RESULTS

Patient characteristics are presented in the Table as mean ± SD. At study enrollment, mean age of the study participants was 12.8 ± 1.4 years, and 55% of the subjects were female. The mean body mass index (BMI) was 30.8 ± 5.9 (males 31.7 ± 6.9, females 30.1 ± 5.0; P = .237), and mean percent body fat (%BF) was 35.8 ± 5.5 (males 36.0 ± 6.3, females 35.6 ± 4.9; P = .462). Mean measurement of VO<sub>2max</sub>/kgLBM at study initiation was 62.0 ± 16.4 mL/kg/min for females, 68.3 ± 16.4 mL/kg/min for males (P = .215), and 64.8 ± 21.8 mL/kg/min for the combined group. Mean fasting insulin was 24.3 ± 16.1 μIU/mL (males 23.9 ± 17.1 μIU/mL, females 24.6 ± 15.4 μIU/mL, P = .665; normal values 4-24 μIU/mL). Mean HOMA<sub>IR</sub> was 5.1 ± 2.3 for females and 4.9 ± 2.7 for males (P = .872), and 5.0 ± 2.5 for the combined group.

For all subjects, univariate analysis revealed highly significant correlations between FI and VO<sub>2max</sub>/kgLBM (r = -0.42, P < .001; Fig 2) and FI and %BF (r = .33, P < .001; Fig 3), indicating that lower CVF and higher body fat had a highly significant association with higher fasting insulin levels. When HOMA<sub>IR</sub> was used as the indicator of insulin sensitivity, the correlation with VO<sub>2max</sub>/kgLBM remained significant (r = -0.27, P = .001), but the correlation with %BF was less robust (r = .18, P = .07).

Among all subjects, %BF and VO<sub>2max</sub>/kgLBM were strongly and negatively correlated with each other (r = -0.57 (P < .001). Consequently, bivariate regression analysis was performed to correct for this interaction. With this model, for males and females combined, both %BF (P = .008 vs FI, P = .035 vs HOMA<sub>IR</sub>) and VO<sub>2max</sub>/kgLBM (P < .001 vs FI, P = .009 vs HOMA<sub>IR</sub>) were significant independent predictors of



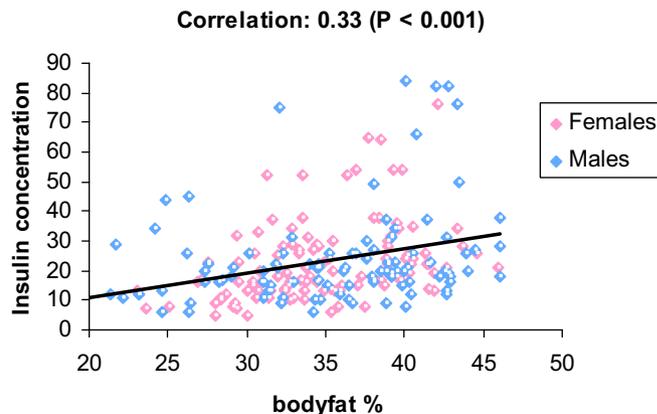
**Figure 2.** Relationship between fasting insulin levels ( $\mu\text{IU}/\text{mL}$ ) and cardiovascular fitness ( $\text{VO}_{2\text{max}}/\text{kgLBM}$  measured in  $\text{mL}/\text{kg}/\text{min}$ ) in overweight middle school children.

FI levels and insulin sensitivity by  $\text{HOMA}_{\text{IR}}$ . Thus in this group of male and female children combined,  $\text{VO}_{2\text{max}}/\text{kgLBM}$  and %BF were independent predictors of insulin sensitivity, and  $\text{VO}_{2\text{max}}/\text{kgLBM}$  was a better predictor of insulin sensitivity than %BF. Cross-validation studies confirmed a smaller mean squared error of predicted versus observed FI values for  $\text{VO}_{2\text{max}}/\text{kgLBM}$  ( $12.99 \mu\text{IU}/\text{mL}$ ) than for %BF ( $14.51 \mu\text{IU}/\text{mL}$ ), indicating that in this study group  $\text{VO}_{2\text{max}}/\text{kgLBM}$  was a better predictor of fasting insulin levels than %BF.

When males ( $n = 48$  subjects, 103 observations) and females ( $n = 58$  subjects, 119 observations) were analyzed separately, the sex-specific effect of %BF on FI ( $r = .37, P = .005$  for females;  $r = .30, P = .031$  for males) was significant for both sexes. In males,  $\text{VO}_{2\text{max}}/\text{kgLBM}$  had a clearly stronger relationship to FI ( $r = -0.52, P < .001$ ) and  $\text{HOMA}_{\text{IR}}$  ( $r = -0.417, P = .0032$ ) than %BF ( $r = .29$ ). For females,  $\text{VO}_{2\text{max}}/\text{kgLBM}$  retained a highly significant inverse correlation with FI ( $r = -0.38, P = .004$ ) equal in strength to that of %BF. However, there was no correlation between %BF and  $\text{HOMA}_{\text{IR}}$  ( $P = .2294$ ) or between  $\text{VO}_{2\text{max}}/\text{kgLBM}$  and  $\text{HOMA}_{\text{IR}}$  ( $P = .35$ ) in females. Further, when  $\text{VO}_{2\text{max}}$  over total body mass (rather than LBM) was used, multivariate regression analysis (ie, regression model of FI levels on  $\text{VO}_{2\text{max}}/\text{kg}$  total body mass or %BF) showed the association with FI to be significant for males ( $P = .001$ ) but not for females ( $P = .214$ ).

## DISCUSSION

This study shows that, like adults, cardiovascular fitness is an equal, perhaps even more important predictor of insulin sensitivity than is fatness in children. Differences in the interactions between CVF and %BF with sex have been noted



**Figure 3.** Relationship between fasting insulin levels ( $\mu\text{IU}/\text{mL}$ ) and percent body fat in overweight middle school children.

previously.<sup>16</sup> Our data suggest that the adverse effects on FI levels were most profound in males with high %BF and low CVF. In one prior study of older adolescents, when CVF (assessed by sub-maximal  $\text{VO}_2$  consumption) and %BF were analyzed together, for boys, both CVF and %BF retained predictive value for FI levels, whereas for girls, only %BF was significantly predictive of FI levels.<sup>17</sup> In our study of younger children, obese females also showed statistically more robust correlations between %BF and FI levels than between  $\text{VO}_{2\text{max}}/\text{kgLBM}$  and FI, and less robust (but still significant) correlations than males between  $\text{VO}_{2\text{max}}/\text{kgLBM}$  and FI. The preservation of CVF as a predictor of FI in females in our study could be related to the use of  $\text{VO}_{2\text{max}}/\text{kgLBM}$  as a more precise measurement of CVF than  $\text{VO}_{2\text{max}}/\text{kg}$  total body weight. Studies of children whose fitness was assessed by submaximal aerobic capacity show that the major influence of body weight on  $\text{VO}_{2\text{max}}$  is explained by LBM, that fat mass does not have any effect on  $\text{VO}_{2\text{max}}$ , and that fatness and  $\text{VO}_{2\text{max}}$  should be considered independent entities.<sup>18</sup> Expressing  $\text{VO}_{2\text{max}}$  per total body mass, rather than LBM, confounds the expression of fitness, especially when discussing metabolic activity, by incorporating fatness, and thereby relatively inert tissue, into the calculation of  $\text{VO}_{2\text{max}}$ . The slightly lower  $\text{VO}_{2\text{max}}/\text{kgLBM}$  in combination with a comparable %BF in females may explain why the association of FI with  $\text{VO}_{2\text{max}}/\text{kg}$  total body mass was not statistically significant, in contrast to males. And whereas there were relatively small differences in body composition (ie, either %BF, kg of LBM, or the ratio of %BF to LBM) between boys and girls in this group of prepubertal and early pubertal children, greater discrepancies in %BF with advancing puberty could be expected to further confound the expression of fitness as  $\text{VO}_{2\text{max}}/\text{total body mass}$ .

It is important to note particular characteristics of the study group and limitations of this report. First, even though all children had  $\text{BMI} > 95$ th percentile, only mild elevations in fasting insulin levels were observed. On the one hand, it is noteworthy that significant effects of CVF on FI were still evident within a fairly narrow range of observed FI levels. On the other hand, it is unknown whether these mild elevations

in FI levels can be confidently associated with the morbidities of hyperinsulinemia cited above. Second, the sample size of the study (106 subjects) was still fairly small, and we used repeated measurements in the same subjects. Finally, we relied primarily on FI levels and, to a lesser extent, calculated HOMA<sub>IR</sub> as indicators of insulin sensitivity. Although not as diagnostic for insulin resistance as frequently sampled testing of glucose and insulin, both fasting insulin levels and HOMA<sub>IR</sub> have been validated as strongly correlated with frequently sampled intravenous glucose tolerance test in obese children and adolescents.<sup>13</sup> The lower strength of correlation between %BF and VO<sub>2max</sub> and HOMA<sub>IR</sub> compared with FI levels in this study was unexpected. However, FI levels have been found to have greater precision than HOMA<sub>IR</sub> alone in some studies.<sup>19</sup>

Improved physical fitness is clearly effective in improving insulin sensitivity in adults,<sup>20</sup> but most adults do not achieve the Surgeon General's recommended 30 minutes of moderate physical activity on most days of the week.<sup>21</sup> Childhood is a critical period for nurturing lifetime activity behavior<sup>22,23</sup> and an attractive starting point for collaborative effort is the school setting, where both active and passive decisions regarding physical activity, food choices, and attendance can be reasonably controlled and programmatically altered. Still, skepticism about the importance and feasibility of changing fitness levels in children has limited acceptance and application of policies and programs required to achieve this goal. We and others have shown that school-based programs can significantly improve cardiovascular fitness and reduce fasting insulin levels in overweight children.<sup>14,24,25</sup>

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