Short report

**LMNA** gene mutation as a model of cardiometabolic dysfunction: From genetic analysis to treatment response

V. Chirico a,*, V. Ferràu a, I. Loddo a, S. Briuglia a, M. Amorini a, V. Salpietro a, A. Lacquaniti b,c, C. Salpietro a, T. Arrigo a

a Department of Pediatric Sciences, University of Messina, Messina, Italy
b Department of Internal Medicine, University of Messina, Messina, Italy
c Department of Internal Medicine, Mediterranean Institute for Transplantation and Advanced Specialized Therapies, ISMETT, University of Pittsburgh Medical Center, Palermo, Italy

Received 28 September 2013; received in revised form 18 December 2013; accepted 19 December 2013
Available online 28 January 2014

Abstract

Aim. – This report highlights the metabolic, endocrine and cardiovascular comorbidities in a case of familial partial lipodystrophy (FPLD), and also evaluates the efficacy and safety of metformin therapy.

Methods. – Mutational analysis was carried out of the **LMNA** gene in a teenage girl with an FPLD phenotype. Insulin resistance, sex hormones and metabolic parameters were also evaluated, and echocardiography, electrocardiology and 24-h blood pressure monitoring were also done.

Results. – The patient showed atypical fat distribution, insulin resistance and hypertrophic cardiomyopathy. Physical examination revealed muscle hypertrophy with a paucity of fat in the extremities, trunk and gudetal regions, yet excess fat deposits in the face, neck and dorsal cervical region. **LMNA** sequencing revealed a heterozygous missense mutation (c.1543A>G) in exon 9, leading to substitution of lysine by glutamic acid at position 515 (K515E). Moderate hypertension and secondary polycystic ovary syndrome were also assessed. Treatment with metformin resulted in progressive improvement of metabolic status, while blood pressure values normalized with atenolol therapy.

Conclusions. – Very rapid and good results with no side-effects were achieved with metformin therapy for FPLD. The association of an unusual mutation in the **LMNA** gene was also described.

© 2014 Elsevier Masson SAS. All rights reserved.

Keywords: Familial partial lipodystrophy; Metabolic syndrome; Polycystic ovarian syndrome; Hyperinsulinism; Amenorrhoea; FPLD

1. Introduction

Familial partial lipodystrophy (FPLD) is an autosomal-dominant disorder due to heterozygous missense mutations in the lamins A/C (**LMNA**) gene. This gene, mapped to chromosome 1q21.2-q21.3, encodes a structural protein of the nuclear envelope that regulates function of the nuclear pores and DNA replication. Missense mutations of the **LMNA** gene can affect nuclear function, and may also result in apoptosis and prematur e cell death of adipocytes, thereby causing lipodystrophy [1]. FPLD-linked **LMNA** mutations are mostly located within a highly conserved region (in exon 8) of the gene coding for the proximal C-terminal domain of A-type lamins. The first identified FPLD mutations of the **LMNA** gene were heterozygous missense substitutions at amino acid 482 [2], whereas FPLD features with overlapping symptoms or atypical presentations have been attributed to other heterozygous **LMNA** mutations [3,4].

FPLD has an estimated prevalence of one in every 200,000 individuals, and is characterized by the gradual appearance at puberty of peripheral fat loss, excess accumulation of fat around the neck and chin, perivisceral adiposity and muscle hypertrophy predominantly in the lower limbs, giving the patient a Cushingoid appearance [5,6]. The disorder is also characterized by metabolic alterations, such as insulin resistance, often leading to diabetes, and severe hypertriglyceridaemia with an increased risk of acute pancreatitis [3]. In addition, atherosclerotic processes and high blood pressure can be seen in these patients, while severe liver steatosis is commonly observed [6,7].

Although insulin resistance is a key feature in FPLD, treatment with insulin sensitizers such as metformin and thiazolidinediones has produced conflicting results. Owen et al. [8]
reported no improvement in glycaemic control with worsening of the lipid profile, while others have reported better responses with glitazones than with metformin [9–11].

However, thiazolidinediones such as pioglitazone are not available in some countries, including France and Germany [12], although other drugs such as glucagon-like peptide (GLP)-1 analogues and 11β-hydroxysteroid dehydrogenase type 1 (11β-HSD1) inhibitors may represent future pharmacological treatments. In particular, leptin replacement therapy is a promising option for metabolic complications related to lipodystrophy. The efficacy of metreleptin therapy has been demonstrated in patients with leptin levels above the seventh percentile, with positive effects on lipid profile [13]. However, the current therapeutic options for managing metabolic abnormalities are mainly limited to drugs for the treatment of hypertension, diabetes and dyslipidaemia.

The present report describes the identification and response to treatment of a teenage girl with FPLD, and also its association with a new mutation of the LMNA gene.

2. Case report

A 16-year-old girl was referred to our paediatric endocrine department for evaluation of amenorrhoea, hirsutism and high blood pressure. Her karyotype was 46,XX. Her childhood was uneventful, with adrenarche at age 11 years and menarche at age 12, followed by regular cycles for 3 years. Mild hirsutism, leading to a full beard that needed shaving daily and significant hair growth on her chest, abdomen and back, was associated with menstrual irregularities. At presentation, she had a masculine morphology with a body mass index (BMI) of 25.5 kg/m², and puberty was complete (Tanner stages B5 and P5). Applying the Ferriman–Gallwey method [14], the patient had a score of 13. Moderate acanthosis nigricans of the neck and axillae was also observed.

The patient showed a marked loss of subcutaneous fat in the extremities, leading to prominent muscle contours, but excess fat around the face and submental and dorsal cervical regions, giving her a Cushingoid appearance. Hypertrophy of the sternocleidomastoid and trapezius muscles was also noted bilaterally, and magnetic resonance imaging (MRI) confirmed the loss of adipose tissue in the extremities, with an excess accumulation in the dorsal cervical and intermuscular regions (Fig. 1).

Moderate hepatic steatosis was diagnosed by ultrasound examination. The patient’s clinical history, hormone tests and ovarian morphology on echography established the diagnosis of polycystic ovarian syndrome (PCOS), while 24-h blood pressure monitoring revealed evidence of moderate systolic and diastolic hypertension. Echocardiography showed left ventricular hypertrophy (LVH), whereas electrocardiography revealed no cardiac conduction abnormalities.

Antihypertensive therapy was started with atenolol at 100 mg/day. Laboratory evaluation revealed increased total serum testosterone, but low levels of sex hormone-binding globulin (SHBG) and progesterone. Serum prolactin, thyroid hormones, growth hormone, follicle-stimulating hormone and
Table 1
Anthropometric, hormonal and biochemical profiles of our patient at baseline and during follow-up.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Baseline</th>
<th>Normal values</th>
<th>After 6 months of treatment</th>
<th>After 12 months of treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td>BMI, kg/m²</td>
<td>25.5</td>
<td>18.5–24.9</td>
<td>25</td>
<td>24.7</td>
</tr>
<tr>
<td>Ferriman–Gallwey score</td>
<td>13</td>
<td>5</td>
<td>13</td>
<td>12</td>
</tr>
<tr>
<td>Systolic BP, mmHg</td>
<td>140</td>
<td>&lt;125</td>
<td>120</td>
<td>110</td>
</tr>
<tr>
<td>Diastolic BP, mmHg</td>
<td>85</td>
<td>&lt;70</td>
<td>80</td>
<td>60</td>
</tr>
<tr>
<td>Serum glucose, mg/dL</td>
<td>95</td>
<td>65–100</td>
<td>90</td>
<td>87</td>
</tr>
<tr>
<td>T120 serum glucose, mg/dL</td>
<td>98</td>
<td>&lt;140</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Insulin, μU/mL</td>
<td>37</td>
<td>&lt;10</td>
<td>20</td>
<td>9</td>
</tr>
<tr>
<td>T120 insulin, μU/mL</td>
<td>169</td>
<td>&lt;75</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>HOMA-IR, %</td>
<td>8.6</td>
<td>&lt;2.5</td>
<td>4.4</td>
<td>1.9</td>
</tr>
<tr>
<td>HbA1c, %</td>
<td>5.2</td>
<td>&lt;6</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Triglycerides, mg/dL</td>
<td>333</td>
<td>&lt;150</td>
<td>200</td>
<td>151</td>
</tr>
<tr>
<td>Total cholesterol, mg/dL</td>
<td>205</td>
<td>&lt;180</td>
<td>175</td>
<td>170</td>
</tr>
<tr>
<td>HDL cholesterol, mg/dL</td>
<td>40</td>
<td>&gt;45</td>
<td>49</td>
<td>63</td>
</tr>
<tr>
<td>LDL cholesterol, mg/dL</td>
<td>123</td>
<td>&lt;100</td>
<td>85</td>
<td>78</td>
</tr>
<tr>
<td>Serum prolactin, mIU/L</td>
<td>196</td>
<td>55–425</td>
<td>185</td>
<td>203</td>
</tr>
<tr>
<td>FT3, pg/mL</td>
<td>3.8</td>
<td>2.3–4.2</td>
<td>3.5</td>
<td>3.9</td>
</tr>
<tr>
<td>FT4, μM</td>
<td>16.8</td>
<td>8.5–17.5</td>
<td>15.2</td>
<td>16.1</td>
</tr>
<tr>
<td>TSH, mU/L</td>
<td>1.2</td>
<td>0.1–3.5</td>
<td>1.5</td>
<td>2.1</td>
</tr>
<tr>
<td>Growth hormone, ng/dL</td>
<td>6.5</td>
<td>&lt;10</td>
<td>7.3</td>
<td>8.2</td>
</tr>
<tr>
<td>FSH, mIU/mL</td>
<td>4.2</td>
<td>2.5–10</td>
<td>5.2</td>
<td>4.9</td>
</tr>
<tr>
<td>LH, mIU/mL</td>
<td>10.6</td>
<td>2–15</td>
<td>11.5</td>
<td>11.8</td>
</tr>
<tr>
<td>Cortisol, μg/dL</td>
<td>15.4</td>
<td>5–20</td>
<td>12.7</td>
<td>14.4</td>
</tr>
<tr>
<td>Testosterone, ng/dL</td>
<td>82.54</td>
<td>6–48</td>
<td>51</td>
<td>38</td>
</tr>
<tr>
<td>Androstenedione, ng/mL</td>
<td>5.10</td>
<td>0–2.8</td>
<td>3.1</td>
<td>2.5</td>
</tr>
<tr>
<td>SHBG, nmol/L</td>
<td>6</td>
<td>20–85</td>
<td>22</td>
<td>25</td>
</tr>
<tr>
<td>Ovarian follicles, n</td>
<td>15</td>
<td>–</td>
<td>9</td>
<td>7</td>
</tr>
<tr>
<td>Ovarian volume, cm³</td>
<td>23</td>
<td>–</td>
<td>12</td>
<td>6</td>
</tr>
</tbody>
</table>

BMI: body mass index; BP: blood pressure; HOMA-IR: homeostasis model assessment for insulin resistance; Hba1c: glycated haemoglobin; T120: after 120 min (2h) of oral glucose tolerance test; HDL/IDL: high-density/low-density lipoprotein; FT3: free triiodothyronine; FT4: free thyroxine; TSH: thyroid-stimulating hormone; FSH: follicle-stimulating hormone; LH: Luteinizing hormone; SHBG: sex hormone-binding globulin.

a Metformin (1700 mg/day), atenolol (100 mg/day), no lipid-lowering drugs.

b Calculated from ovary with the greater number of follicles.

c Calculated from the homolateral ovary.

Luteinizing hormone were all normal. Hypercortisolism was absent. The physiological response of 17-hydroxyprogesterone (17OHP) to adrenocorticotropic hormone (ACTH) was also assessed.

Significant insulin resistance was revealed by homeostasis model assessment (HOMA-IR), calculated as the product of fasting insulin level and fasting glucose level (mM), divided by 22.5. In addition, a 120 min oral glucose tolerance test (OGTT) using 75 g of glucose was performed after a 12-h overnight fast. At baseline, glucose level was 95 mg/dL; after 30 min, it was 102 mg/dL; after 60 min, 97 mg/dL; after 90 min: 98 mg/dL; and after 120 min, 98 mg/dL. At the same time, insulin levels were 37 μU/mL, 51 μU/mL, 101 μU/mL, 139 μU/mL, and 169 μU/mL, respectively. Glucose tolerance was normal, whereas fasting and glucose-stimulated insulin levels were high. Glycated haemoglobin A1c (Hba1c) level was in normal range (Hba1c: 5.2%).

Elevated serum liver enzymes were revealed, with no obvious cause of liver cytosis. The lipid profile showed high serum triglycerides and total cholesterol, and low serum high-density lipoprotein (HDL) cholesterol concentrations. Leptin level was 7.5 ng/mL at baseline.

In view of her characteristic physical appearance and evidence of metabolic abnormalities related to insulin resistance, FPLD was suspected and investigated. Sequencing the LMNA gene revealed a heterozygous missense mutation (c.1543 A>G) in exon 9, leading to substitution of lysine by glutamic acid at position 515 (K515E), thus confirming the diagnosis of FPLD. Parental samples were not obtainable, making it impossible to determine whether the change was de novo or inherited. Metformin at a dose of 1700 mg/day was initiated.

2.1. Follow-up period

Reduced insulin and fasting glucose levels and an improved lipid profile were observed after 6 months of treatment. Correction of amenorrhoea was also noted. However, after 12 months, there were no significant changes in BMI and body fat distribution, although acanthosis nigricans had disappeared, and glucose tolerance and insulin sensitivity improved. Liver function tests slowly improved with a reduction in liver volume, as evaluated by ultrasound. No side-effects of therapy were reported. Table 1 shows the patient’s anthropometric and laboratory parameters at baseline and during the follow-up.
3. Discussion

The aim of the present report was to share the specific characteristics of a patient with FPLD-linked heterozygous LMNA gene mutation at metabolic, endocrine and cardiac levels. The precise aetiology of insulin resistance and hyperinsulinaemia in FPLD is not clear, but is most likely the consequence of ectopic fat accumulation in non-adipose tissues. Also, these metabolic complications are qualitatively the same as or similar to those observed in obese patients. Thus, this type of lipodystrophy might represent a human model of ‘metabolic syndrome’.

The severity of hypertriglyceridaemia and hyperandrogenism in FPLD was strictly related to insulin resistance. In fact, normalization of lipid and hormone profiles was achieved after improvement of insulin and glucose metabolism through metformin administration.

The goals of therapy in FLDP patients are to improve the metabolic profile and reduce excess fat accumulation in non-adipose tissues. Lifestyle modification is a necessary first step, although pharmacological treatment is often required. Several lines of evidence suggest that a good therapeutic response can be obtained with oral hypoglycaemic agents.

The present case study has demonstrated that progressive improvements in sex-hormone levels, lipid profile, hyperinsulinaemia, insulin resistance and resolution of amenorrhoea were achieved by administration of metformin, albeit with no changes in weight or body fat distribution. There was also normalization of ovarian morphology as assessed by ultrasound.

PCOS is strictly related to changes in insulin levels, while metformin acts through hypoglycaemic and insulin-sensitizing effects, and also through direct effects on insulin-mediated steroiogenesis and glucose uptake in ovarian cells [15]. However, in non-genetic-related PCOS, results with metformin have been mixed [16]. Conflicting metformin results were reported in women with FPLD by Gambineri et al. [9], who saw no clinical improvements after several years of uninterrupted therapy with metformin, whereas pioglitazone led to progressive improvement. Metformin treatment of a young girl who had lipodystrophy as well as insulin resistance, hyperinsulinism and a high risk of diabetes in adulthood could be also considered a preventative strategy. This pharmacological intervention was closely associated with lifestyle education, which is insufficient on its own and difficult to maintain. However, further studies are now needed to establish the true value of metformin in preventing diabetes development in paediatric patients.

Cardiac evaluation of our patient also revealed LVH that was probably related to her moderate hypertension, although neither dilated cardiomyopathy nor cardiac conduction system abnormalities were found, despite the fact that such cardiac complications have been reported in FPLD cases [17]. Atherosclerosis represents another common comorbidity of FLPD and is closely related to a wide range of proatherogenic metabolic disturbances (such as dyslipidaemia, hyperinsulinaemia, hypertension and diabetes). In addition, LMNA mutations exert direct proatherogenic effects on endothelial cells, which could contribute to these patients’ early atherosclerosis [18].

Our patient’s blood pressure was managed with a beta-blocker. However, LVH remained unchanged after 1 year of treatment. The condition is probably secondary to hypertension, although it has been demonstrated that FPD patients are characterized by primitive cardiomyopathy of various severity [19].

There are no management guidelines for patients with cardiomyopathy due to LMNA gene mutations. Also, whether medical treatment using diuretics, beta-adrenergic blockers or angiotensin-converting enzyme inhibitors are effective for cardioprotection remains unclear.

In our present case, there was an LMNA gene mutation at c.1543 A>G (K515E) in exon 9. Interestingly, our patient had a similar pattern of body fat loss as typically seen in patients with FPLD harbouring R482Q, R482W and R482L mutations in exon 8. In addition, it is known that heterozygous substitutions of LMNA/arginine-482 (R482) are the cause of more than 80% of LMNA-linked FPLD cases, whereas non-R482 mutations are usually associated with a less severe form of lipodystrophy [20].

The LMNA gene has been extensively sequenced, and several mutations have been described in a gene-specific database (online at http://www.dmd.nl/nmdb/home.php?select_db=LMNA). However, our patient also showed an unusual mutation, already been described in adults, characterized by the accumulation of fat in the face and neck, but with a loss of subcutaneous fat in the arms, legs and trunk, beginning at puberty. Although this mutation is most probably a rare event, it could be associated with a better prognosis, as evidenced by the excellent response to metformin seen during the follow-up period.

In conclusion, PCOS with severe hyperandrogenism, acanthosis nigricans and insulin resistance in non-obese women calls for molecular studies of the LMNA gene. Our present data suggest significant therapeutic efficacy with metformin for the treatment of hyperandrogenism and ovulatory dysfunction in PCOS associated with partial lipodystrophy and severe insulin resistance. Cardiac examination should also be carried out in FPLD patients.

Disclosure of interest

The authors declare that they have no conflicts of interest concerning this article.

Funding source: This research did not receive any specific grant from any funding agency in the public, commercial or not-for-profit sector.

Financial disclosure: The authors have no financial relationships relevant to this article to disclose.

Appendix A. Supplementary material

Supplementary materials (French summary) associated with this article can be found at http://www.sciencedirect.com at http://dx.doi.org/10.1016/j.diabet.2013.12.008.

References