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Impact of ER gene polymorphisms on overweight and obesity in Down Syndrome

Research Article

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Abstract: The impact of ER XbaI and PvuII α gene polymorphisms on overweight and obesity were studied in 77 subjects with Down Syndrome (DS), of which 32 were children (18 boys, 14 girls), mean age 8.7 ± 2.3 years, and 45 adolescents (28 boys, 17 girls) mean age 14 ± 2.5 years. Their lifestyle was compared to 40 healthy age-matched controls. DS subjects had significant lesser physical activity than controls ($p<0.05$) and a lower caloric intake than the recommended requirements, which was significantly lesser than controls ($p<0.05$). Body Mass Index (BMI), Arm Circumference (AC) and Triceps Skinfold Thickness (TST) were significantly higher in DS subjects than controls ($p<0.05$), while metabolic and cardiovascular parameters were not significantly different between the groups ($p>0.05$). The frequency of ER genotypes in DS subjects was compared with the healthy controls, finding that there was a high prevalence of XXER genotype in DS subjects. Children and adolescents with DS, lacking ER XbaI site, showed significantly higher BMI and body fat distribution than other XbaI genotypes. The lack of ER XbaI site can indicate added risk of obesity in DS. No differences in metabolic and cardiovascular parameters were observed among ER genotypes. However, childhood obesity is associated with increased cardiovascular risk.

Keywords: *Down Syndrome (DS) • Obesity • Overweight • XbaI ER polymorphism • PvuII ER polymorphism • Cardiovascular risk*

1. Introduction

People with Down Syndrome (DS) are frequently overweight or obese [1-3].

Like the general population, obese children and adolescents with DS have a greater tendency toward obesity in adulthood with increased risk of cardiovascular and metabolic diseases [4,5].

In the general population obesity has been related to many biological and environmental factors such as endocrine, metabolic disorders and unhealthy lifestyle. Recent studies have shown that several genetic mutations can also be associated with obesity [6]. It has been demonstrated that monogenic alterations with failure in serum leptin concentrations such as DG133 deletion, mutations in the melanocortin- 4 receptor gene (MC4R) and proopiomelanocortin (POMC) are associated with obesity [6].

Estrogen has effects on body fat distribution, a common risk factor of cardiovascular alterations in post-menopause. These effects are mediated by the estrogen receptor α gene (ER α), a transcription-dependent factor ligand with two important polymorphisms: PvuII which is located at 0.4 Kpb upstream of exon 2 and XbaI which is located in the intron, approximately 50bp from the PvuII polymorphic site [7]. It has been shown that ER α absence causes adipocyte hyperplasia and hypertrophy in white adipose tissue and is accompanied by alterations in glucose metabolism in male and female mice [8]. The association of polymorphisms in the ER α gene with obesity has been reported particularly in women [7,9].

Knowing something more about associations between ER gene polymorphisms and obesity could enhance current research in preventive treatment of the disorder, and help avoid the risk of cardiovascular and metabolic disease in the general population.

The study of associations between ER genotypes and obesity in DS is particularly interesting because of the high frequency of overweight individuals found with this genetic disease.

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The purpose of this study was to determine the frequency of various ER genotypes in a DS population compared to healthy controls, and its associations with obesity, and cardiovascular and metabolic disorders. Caloric intake and physical activity were also evaluated.

2. Material and Methods

This study included 77 out patients with DS from the Campania region in Italy, 32 of which were children (18 boys and 14 girls) mean age 8.7 ± 2.3 years, and 45 adolescents (28 boys and 17 girls) mean age 14 ± 2.5 years, considering that the onset of pubertal development was evaluated, for males, when a testicular volume was > 4 ml, and for females, the Tanner stage B2 [10]. Exclusion criteria included untreated heart disease before the age of one year, hypothyroidism or any chronic or intercurrent illness. Informed consent was obtained from parents. Procedures followed were in accordance with the ethical standards.

A dietary assessment was performed in DS subjects and 40 age-matched healthy controls of both sexes (15 children, 25 adolescents). Parents were instructed on measuring techniques and serving size, which they recorded for 2 weeks, also keeping track of meal or snack times, and the type of food and preparation methods using household measures. Determining caloric intake was performed with the assistance of a nutritionist as described by Sharav and Bowman [11]. The percentage of caloric intake in both, DS and controls, was compared to the recommended energetic requirement for height, because children with DS are usually short for age [11]. Parents were then requested to compile the Telama questionnaire for physical activity over a 2-week period [12]. They also recorded the participation in sports, such as swimming, skating and gymnastics. The daily number of hours that DS subjects spent in physical activity were estimated [11]. Using a 1 to 5 graded scale, the physical activity of DS subjects was compared to the 40 healthy age-matched controls for movement when playing and/or walking [11]. The score was reported for each patient and control by a same relative.

Anthropometric, metabolic and blood pressure values were evaluated in both DS subjects and controls.

For DS subjects the anthropometric variables were evaluated on the growth charts for DS from birth to 18 years [13].

Body mass index (BMI) was calculated as kg/m^2.

Body fat distribution was related to triceps skinfold thickness (TST) measured with Heberden skinfold callipers and arm circumference (AC).

Patients were considered overweight when their BMI was > 85th < 95th percentile. They were defined as obese when their BMI was ≥ 95th percentile.

Glucose metabolism was evaluated using a glucose oxidase method that measures fasting plasma glucose concentrations.

Fasting cholesterol, HDL-cholesterol and triglycerides were evaluated by enzymatic-colorimetrical method, LDL-cholesterol was determined by Friedewald's method [14].

Systolic and diastolic blood pressure (SBP and DBP) were evaluated by repeated measurements at the same time of day, using a mercury sphygmomanometer in the seated position. Children were considered hypertensive when values were > 95th percentile compared with percentile curves [15].

Genomic DNA was extracted from whole blood and analyzed for polymorphisms in the ER gene after amplification by PCR. Primers designed to amplify intragenic polymorphic PvuII and XbaI sites of the ER gene were the following: forward, 5-CTGCCACCCTATCTGTATCTTTTCCTATTCTCC-3'; and reverse, 5-TCTTTCTCTGCCACCCTGGCGTCG ATTATCTGA-3'[16]. Amplification conditions were as follows: 94 ºC for 5 min followed by 35 cycles at 94 ºC for 60 s, 61 ºC for 60 s, and 72 ºC for 90 s, and a terminal extension at 72 ºC for 10 min. After amplification, the PCR product (1.3 Kb) was digested with restriction endonucleases PvuII and XbaI and eletrophoresed in a 1.2% agarose gel. The relative genotypes were classified as XX homozygotes (absence of XbaI site), Xx heterozygotes and xx homozygotes (presence of XbaI site); PP homozygotes (absence of PvuII site), Pp heterozygotes and pp homozygotes (presence of PvuII site).

Children and adolescents with DS were categorized by XbaI and PvuII ER gene polymorphisms.

Statistical analysis for the comparison of data was performed using the dependent *t*-test.

Comparison of anthropometric, metabolic and blood pressure values between DS subjects and relative controls (40 subjects), as well as among DS subjects belonging to various XbaI and PvuII genetic groups was performed by analysis of variance (ANOVA).

The different frequency of ER genotypes in DS and in controls (15 children: 8M-7F; 25 teen-agers: 14M-11F) was evaluated by χ^2 test.

Significant values were set at $p \leq 0.05$.

The relation between XbaI and PvuII polymorphisms was examined.

Table 1. Characteristics of subjects with Down Syndrome Comparison with control subjects.

	CHILDREN		MALE TEEN-AGERS		FEMALE TEEN-AGERS	
	Down	Controls	Down	Controls	Down	Controls
Number of patients	32 (18M-14F)	15 (8M-7F)	(28M)	(14M)	(17F)	(11F)
Age (years)	8.4 ±1.7	9.1±1.9	14.9±1.2	14.1±1.5	14 ±1.5	13.7±1.8
BMI (Kg/ m^2)	21.2±3.4 (50^{th} – 85^{th} C)	17.9±1.76 (25^{th} – 75^{th} C)	23.7±2.8(75^{th} -85^{th} C)	19.98±0.88(25^{th} – 75^{th} C)	23.7 ±2.5(75^{th} -85^{th} C)	19.39±1.83(10^{th} – 75^{th} C)
AC (mm)	219±20 (50^{th} – 85^{th} C)	185±7.15 (25^{th} – 75^{th} C)	269±30 (75^{th} -85^{th} C)	232±10.5(25^{th} – 75^{th} C)	272 ±28(75^{th} -85^{th} C)	248±12(10^{th} – 75^{th} C)
TST (mm)	17.6±1.5 (50^{th} – 85^{th} C)	11.8±1.2 (25^{th} – 75^{th} C)	18.4±3.5 (75^{th} -85^{th} C)	11.9±2.2(25^{th} – 75^{th} C)	24.3 ±4.5(75^{th} -85^{th} C)	17.2±2.4(10^{th} – 75^{th} C)
TCHO (mg/dl) n.v. 50-220 mg/dl	164±16	162±15.7	163±23	164±22.5	160 ±20	162±18.9
HDL-CHO (mg/dl) n.v.	42.1±4.2	42.5±3.7	40.8 ±3.6	42±2.8	41.8 ±4.3	42±3.5
LDL-CHO (mg/dl) n.v.	114.1±18.4	112±19.1	112±12	110±13.2	108 ±20	110±18.5
TG (mg/dl) n.v. 50-200mg/dl	62.5±14	63.1±12	74±9.2	76±7.5	68.8 ±7.5	69±8.1
GLU (mg/dl) n.v. 70-115 mg/dl	85±10	87±7	90±15	94±10.5	92±12	90±15.2
SBP (mmHg)	105±10	102±7	115±10	110±10	115±8	110±8
DBP (mmHg)	68±8	67±6	72±5	70±5	70±5	68±5

BMI= Body Mass Index; AC = Arm Circumference ; TST= Triceps Skinfold Thickness;
TCHO= Total Cholesterol; HDL-CHO= High Density Lipoproteins – Cholesterol;
LDL-CHO = Low Density Lipoproteins – Cholesterol; TG = Triglycerides; GLU= Glucose;
SBP = Systolic Blood Pressure; DBP= Diastolic Blood Pressure.

BMI, AC, TST: children, male and female teen-ages vs controls: $p<0.05$
TCHO, HDL-CHO, TG, GLU: DS children, male and female teen-ages vs controls: $p>0.05$
SBP, DBP: DS children, male and female teen-agers vs controls: $p>0.05$

Table 2. Anthropometric, metabolic and blood pressure values in children with Down Syndrome belonging to various ER genotypes.

	XX	Xx	xx	PP	Pp	pp
Number of patients	12 (7M 5F)	16 (9M 7F)	4 (2M 2F)	6 (3M 3F)	16 (9M 7F)	10 (6M 4F)
Age	8.2 ± 1.7	8.9 ± 2.1	8.5 ± 1.9	7.9 ± 2.1	8.2 ± 1.7	8.4 ± 1.9
BMI (kg/m^2)	23.3 ± 1.5	20.4 ± 2.8	19.8 ± 3.4	22.1 ± 3.4	22.7 ± 2.8	22.4 ± 2.9
AC (mm)	245 ± 10.7	210 ± 15	195 ± 12.1	200 ± 15	215 ± 15	205± 10
TST (mm)	19.5 ± 2.2	16.2 ± 1.7	15.5 ± 1.9	17.2 ± 3.1	16.9 ± 2.7	16.5 ± 1.8
TCHO (mg/dl) Range 50-220 mg/dl	165 ± 22	155 ± 10	155 ± 15	158 ± 10	155 ± 12	158 ± 15
HDL-CHO (mg/dl) Range 35-75 mg/dl	38.5 ± 4.8	44.3 ± 5.4	45 ± 3.2	41 ± 6.5	43.6 ± 5.8	42.8 ± 4.3
LDL-CHO (mg/dl) Range 0-160mg/dl	116.6 ± 18.7	125.14 ± 26.4	92.98± 16.3	121.32 ± 9.1	119.21± 16.3	109.39 ± 27.5
TG (mg/dl) Range 50-200mg/dl	65 ± 10	60 ± 15	62 ± 10	61 ± 10	62 ± 12	60 ± 10
GLU (mg/dl) Range 70-115 mg/dl	85±10	80±7	82±5	90±5	82±8	85±10
SBP (mmHg)	105±10	105±7	105±8	105±10	105±8	105±9
DBP (mmHg)	70±6	68±5	67±3	66±6	66±6	68±8

BMI= Body Mass Index; AC= Arm Circumference; TST = Triceps Skinfold Thickness;
TCHO= Total Cholesterol ; HDL-CHO=High Density Lipoproteins- Cholesterol; LDL-CHO = Low Density Lipoproteins- Cholesterol;
TG = Triglycerides; GLU= Glucose;
SBP= Systolic Blood Pressure; DBP= Diastolic Blood Pressure;

BMI, AC, TST : XX vs Xx $p < 0.05$
XX vs xx $p<0.05$
Xx vs xx $p>0.05$
TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : XX vs Xx $p> 0.05$, XX vs xx $p> 0.05$, Xx vs xx, $p>0.05$
BMI, AC, TST, TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : PP vs Pp, PP vs pp, Pp vs pp, $p>0.05$

3. Results

Characteristics of children and adolescents with DS compared with controls are shown in Table 1.

Anthropometric values were in DS children between 50th and 85th percentile, in DS teenagers of both sexes between 75th and 85th percentile. Children and male teenagers of control groups showed anthropometric values between 25th and 75th percentile, female teenagers between 10th and 75th percentile.

Significant differences were observed in anthropometric measurements between DS subjects and relative controls (p <0.05).

Non significant differences of metabolic and blood pressure values were observed between DS subjects and controls (p>0.05) (Table 1).

Analysis of the energetic intake of the children with DS and controls showed that the DS subjects were receiving 78±7.5% of the recommended intake for height and the controls were receiving 85.2±12% intake for height (p<0.05).

Male teenagers were receiving 78.5±6.3 % of the recommended energetic requirements for height and controls were receiving 83±7% intake (p<0.05). Female teenagers were receiving 76±7% intake for height and controls 84±5% of the recommended requirement for height (p<0.05).

Analysis of the activity questionnaire, using the dependent t-test, showed that the DS subjects had significantly less physical activity than controls closest in age: children t = -7.8, p< 0.05; adolescent males t = -4.5, p< 0.05; adolescent females t = -2.6, p< 0.05.

Analysis of the frequency of XbaI ER genotypes, in DS subjects, showed 26 patients with XX, 41 with Xx and 10 with xx ER genotype (Tables 2-4). In controls 5 subjects had XX ER genotype, 29 Xx ER genotype, 6 xx ER genotype. Statistical analysis showed that the frequency of XX ER genotype was significantly higher in DS subjects than controls (χ^2 = 5.07, p<0.05), while non significant differences were observed among the other XbaI ER genotypes. (χ^2 = 3.29 and 0.00 respectively, p>0.05).

Frequency of PvuII ER genotypes in DS subjects are reported in Tables 2, 3 and 4.

In the control groups, 4 subjects had PP, 30 Pp and 6 pp ER genotype. Non significant differences of various PvuII genotypes were observed between DS subjects and controls. (χ^2 = 1.112, 3.65, 1.97 respectively, p>0.05).

Table 3. Anthropometric, metabolic and blood pressure values in female teen-agers with Down Syndrome belonging to various ER genotypes.

	XX	Xx	xx	PP	Pp	pp
Number of patients	6	8	3	4	9	4
Age (years)	14.1±1.2	15.2±1.8	14.1±1.2	14.8±1.5	14.8±1.5	15.1±1.6
BMI (kg/m^2)	28.6±1.8	24.5±1.9	19.4±2.3	23.8±1.9	24.5±1.2	24.2±2.2
AC (mm)	315±25	260±25	248±20	240±10	240±22	235±25
TST(mm)	30.8±2.1	24.5±1.8	18.2±2.5	24.8±3.1	22.5±3.8	20.6±3.9
TCHO (mg/dl) Range 50-220 mg/dl	170±25	155±20	150±15	165±15	158±20	160±20
HDL-CHO (mg/dl) Range 35-75 mg/dl	36.2±4.9	42.5±3.4	44.2±3.8	45.2±1.9	43.8±2.5	44.9± 3.2
LDL-CHO (mg/dl) Range 0-160mg/dl	115±13	106±12	103±12	112±18	102±11	107±17
TG (mg/dl) Range 50-200mg/dl	75.9±5.9	65.8±10.1	68.1±7.9	69.9±7.5	67.9±8.4	65.8±5.7
GLU (mg/dl) Range 70-115 mg/dl	94±10	90±10	85±5	95±8	94±10	90±10
SBP (mmHg)	115±7	115±8	113±6	115±8	115±7	113±6
DBP	69±4	71±4	70±5	70±5	68±3	72±3

BMI= Body Mass Index; AC= Arm Circumference; TST= Triceps Skinfold Thickness;
TCHO= Total Cholesterol; HDL-CHO= High Density Lipoproteins – Cholesterol; LDL-CHO = Low Density Lipoproteins – Cholesterol;
TG= Triglycerides; GLU= Glucose;
SBP= Systolic Blood Pressure; DBP= Diastolic Blood Pressure.
BMI, AC, TST: XX vs Xx p <0.05
XX vs xx p< 0.05
BMI, TST: Xx vs xx p< 0.05
AC: Xx vs xx p>0.05
TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : XX vs Xx p> 0.05, XX vs xx p> 0.05, Xx vs xx p>0.05
BMI, AC, TST, TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : PP vs Pp, PP vs pp, Pp vs pp, p>0.05

However, non significant differences were observed between genders in both DS subjects and controls (p>0.05).

Relationships of anthropometric measurements, metabolic levels and blood pressure values with XbaI and PvuII ER polymorphisms of children, adolescent males and females are shown in Tables 2-3 and 4. In children and adolescents of both sexes belonging to ER XX genotype, anthropometric measurements were significantly higher than other XbaI genotypes (p<0.05), while non-significant differences of serum metabolic parameters and blood pressure values were observed (p>0.05).

In adolescents of both sexes with ER XxbaI genotype, BMI was significantly higher than subjects belonging to ER xxbaI genotype (p<0.05). Adolescent girls with this ER genotype showed significantly higher TST than girls with ER xxbaI genotype (p<0.05).

Subjects with various PvuII genotypes showed non-significant differences in anthropometric, metabolic and blood pressure parameters.

All possible linkages between XbaI and PvuII polymorphisms were detected. Xx/Pp was the most frequent association (21 cases), followed by XX/pp (11 cases), XX/Pp (10 cases), Xx/PP (10 cases), Xx/pp (10 cases), xx/Pp (9 cases), XX/PP (5 cases), xx/pp (1 case).

Non significant differences of anthropometric, metabolic and blood pressure values were observed between DS subjects belonging to evaluated XbaI / PvuII associations (data not shown).

4. Discussion

Childhood obesity is a predictive index of adult disability and mortality because of its association with later cardiovascular and metabolic disease. Adipocytes increase in number especially during the first year of life, but this increase continues, although at a lower rate, throughout puberty, so that in adolescence, during weight reduction, the size, but not the number of adipocytes decreases. Obesity may be the result from increases in numbers or size of fat cells.

Hence, if obesity starts in the first years of life, it is likely to persist. Similarly, early over-nutrition may lead to rapid weight gain and to obesity. It has been shown that genetic and environmental factors are related to obesity. Genetic predisposition to obesity is the result of allele variability that influences energetic equilibrium. Furthermore, genetic factors intervening in metabolism and behaviour, influence energetic intake, physical activity and consequently, BMI variability.

Table 4. Anthropometric, metabolic and blood pressure values in male teen-agers with Down Syndrome belonging to various ER genotypes.

	XX	Xx	xx	PP	Pp	pp
Number of patients	8	17	3	5	15	8
Age (years)	15±1.8	14.8±1.5	15.4±1.7	14.5±1.2	15.3±1.7	14.1±1.3
BMI (kg/m^2)	26.2±2.8	23.1±1.5	19.4±0.9	22.5±1.9	23.5±2.1	23.8±2.4
AC (mm)	310±15	265±20	252±15	250±10	245±12	238±15
TST (mm)	23.8±1.1	17.1±1.9	14.1±2.1	22.1±2.8	20.4±3.5	19.3±3.6
TCHO (mg/dl) Range 50-220 mg/dl	168±25	160±15	155±22	160±15	160±12	155±15
HDL-CHO (mg/dl) Range 35-75 mg/dl	35.5± 5.8	45.8±6.1	46.1±4.1	42.5±2.8	44.2±2.9	41.6±3.8
LDL-CHO (mg/dl) Range 0-160mg/dl	120±14	108±15	105±16	110±15	106±14	100±12
TG (mg/dl) Range 50-200mg/dl	84.5±6.7	68.5±9.2	65.4±8.5	95±10	90±10	85 ±10
GLU (mg/dl) Range 70-115 mg/dl	95±10	90±7	85±10	87±12	80±10	85±10
SBP (mmHg)	115±10	110±12	112±10	113±9	115±8	115±10
DBP (mmHg)	72±5	72±3	73±3	73±3	72±3	72±5

BMI= Body Mass Index; AC= Arm Circumference; TST= Triceps Skinfold Thickness;
TCHO= Total Cholesterol; HDL-CHO= High Density Lipoproteins – Cholesterol; LDL-CHO = Low Density Lipoproteins – Cholesterol;
TG= Triglycerides; GLU= Glucose;
SBP= Systolic Blood Pressure; DBP= Diastolic Blood Pressure.

BMI, AC, TST: XX vs Xx $p < 0.05$
XX vs xx $p < 0.05$
BMI: Xx vs xx $p < 0.05$
AC, TST: Xx vs xx $p > 0.05$
TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : XX vs Xx $p > 0.05$, XX vs xx $p > 0.05$, Xx vs xx $p > 0.05$
BMI, AC, TST, TCHO, HDL-CHO, LDL-CHO, TG, GLU, SBP, DBP : PP vs Pp, PP vs pp, Pp vs pp, $p > 0.05$

Fifty to eighty percent of obesity is determined by genetic factors [17].

Because many genes are involved in the development of obesity, it is a polygenic disease. Overweight or obesity, are rarely directly caused by a monogenic mutation. However, in some syndromes such as DS and Prader-Willi Syndrome, the pathogenesis of obesity is still unclear.

In 80% of DS children problems related to food or feeding have been observed [18].

Moreover, lifestyle changes in children with DS reduced but did not eliminate the incidence of obesity [18], suggesting a multivariable pathogenesis.

Our findings on anthropometric characteristics and lifestyle of DS subjects are in agreement with other reports that showed a frequent tendency to obesity, low energetic intakes and inadequate physical activity in prepubescent children [1-3,11,19]. Furthermore, the children with DS of both sexes regardless of the ER genotype showed significantly higher BMI and body fat distribution values than controls.

As in the general population, for DS subjects, greater importance should be placed on prevention of obesity through diet and physical activity. Therefore, in this syndrome, the metabolism is down-regulated by lowered basal metabolic rate [20-22]. Our study shows a relationship between lack of ER XbaI site and obesity in both genders. The high frequency of XXER genotype in our cohort of patients may suggest a role of this genotype in developing obesity in DS. These findings are in agreement with other recent studies: Okura et al. [7] found that middle aged women with lack of ER Xba I site had greater BMI and body fat distribution than those with the site. Similarly, other authors have reported a gender specific association of ER XbaI polymorphism with variation in BMI and body fat distribution [9].

An associated lack of ER XbaI site and overweight has also been reported in thalassemic patients of both genders [23]. A recent study on Chinese families using a quantitative disequilibrium test failed to support associations of the ER α gene polymorphisms and BMI [24]. The interaction of various genetic and environmental factors could explain the different results in the Chinese study.

In agreement with another recent study we found negative associations between ER gene polymorphysms and metabolic or cardiovascular disorders [25]. In healthy populations, childhood obesity is associated with severe risk factors from later heart and chronic degenerative disorders [5].

Our findings suggest that the pathogenesis of obesity in DS is multivariable. Low metabolic rate with subnormal enzymatic activity, unhealthy lifestyle particularly characterized by little physical activity and allele variability are possible causes of obesity in these subjects.

In addition, by subdividing DS subjects in ER genotypes, the increased frequency of XXER genotype and the frequent association of this polymorphism with obesity and overweight in DS ought to be considered indicators for additional risk, although these findings may not guarantee metabolic and cardiac complications and several other genetic loci could be of potential relevance to obesity [6]. The mechanism by which estrogen regulates adipose tissue is unclear; it is possible a relationship with leptin production or leptin receptor expression [8].

Moreover it has been explored direct *in vitro* effects of both estrogens and androgens on ob expression and leptin secretion in human adipose tissue. It has been shown that in men the dihydrotestosterone induced a reduction in leptine secretion and *ob* mRNA level, while in women, 17 beta-estradiol, estrogen precursors, testosterone and dehydroepiandrosterone induced an increase in leptin secretion [26].

In conclusion this study points out that starting in childhood, DS children tend to be overweight and obese, regardless of the ER genotype. Furthermore, it raises the possibility that the lack of the ER XbaI site contributes to obesity. This finding is particularly interesting because of the increased frequency of XX ER genotype we found in DS subjects.

However, many other biological, environmental and genetic factors need to be investigated to better understand obesity occurrence in this complex syndrome.

A multicenter study on a larger population would be interesting.

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