

Correlation between Subfascial Fat Loss and Decrease in Serum Insulin Levels in Obese Teenagers during 4-Week Weight-Reduction Program

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ABSTRACT

Objective: To determine the association between reductions of fat from subcutaneous, intermuscular and subfascial compartment at thighs and improvement of serum insulin level.

Methods: A longitudinal, clinical intervention study of restricted energy with exercise was performed in 20 healthy, overweight teenagers (age: 10-15 y, BMI: 26-40 kg/m²). Fat distribution parameter (by dual-energy X-ray absorptiometry and computerized axial tomography at thigh), serum insulin, body weight were assessed at baseline and after 4 weeks.

Results: There are significant reductions in bodyweight, BMI, serum insulin, FGIR subfascial fat and intermuscular fat after 4 weeks. Loss of subfascial fat at thighs is significantly associated with decreasing levels of serum insulin in those who have initially abnormal high levels.

Conclusion: Losses of subfascial fat at thighs is associated with improvement of insulin levels in obese teenagers who are on 4 weeks of dietary restriction and moderate exercise.

Keywords: Computed tomography; obese teenager; subfascial fat; serum insulin; weight-reduction program

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There have been growing concerns that obesity is the most serious health problem at present. So far, it is one of the leading causes of death worldwide.¹ It is the major contributing factor to the increase of type II diabetes mellitus.² Now, childhood obesity has reached epidemic levels in the developed countries.³ More and more children are diagnosed of type II diabetes mellitus due to obesity.^{4,5} Also, the metabolic syndrome that is associated with obesity such as hyperinsulinemia, hyperlipidemia, impaired glucose metabolism and hypertension is quite common.^{6,7} The metabolic syndrome in adults is also more frequently found in obese children.⁸⁻¹⁰ This long-standing condition without appropriate management will result in an increase of mortality of these children in their early years.

Maintaining high levels of vigorous activity and low levels of TV viewing may help reduce the increasing prevalence of childhood obesity.¹¹ Physical activities attenuate this metabolic syndrome. Hence, they are recommended as an adjunct to a weight-reducing diet in the treatment of obesity.¹⁰ Previously we have demonstrated that a 4-week program of dietary restriction and intensive exercise during the summer as a day-time camp for obese children resulted in remarkable weight losses.¹² Most of the weight loss were from fat compartment, while maintaining their lean body masses. Therefore, the program was well accepted by the participants. Also, all participants complied with the program, and there were no complications during the course. We thus recommended this program in the summer vacation for starting of weight reduction for obese children.

Goodpaster and colleagues¹³ showed that during a moderate exercise, obese and sedentary men could increase the rates of fatty acid oxidation from nonplasma sources, presumably from intramuscular sources and

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reduce the rates of carbohydrate oxidation. Also earlier, they had showed that the subfascial fat and intramuscular fat at the thighs were good markers of insulin resistance in obesity and diabetes mellitus, although the intramuscular fat was much smaller than subcutaneous fat at the thigh.¹⁴

We, therefore, organized a 4-week program of weight reduction by dietary restriction with moderate exercise for moderate to severe cases of obesity in children. The main purpose of this study was to determine any association between reductions of fat from the intermuscular, subfascial and subcutaneous compartment at the thighs and the improvement of serum insulin level in obese teenagers.

MATERIALS AND MEHODS

Subjects

All subjects were healthy, except for their overweight (body mass index (BMI; in kg/m²) > 26. The children were between 10 and 15 years old. All subjects were thoroughly interviewed and examined to rule out any major medical disorders. Twenty healthy obese (eight males, twelve females) teenagers met all criteria and were recruited into the study. All methods and procedures for the study were approved by the Institutional Review Board of the Faculty of Medicine Siriraj Hospital, Mahidol University. At least one of parent of each participant provided a written informed consent for allowing his/her child to enter the study.

Program

The program was organized at the Clinical Research Center (CRC), Department of Pediatrics, Faculty of Medicine Siriraj Hospital, Mahidol University. A 4-week program, Monday through Friday, 8:00 a.m. till 16:00 p.m. was scheduled from March 30, 2003 to April 24, 2003. The program was run everyday, except on Wednesday, the daily schedule is as follows:

8:00-10:00 a.m.	: Nutrition education
10:00-Noon	: Swimming
Noon-1:00 p.m.	: Lunch
1:00-3:00 p.m.	: Group therapy & Psychotherapy
3:00-4:00 p.m.	: Aerobic dance

Every Wednesday, all participants joined a one-day tour around the Bangkok Metropolitan area. Besides sight-seeing, games were played by participants while on the bus and on the ground out-side the hospital. Each participant was assumed to have 2.5 to 3 hours of exercise daily which was estimated to consume of 500 kilocalories.

Education

Education in nutrition and exercise were held daily. Food exchange lists were taught at the beginning of the program. Practices for estimation of amount of energy and protein in each food were done from the second week. Benefits and techniques of exercise were likewise taught daily.

Group therapy

During the program, group therapy was set up for all participants by a team of pediatric psychiatrists and psychologists from the Department of Pediatrics, Faculty of Medicine Siriraj Hospital for at least two hours

daily, except Wednesday. The aim of this session was to change the participants' habits of eating and to improve their self-esteem.

Diets

During the first and second weeks of the program, all participants were supplied with food from our team for every meal, with a daily intake of 900 and 800 kcal respectively, and with protein intake of 50 grams daily. Participants were also advised not to take additional food at home. After the third week, they were advised to have dinner at home with 300 kcal, 20 grams protein for the meal; while during the day, they had breakfast and lunch at the hospital with a total of 500 kcal, 30 grams protein. Participants were taught to estimate the amount of energy and protein from the food given. They were advised to have breakfast of 200 kcal with 10 grams of protein at home during the fourth week. At lunch, all participants helped themselves to get food of 300 kcal, 20 grams of protein for their meals, under our close supervision. Then they were advised to have dinner at home of 300 kcal with 20 grams of protein for the meal.

Anthropometry

On the first and last day of the program, subjects' body weight, height, were measured. Their weight was measured with a Detecto weight balance. The height was also measured with height measurement of Detecto. The body weights were taken at 8:00 a.m. before meal everyday.

Dual-energy X-ray absorptiometry

Whole body fat mass and fat-free mass were assessed with dual-energy X-ray absorptiometry (DEXA) using Lunar Prodigy (software version 3.10.025, Lunar, Madison, WI) for each subject at day 0 and Day 28 of the program.

Computed tomography

Computed tomography of the right mid-thigh of each participant at the same position was done before and after the 4-week program. The intermuscular fat, subfascial fat and subcutaneous fat at and Day 0 and Day 28 of the program were analyzed with a computerized AUTOCAD 2004 program. Loading photos after scanning them in the program, we needed to indicate the units and decimal to measure as centimeters in order to be relevant to the units that received from the photos by film CT scan with the menu order for Format/Units. The length of the photos that come from scanning by the program, indicating from the photos by film CT scan, would consist of the distance for reference as centimeters so that it could match with the photos in the program and be able of find the size of different areas. The size and the cross-sectional areas of the subcataneous fat, subfascial fat, intermuscular fat, muscle, bone and skin would be calculated with the photo function in the program. Different sizes at the same position were measured by the program to compare the results with the previous films.

Metabolic testing

All tests are performed in the morning after a 12 h overnight fast. Fasting blood sugar, insulin level and lipid profiles were checked before and after the program.

TABLE 1. Clinical characteristics and baseline laboratory results of 20 subjects the program.

	Mean (SD)
Gender (M/F)	8/12
Age (y)	12.9 (1.7)
Weight (kg)	97.7 (16.4)
Height (cm)	157.6 (9.7)
BMI (kg/m ²)	31.8 (4.6)
Blood pressure (normal/abnormal)	8/12
Serum insulin (mmol/L)	29.7 (12.4)
Plasma glucose (mmol/L)	4.9 (0.4)
Cholesterol (mmol/L)	4.9 (1.1)
LDL cholesterol (mmol/L)	3.0 (0.8)
HDL cholesterol (mmol/L)	1.3 (0.4)
Triglyceride (mmol/L)	1.2 (0.6)
Oral glucose tolerance test (normal/abnormal)	8/12

Fasting glucose-to-insulin ratio (FGIR) was to measure insulin resistance, with insulin resistance diagnosed when values were < 7.¹⁵ Oral glucose tolerance test was done on each participant before entering the program.

Statistical analyses

Differences between means are evaluated using the paired Student t-test. Correlations between changing serum insulin levels after the program in those who have abnormally high initial values with improvement of bodyweight, BMI, body fat, percent of change in the subcutaneous, subfascial and intermuscular fat at the thighs after the program are evaluated by Spearman's correlation coefficient. The statistical significance level selected for all tests is $P < 0.05$.

RESULTS

All twenty participants could attend full course of the 4-week program. Nineteen of them had CT scan of right mid-thigh at Day 0 and Day 28. Only fifteen participants had completed results of pair serum insulin at Day 0 and Day 28. However, only ten of the fifteen had abnormal insulin levels initially. Then these ten pairs insulin levels were chosen to compare with other data. There were no adverse incidence occurred during

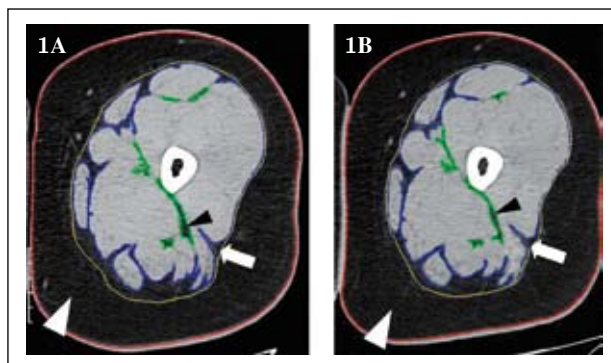


Fig 1A. A mid-thigh CAT film in a subject at Day 0, illustrating compartments of subcutaneous (SCF, large white arrowhead), subfascial (SFF, white arrow), and intermuscular (IMF, large black arrowhead) fat. All the lines were drawn by program AUTOCAD 4.

Fig 1B. Representative mid-thigh CAT film in the same subject at Day 28, showing decreases in SFF and IMF.

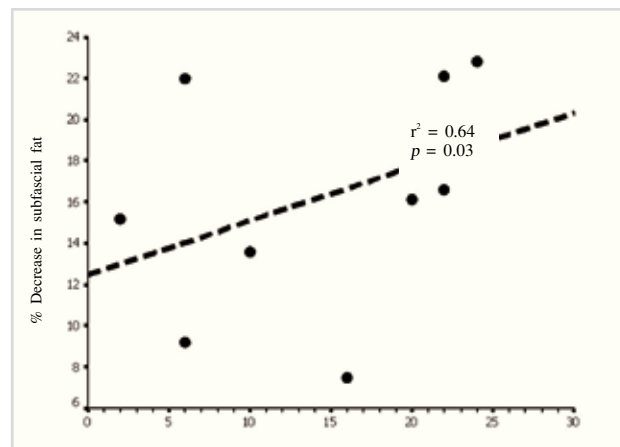


Fig 2. Association of decrease in percents of subfascial fat with decrease in insulin level.

the 4-week weight-reduction program. Parents and participants were very satisfied with the program. However, some parents reported of low compliances in their children with the food restriction during at home. However, they could completely attend the program.

Table 1 showed clinical characteristics and some screening laboratory results of twenty participants before entering the program. There were twenty teenagers, eight were male and twelve were female, participating in this weight- reduction program. Their age ranges were from 10 to 15 years. Their body mass index (BMI) ranged from 26 to 40. Twelve of them had hypertension, sixteen were considered hyperinsulinemia (insulin level > 15 mmol/L), and twelve had abnormal glucose tolerance tests.

Fig 1A and Fig 1B were CT scans of mid-thigh of a participant at Day 0 and Day 28, respectively. Fig 1A illustrates compartments of subcutaneous (SCF, large white arrowhead) subfascial (SFF, white arrow), and intermuscular (IMF, large black arrowhead) fat. All the lines were drawn by AUTOCAD 4 program. Figure 1B showed some decreases in SFF and IMF.

Table 2 compared the clinical outcomes of the participants between values of Day 0 and those of Day 28. There were three participants who had stable bodyweights throughout the program which were marked as low compliances with dietary restriction. The others had weight reduction ranging from one to five kilograms. Their BMI reductions ranged from 0.1 to 2.5 kg/m². At Day 28 of the program, bodyweights, BMI, serum insulin levels, fasting glucose-to-insulin ratio (FGIR), the subfascial fat and intermuscular fat at the right mid-thighs and total body fat, were significantly reduced compared with the entering values. Only the values of the subcutaneous fat of the right mid-thighs of Day 0 and Day 28 were not significantly different. Although there were improvements of serum insulin levels, the mean values of FGIR still were lower than seven which indicated insulin resistance.

Studies associations of reductions in those who had abnormally high initial insulin levels (insulin level > 15 mmol/L) with changes in bodyweights, body mass indexes, body fat, percent changes of fat in the subcutaneous, subfascial and intermuscular compartment after the program are shown in Table 3. There was an association only in the subfascial fat compartment.

Fig 2 depicts the association of changes in percents

TABLE 2. Clinical outcomes from pre-and post program of 20 participants between pre- and post 4-wk weight reduction program[†].

	Day		p-value
	0	28	
Weight (kg)	79.7 ± 16.4	76.9 ± 15.4	< 0.001
BMI (kg/m ²)	31.8 ± 4.6	30.7 ± 4.1	< 0.001
Serum insulin (mmol/L)	30.1 ± 10.8	18.8 ± 8.2	< 0.005
FGIR [‡]	4.1 ± 3.2	5.3 ± 3.4	< 0.001
Fat compartments of mid-thigh			
Subcutaneous fat (cm ²)	170.1 ± 51.6		
Subfascial fat (cm ²)	18.8 ± 5.0	160.1 ± 50.7	NS
Intermuscular fat (cm ²)	3.1 ± 2.4	15.6 ± 4.8	< 0.001
Total body fat (%)	45.5 ± 5.7	1.9 ± 1.7	< 0.001
		43.9 ± 5.5	< 0.05

[†]Means ± SD

[‡]FGIR, fasting glucose-to-insulin ratio

NS: not significant

of subfascial fat with serum insulin levels.

DISCUSSION

A 4-week weight reduction program during the summer is very applicable. Not only that dietary restriction and exercise were provided to the participants, but also they were taught nutritional values of foods and food exchange lists during the weight reduction program. Previously we have demonstrated that the participants had significantly better knowledge about nutrition after the program.¹⁶ Moreover, we also showed that the participants' lung functions were much improved as well as much reduction of body fat after the program.¹⁷ Also, during the 4-week weight reduction program, the participants received two sessions of aerobic exercise, one in the morning and the other in the afternoon. Each session was expected to burn up 300-400 kilocalories of each participant. The levels of exercise in each session and the numbers of days of exercise are significant to the reduction of subfascial and intramuscular fat. From our program, two sessions per day of moderate exercise and for four weeks were long enough to improve insulin levels in the participants. It appeared that peer-to-peer induction in weight reduction was also efficient during the program. Those who had experiences in group exercises before joining the weight reduction training program had a better chance of success in the program.¹⁸ The reduced fat deposition at thighs in our study is the same as Ryan's study¹⁹, although both were

TABLE 3. Association of reductions in serum insulin level with changes in body weights, BMI, body fat, percent changes of subcutaneous (SCF), subfascial (SFF) and intermuscular (IMF) fat after the program.

	Insulin levels	
	r	p-value
Body weight	-0.32	0.34
BMI	-0.30	0.37
Body fat	-0.35	0.29
SCF	0.18	0.60
SFF	0.64	0.03*
IMF	0.12	0.73

*Significant difference

done in different age groups.

From CT of the thigh, fat accumulation within the thigh is subcutaneous (SCF), that accumulates beneath the fascia lata (subfascial, SFF) and within the muscle (intramuscular, IMF). It was shown that increased muscle lipid content was associated with the severity of insulin resistance.^{20,21} Although the subfascial and inter-muscular fat are much smaller than the subcutaneous, they are good markers for insulin resistance in obesity and diabetes mellitus.¹⁴ With the assistance of computer program (AUTOCAD 4), it is very reliable to measure any minimal changes in three fat-compartmental changes of CT scan at midthighs. The increases in SFF

and IMF were shown to be associated with increased total and central obesity and reduced physical fitness, independent of age.²² Exercise increases the capacity of muscle for lipid utilization²³, which may partly explain the significant loss of SFF in our study.

Accumulation of fat in several internal organs in obese subjects had been suggested to be associated with insulin resistance. However, accumulation of fat in the pancreas does not impair insulin secretion in nondiabetic teenagers.²⁴ Bonen et al.²⁵ showed that triacylglycerol accumulation in human obesity and type 2 diabetes was associated with increased rating of skeletal muscle fatty acid transport and contributed to insulin resistance. Insulin resistance and hyperinsulinemia were associated with hypoadiponectemia in obesity and type 2 diabetes.²⁶ Whereas Weiss et al.²⁷ showed that low adiponectin levels in adolescent obesity is a marker of increased intramyocellular lipid accumulation. However, the relation between adiponectin level and amount of intermuscular fat at thigh has not been demonstrated. Thus, it is still dubious that improvement of adiponectin level should inversely be related with of subfascial fat at the thigh.

There may be a normal limit of subfascial fat in each subject. The value above that limit will result in hyperinsulinemia. When the amount of subfascial fat is reduced to below normal limit, the serum insulin level will decrease to normal value. Although there is no association between intercellular fat and insulin level, intercellular fat might play some roles contributing to hyperinsulinemia. Further researches to define the normal fat content in the subfascial compartments in each individual are needed in order to predict when insulin resistance will occur. Thus, in this study, we have shown that reducing subfascial fat at the mid-thigh area is correlated with the improvement of hyperinsulinemia. Also, the estimation of subfascial fat at the mid-thigh area by CT scan by an appropriate program is accurate. We recommend the use of this method to follow up the improvement of fat accumulation, instead of the total body fat.

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