Brief Genetics Report

Insulin Resistance Syndrome in Subjects With Mutated RING Finger Protein TRIM37

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We evaluated the glucose and lipid metabolism in 65 patients (aged 1.1–55 years) with mulibrey (muscle-liver-brain-eye) nanism (MUL), which is a monogenic disorder with prenatal-onset growth failure and typical clinical characteristics. MUL is caused by mutations in the *TRIM37* gene, encoding a peroxisomal protein (TRIM37) with E3 ubiquitin-ligase activity. The subjects underwent clinical evaluation, abdominal ultrasonography, and laboratory measurements, including a 3-h oral glucose tolerance test. The results showed a dramatic change in glucose and lipid metabolism with age in MUL subjects. While the children had low fasting glucose and insulin levels, 90% of the adults had high fasting and postload insulin values (up to 1,450 mU/l). A 10-fold decrease in the fasting glucose-to-insulin ratio and a 4-fold decrease in whole-body insulin sensitivity index were observed. Insulin resistance, fatty liver, high serum leptin, hypertension, and acantosis nigricans were already evident in many slim prepubertal children. Half of the adults had type 2 diabetes, and an additional 42% showed impaired glucose tolerance. Seventy percent fulfilled the National Cholesterol Education Program criteria for metabolic syndrome. The peroxisomal targeting and the functional link of TRIM37 to the ubiquitin-proteosome pathway may provide novel clues to the development of metabolic syndrome. *Diabetes* 54:3577–3581, 2005

Insulin resistance syndrome (IRS) or metabolic syndrome, including central obesity, hyperinsulinemia, abnormal glucose tolerance, hyperuricemia, hypertriglyceridemia, and a propensity to atherosclerotic cardiovascular disease, has become the major health problem in industrialized countries (1). Epidemiological and clinical studies (2–4) have revealed that subjects who had a detrimental fetal environment and were born small for gestational age (SGA) have an increased risk of IRS as an adult. The pathophysiology of the development of IRS and type 2 diabetes in these subjects or in the general population is mostly unknown (4–6).

In this work, we studied glucose and lipid metabolism in patients with mulibrey (muscle-liver-brain-eye) nanism (MUL; MIM 253250), constituting a homogenous group of SGA subjects. MUL is an autosomal recessive disorder with prenatal-onset growth failure, minor dysmorphic features, and cardiopathy but no major neurological handicap (7,8). The disorder is caused by mutations in the *TRIM37* gene located on chromosome 17q22-q23 and coding for a novel member, TRIM37, of the tripartite motif (TRIM: comprising RING, B-box, and coiled-coil domains) protein family (9,10). Today, ~130 patients are known worldwide, 88 of them from Finland. Sporadic cases have been reported from different ethnic groups all over the world (7,11). Previously, no metabolic defect has been identified in MUL.

RESEARCH DESIGN AND METHODS

The study group included 65 MUL patients (37 female) aged 1.1–55 years. Sixty-three were homozygous for the Finnish founder mutation (c.493–2A>G), and two were compound heterozygotes for the founder and a c.2212delG mutation. Both mutations predict a truncated TRIM37 protein (9,10). All but three of the MUL patients (95%) were born SGA, with a mean birth length and weight of 45 cm (−3.0 SD score [SDS]) and 2,300 g (−2.7 SDS), respectively. None presented catch-up growth, and the median final height was 137 cm (−4.7 SDS) for female and 152 cm (−3.6 SDS) for male subjects (Table 1). Six patients received growth hormone therapy at the time of blood sampling.

All patients underwent a physical examination with assessment of height, weight, and pubertal stage (according to the criteria of Tanner) (12) and ultrasound evaluation of the liver. Weight was presented as weight for height (percent deviation from the age-specific median weight for height of Finnish standards) (13), since it revealed the weight changes at different ages better than the BMI. The blood pressure was measured three times while the subjects were seated, and the last two measurements were averaged for analysis.

Blood glucose (venous whole-blood glucose) and serum insulin were measured after an overnight fast with concomitant measurement of serum leptin and plasma uric acid, alanine aminotransferase, aspartate aminotransferase, γ-glutamyl transferase, and triglycerides as well as plasma total, HDL, and LDL cholesterol. All but six young patients (aged from 1.1 to 5.0 years) underwent a 3-h oral glucose tolerance test (OGTT; glucose load 1.75g/kg, maximum 75 g) with measurements of blood glucose and serum insulin. Thirty of the patients had a second OGTT and laboratory follow-up 2–5 years after their first test. The OGTT was used to classify the subjects as having normal glucose tolerance, impaired glucose tolerance (IGT; the 2-h venous whole-blood glucose value from 6.7 to 9.9 mmol/l.), or type 2 diabetes (fasting venous whole-blood glucose ≥6.1 mmol/l or the 2-h value ≥10.0 mmol/l) on the basis of World Health Organization criteria (14). The degree of insulin sensitivity was determined from the ratio of fasting glucose and insulin levels (15) and the whole-body insulin sensitivity index was calculated from the OGTT values according to Stumvoll (16,17).

Clinical and laboratory parameters were correlated with age and weight for height, and the explanation rate was expressed as an R^2 value. No significant

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IGT, impaired glucose tolerance; IRS, insulin resistance syndrome; MUL, mulibrey (muscle-liver-brain-eye) nanism; OGTT, oral glucose tolerance test; SDS, SD score; SGA, small for gestational age.

TABLE 1
Glucose and lipid metabolism in 65 patients (37 female) aged 1.1–55 years with MUL

Feature	Normal range	1–10 years of age		11–20 years of age		>20 years of age	
		Median (range)	Abnormal value*	Median (range)	Abnormal value*	Median (range)	Abnormal value*
Anthropometrics							
Height	−2.0 ± 2.0 SDS	−4.0 (−6.1 to 1.0)	30/33 (90)	−3.4 (−5.5 to −1.4)	26/27 (94)	−4.2 (−6.2 to −2.1)	33/35 100)
Weight for height	−20 ± 20%	−17 (−31 ± 16)	0/33 (0)	−8 (−22 ± 75)	5/27 (19)	16 (−24 ± 45)	16/35 (46)
BMI	<91st percentile	14 (11−18)	0/33 (0)	16 (12–32)	2/27 (7)	21 (14–26)	2/35 (6)
Hypertension	<95th percentile	102/65 (85–140/50–65)	10/28 (36)	116/70 (97–140/55–85)	2/11 (18)	139/85 (105–174/70–118)	21/26 (81)
Glucose metabolism							
Fasting blood glucose	4.0–6.1 mmol/l	3.5 (2.2–5.6)	0/33 (0)†	4.4 (2.9–6.1)	1/27 (4)†	4.6 (3.8–10.9)	6/35 (17)†
2-h blood glucose	<6.7 mmol/l	6.1 (3.8–10.9)	8/23 (35)	7.0 (4.6–11.2)	14/27 (52)	10.1 (4.2–21.9)	33/35 (94)
HbA_{1c}	4.9–6.0%	4.4 (4.0–5.6)	0/33 (0)	4.4 (4.0–5.7)	0/27 (0)	5.4 (4.6–6.7)	5/35 (14)
Fasting serum insulin	2–15 mU/l	5 (1–22)	4/33 (12)	12 (2–50)	0/27 (0)	25 (3–178)	27/35 (77)
2-h serum insulin	<75 mU/l	29 (3–203)	5/23 (22)	106 (17–551)	17/27 (63)	280 (41–1,100)	22/35 (63)
Peak postload serum insulin	<150 mU/l	61 (14–291)	7/23 (30)	178 (60–620)	18/27 (67)	430 (88–1,450)	31/35 (89)
Insulin sensitivity							
FGIR index	—	0.80 (8.2–0.18)		0.34 (1.95–0.09)		0.21 (1.53–0.04)	
Stumvoll index	—	0.119 (0.134–0.084		0.107 (0.126–0.070)		0.080 (0.122–0.012	
Lipid metabolism							
Serum total cholesterol	<5.0 mmol/l	3.2 (2.3–5.7)	0/33 (0)	3.8 (3.2–5.6)	3/27 (11)	5.3 (3.3–7.3)	22/35 (63)
Serum LDL cholesterol	<3.5 mmol/l	1.7 (0.6–3.1)	0/33 (0)	2.0 (0.9–3.9)	0/27 (0)	2.6 (0.9–4.6)	13/35 (37)
Serum HDL cholesterol	>1.0 mmol/l	1.3 (0.6–2.3)	7/33 (21)	1.2 (2.1–0.7)	7/27 (26)	1.1 (1.7–0.7)	12/35 (34)
Serum triglycerides	<1.7 mmol/l	0.9 (0.4–2.4)	2/33 (6)	1.2 (0.5–2.3)	6/27 (22)	2.4 (0.7–4.7)	25/35 (71)
Serum leptin (μg/l)	—	3.8 (1.7–10.0)		6.8 (1.9–34.4)		16.1 (2.0–49.9)	
Metabolic assays							
Serum aspartate aminotransferase	<35 units/l	51 (36–147)	30/33 (91)	38 (19–74)	16/27 (60)	38 (12–205)	21/35 (60)
Serum alanine aminotransferase	<35 units/l	44 (16–316)	26/33 (79)	40 (19–85)	16/27 (60)	37 (12–109)	17/35 (49)
Serum γ glutamyltransferase	<50 units/l	27 (10–130)	9/33 (27)	40 (14–104)	14/27 (52)	75 (29–382)	29/35 (83)
Serum uric acid	150–350 μmol/l	372 (200–560)	21/33 (64)	340 (208–490)	17/27 (63)	380 (233–505)	16/35 (46)

*Measurements out of the normal range are presented in each group as the abnormal value of the total amount and as percentage (%). †Abnormal fasting venous whole-blood glucose >6.1 mmol/l.

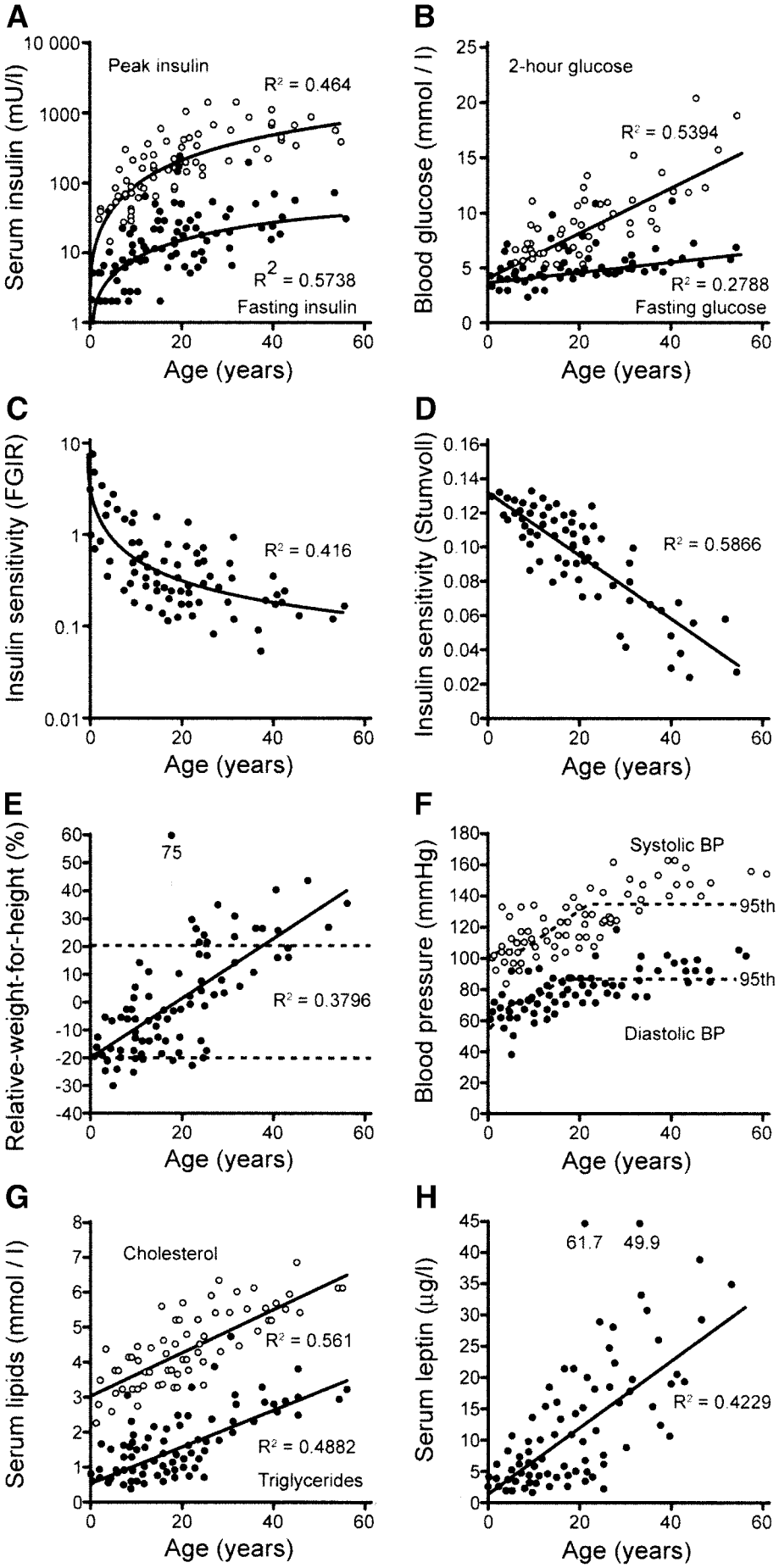

FIG. 1. Glucose and lipid metabolism in 65 MUL patients. *A* and *B*: Serum insulin and blood glucose levels in fasting and OGTT, and the fasting glucose and insulin levels and Stumvoll indexes calculated from these values (15–17) (*C* and *D*). *E*: Weight for height (13). The dashed lines (−20 and +20%) indicate the normal range. *F*: Systolic and diastolic blood pressure (BP). The dashed lines indicate the 95th percentiles at the different age-groups (adjusted to the height) (18). *G* and *H*: Serum total cholesterol, triglyceride, and leptin levels.

changes were noted in the pattern of distribution and in the R^2 value, when all glucose and insulin measurements in fasting (n = 95) or during the OGTT (n = 89) were analyzed, compared with the use of only one measurement form each patient (n = 65). The ethics committee of our hospital approved the study protocol, and all patients or their guardians gave informed consent.

RESULTS

Glucose metabolism in MUL patients showed a dramatic change with age. Patients aged <10 years had low fasting glucose and insulin levels, so that hypoglycemic values (blood glucose <3.4 mmol/l) were recorded in half (14 of 28) of the children (Fig. 1*A* and *B*, Table 1). Fasting glucose concentrations and the 2-h postload glucose concentrations during OGTT increased with advancing age. Among adults (aged >20 years), IGT or type 2 diabetes was diagnosed in 92% of the patients (IGT 42% and type 2 diabetes 50%), indicating serious deterioration in glucose homeostasis in young adult life. The median ages revealing IGT or type 2 diabetes were 18.3 (range 3.1–42.3) and 26.0 (9.5–55.2) years, respectively.

The changes in insulin levels were even more marked; abnormally high fasting (>15 mU/l) and postload peak (>150 mU/l) insulin values were recorded in 90% of the adults (24/26), and the peak insulin values ranged from 138 to 1,450 mU/l (median 430 mU/l) (Fig. 1*A*). Serum basal and postload insulin levels correlated well with the C-peptide levels, and the explanation rate (R^2 value) was 0.6121 and 0.657, respectively. The fasting glucose and insulin levels and Stumvoll indexes showed a 10- and 4-fold decrease with advancing age, indicating the development of severe insulin resistance (Fig. 1*C* and *D*, Table 1).

The development of insulin resistance was accompanied by changes in body composition and lipid metabolism. While prepubertal children were thin, clinically evident abdominal obesity started to develop after puberty, and 42% (14 of 34) of the adults were overweight (weight for height >20%) (Fig. 1*E*). The insulin levels and weight for height, however, correlated poorly, and only 28% (12/43) of the patients with a postload serum insulin level exceeding 150 mU/l were overweight. Serum leptin, total cholesterol, and triglycerides also increased with age (Fig. 1*G* and *H*, Table 1). The leptin levels correlated with the peak insulin levels (R^2 = 0.66) and weight for height (R^2 = 0.606). While growth hormone is known to temporarily increase fasting and postprandial insulin levels, the insulin levels of the six patients receiving growth hormone were not higher than the levels of other subjects in the same age category.

Ultrasonography of the liver showed increased echogenicity suggestive of fatty liver in all adolescents and adults and in 46% of the prepubertal children. Moreover, fatty liver was noted in all patients (n = 12) who underwent a liver biopsy (at the age of 1.8–35 years) and at autopsy in 85% (11/13) of historical MUL patients deceased at the age of 2.0–49 years. Atherogenic vascular changes were noted in two thirds of the autopsied patients. Ninety percent of the adults had elevated γ-glutamyl transferase levels (Table 1).

Acantosis nigricans was present in some children and was a frequent finding postpubertally (80%), particularly in axillae, groins, and the genital region. Serum uric acid was elevated in two thirds of the patients during the period of growth and remained elevated in nearly half of the adults (Table 1). Hypertension (>95th percentile for age and sex) was observed in 81% of the adults (Fig. 1*F*, Table 1), and, overall, 70% of the adults fulfilled the criteria for metabolic syndrome according to the National Cholesterol Education Program Adult Treatment Panel (18).

DISCUSSION

The results indicate that subjects with a rare monogenic disorder, MUL, develop severe IRS in early adulthood. The pathophysiology of this process remains to be solved. While it may be partly explained by poor intrauterine growth, other factors are most likely involved. High risk of IRS has been associated with SGA caused by an unfavorable mother-child environment followed by early postnatal weight gain to obesity (19,20). In MUL, however, the prenatal-onset growth failure is caused by a fetal gene defect, and only a modest weight gain is observed before puberty. Moreover, in our experience the link between SGA and IRS is not definitive, as subjects with Silver-Russell syndrome, who have a phenotype and growth pattern similar to MUL patients, do not show abnormalities in glucose metabolism, as studied by OGTTs (data not shown).

In most monogenic diseases leading to type 2 diabetes, the pathogenic process can be explained by dysfunction of the pancreatic β-cells or obesity (21). Contrary to this, these subjects presented exceptionally high postload serum insulin levels, and fatty liver as well as acanthosis nigricans were often already detectable in slim prepubertal MUL children. Additionally, the weight gain did not correlate with IRS. On the other hand, lipodystrophy with low leptin secretion (21) was not involved in the development of insulin resistance. In MUL, serum leptin levels were unexpectedly high compared with the weight gain. The highly elevated serum leptin levels together with early development of fatty liver seen in MUL suggest that accumulation of liver fat may be a crucial step in the development of IRS in these subjects.

The genetic defects in the Finnish MUL patients predict a nonfunctional truncated TRIM37 protein, a member of the TRIM subfamily of zinc finger proteins (9,10). While the physiological function of TRIM37 in vivo is not known, it is expressed in several tissues (22) and has been localized to peroxisomes in cell cultures (10). More recently, TRIM37 was found to possess TRIM domain–dependent E3 ubiquitin-ligase activity implying defective ubiquitin-dependent degradation of an as-yet-unidentified target protein in the pathogenesis of MUL (23). Interestingly, inherited variation in the peroxisome proliferator–activated receptor γ gene has been implicated in the pathogenesis of type 2 diabetes (24) and also with reduced fetal growth (4). Moreover, recent reports have demonstrated that members of the ubiquitin-proteosome pathway can affect the regulation of insulin signaling cascades and insulin action (6,25). In the light of these findings, the peroxisomal targeting and the functional link of TRIM37 to the ubiquitin pathway may provide novel clues to understanding the insulin action during the fetal period and the development of metabolic syndrome later in life. Thus, MUL could serve as an excellent model for both molecular and clinical studies on the development of type 2 diabetes and metabolic syndrome.

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