Sequencing of the Reannotated *LMNB2* Gene Reveals Novel Mutations in Patients with Acquired Partial Lipodystrophy

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The etiology of acquired partial lipodystrophy (APL, also called "Barraquer-Simons syndrome") is unknown. Genomic DNA mutations affecting the nuclear lamina protein lamin A cause inherited partial lipodystrophy but are not found in patients with APL. Because it also encodes a nuclear lamina protein (lamin B2) and its genomic structure was recently reannotated, we sequenced *LMNB2* as a candidate gene in nine white patients with APL. In four patients, we found three new rare mutations in *LMNB2*: intron 1 −6G→T, exon 5 c.643G→A (p.R215Q; in two patients), and exon 8 c.1218G→A (p.A407T). The combined frequency of these mutations was 0.222 in the patients with APL, compared with 0.0018 in a multiethnic control sample of 1,100 subjects ($P = 2.1 \times 10^{-7}$) and 0.0045 in a sample of 330 white controls ($P = 1.2 \times 10^{-5}$). These novel heterozygous mutations are the first reported for *LMNB2,* are the first reported among patients with APL, and indicate how sequencing of a reannotated candidate gene can reveal new disease-associated mutations.

Mutations in several genes have been found in patients with inherited lipodystrophies, including mutations in *LMNA* (MIM 150330), *PPARG,* and *ZMPSTE24* in partial lipodystrophy and mutations in *BSCL2* and *AGPAT2* in congenital total lipodystrophy.[1–4] However, the potential genetic contribution to acquired partial lipodystrophy (APL, also called "Barraquer-Simons syndrome" [MIM 608709]) and acquired generalized lipodystrophy (AGL) has not been established.[5,6] A family history is usually absent in APL and AGL, whereas a wide range of secondary factors and conditions are often associated.[5,6] For instance, systemic lupus erythematosis, dermatomyositis, hypocomplementemia, and membranoproliferative glomerulonephritis are sometimes seen in patients with APL,[6] whereas panniculitis and various autoimmune diseases are sometimes seen in patients with AGL.[5] An unresolved issue is whether acquired lipodystrophy syndromes have an underlying component of genetic susceptibility.[6] Because such patients are sporadically affected and are typically not members of multigenerational families with multiple affected members, alternative strategies to classic positional cloning are required to find susceptibility genes.

Candidate-gene sequencing is one strategy to implicate a susceptibility gene through identification of rare DNA sequence mutations that are present in affected individuals and absent in healthy individuals. Putative biological function is a common criterion used to select a candidate gene for genomic DNA sequence analysis. For instance, because structural abnormalities in a nuclear envelope protein gene—namely, *LMNA,* encoding lamin A/C—cause familial partial lipodystrophy type 2 (FPLD2 [MIM 151660]), it is possible that mutations might be found in genes that encode related nuclear envelope proteins in other lipodystrophy syndromes. In 2001, we used the genomic information available at that time to develop amplification primers for candidate genes encoding nuclear envelope proteins, including *LBR, LMNB1,* and *LMNB2* (MIM 150341), which encode lamin B receptor, lamin B1, and lamin B2, respectively.[7] We then sequenced these candidate genes in patients with AGL or APL who had no mutations in *LMNA*.[7] Although we identified several common polymorphisms, we found no disease-causing mutations and concluded that sequence variants affecting the nuclear lamina proteome were not likely to be associated with APL or AGL.[7]

Recent observations suggest that early versions of mammalian genome maps underestimated the total numbers of exons. For instance, new computational algorithms have revealed thousands of previously unrecognized exons in mammalian genomes.[8] This understanding has necessitated significant reannotation of cDNA and genomic DNA databases by "stitching" newly identified exons into previously annotated genes. On the basis of this biological reality, we revisited the reannotated genomic structures of nuclear proteome genes[7]; in doing so, we found that *LMNB2* had 6 exons identified in 2001 but 12 exons in 2005 (fig. 1*A*).

Using new primers, we interrogated the coding regions of the reannotated *LMNB2* gene in nine unrelated patients with APL (table 1) whose conditions were diagnosed according to established criteria.[6] Specifically, subjects had gradual onset of bilateral subcutaneous fat loss from the head, neck, upper extremities, and thorax but not the lower extremities.[6] In addition, subjects had supportive

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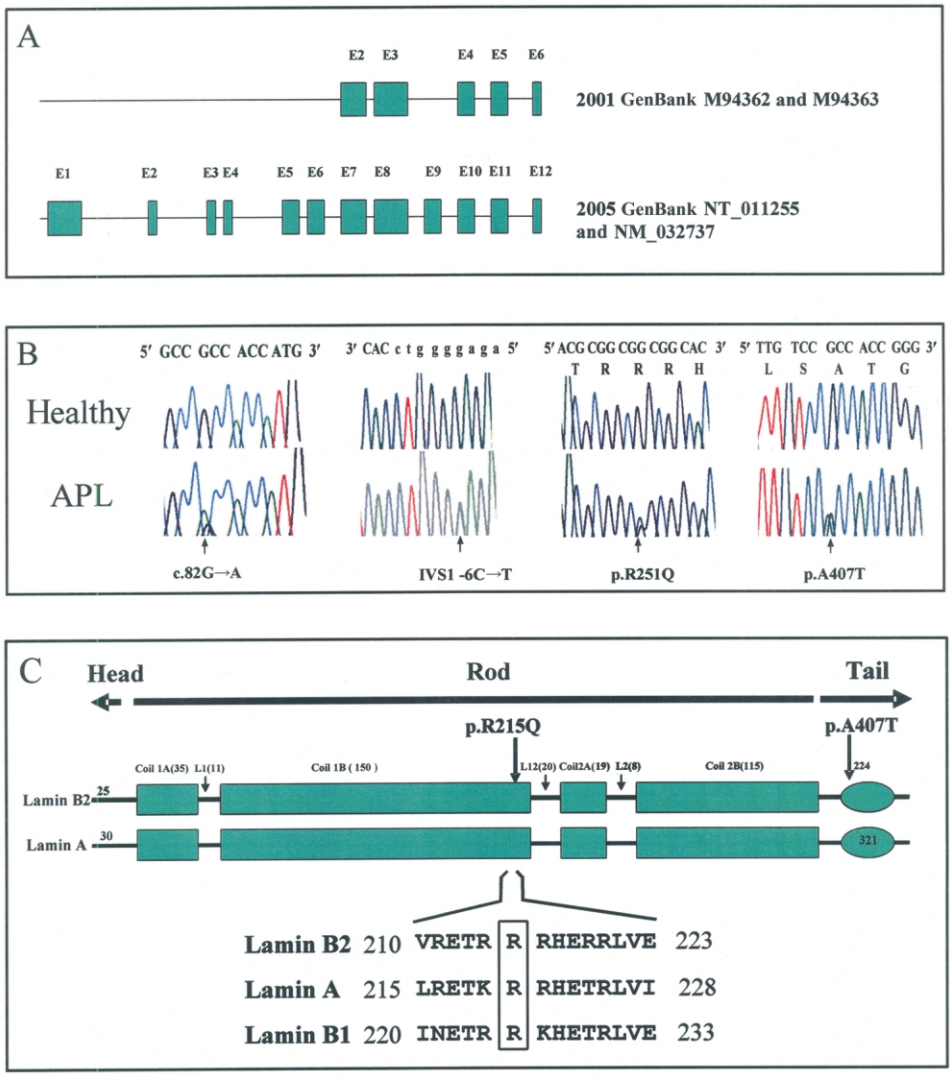

Figure 1. *A,* Genomic structure of *LMNB2* as it was known in 2001 and 2005 (with respective reference sequence numbers indicated). Boxes indicate exons, with the exon number shown above. *B,* Electropherogram tracings of genomic DNA sequences of the *LMNB2* gene from healthy control subjects (top row of sequences) and subjects with APL (lower row of sequences). Normal genomic sequence for each region is shown above the tracings, in addition to 5′ and 3′ orientation. Mutation names are given below the tracings, with arrows pointing to mutant heterozygous sequences. The sequence designated c.82G→A in exon 1 was later determined to be a polymorphism that was seen at relatively high frequency in control subjects of several ethnic backgrounds and was not significantly associated with APL in this sample. *C,* Schematic diagram of head, rod, and tail domains of lamins B2 and A. Subdomains of the rod domain are shown as numbered coil domains and numbered "linker" domains (L). The numbers of amino acid residues for each region of each lamin type are shown in parentheses above the domain maps. The positions of the two *LMNB2* missense mutations are shown (*arrows*). For the p.R215Q mutation within Coil 1B of the highly conserved rod domain, the homologous regions from lamins B2, A, and B1 are shown below the map, with the respective amino acid numbers on each side of this conserved region. R215 in lamin B2 and the corresponding residue in the other lamins are boxed.

diagnostic criteria, including absence of a clear family history of lipodystrophy and onset of lipodystrophy before adulthood. Other types of lipodystrophies, including inherited varieties such as congenital generalized lipodystrophy (CGL [MIM 269700]), FPLD2, or familial partial lipodystrophy type 3 (FPLD3 [MIM 604367]) and acquired varieties such as AGL and HIV-associated lipodystrophy, were excluded. Other associated conditions in the patients with APL are shown in table 1. In addition, one patient had dermatomyositis and alopecia areata, one patient had bilateral carpal tunnel syndrome, one patient was hypothyroid, and the single patient with C3 deficiency and biopsy-proven membranoproliferative glomerulonephritis had received three renal transplants, with subsequent development of secondary and tertiary hyperparathyroidism. All patients were white. All study subjects provided

Table 1. Demographic Clinical Attributes of Nine Patients with APL

Characteristic	Value
Demographic:	
Mean (range) age [years]	55.4 (26–80)
White	9 (100)
Female sex	7 (78)
Family history of lipodystrophy	0 (0)
Mean (range) age at fat-loss onset [years]	15.6 (5–20)
Mean (range) age at diagnosis of APL [years]	26.4 (9–40)
Affected regions with subcutaneous fat loss:	
Face	8 (89)
Neck	9 (100)
Arms	9 (100)
Chest	9 (100)
Abdomen	7 (78)
Thighs	2 (22)
Calves	0 (0)
Associated conditions:	
Complement C3 deficiency	1 (11)
Glomerulonephritis	1 (11)
Dermatomyositis	2 (22)
Treated type 2 diabetes	5 (56)
Treated hypertension	5 (56)
Type IV or V dyslipoproteinemia	8 (89)
Hepatomegaly	5 (56)
Hirsutism, no. (%) of women	3 (43)
Polycystic ovarian disease, no. (%) of women	2 (29)
Early vascular disease (at age <50 years)	2 (22)

NOTE.—Data are no. (%) of patients, unless otherwise indicated.

informed consent for DNA analysis, and the Institutional Review Board of the University of Western Ontario approved the study (protocol 07920E).

No coding sequence or promoter mutation in *LMNA, PPARG, ZMPSTE24, BSCL2,* or *AGPAT2* had been found in genomic DNA sequence of any patient with APL by use of established protocols.[9–11] *LMNB2* was originally sequenced using primer pairs[7] to amplify the five C-terminal exons of the presumed total of six exons from the 2001 reference sequence (fig. 1*A*; GenBank accession numbers M94362 and M94363). However, in 2005, the reannotated *LMNB2* gene in chromosome 19 was shown to have 12 exons (fig. 1*A*; GenBank accession numbers NT_011255 and NM_032737). We made new primers to amplify all 12 exons plus >50 nt at each intron-exon boundary of *LMNB2* (table 2). Standard amplification conditions were used for all reactions, including annealing at 60°C for each of 30 cycles. Sequences were read on a Model 3730 Automated DNA Sequencer (Applied Biosystems). Mutations were confirmed by a second independent sequencing reaction performed on a different day.

The frequencies of *LMNB2* mutations were determined in a multiethnic control sample of 1,100 healthy individuals comprising 330 individuals of white descent, 35 individuals of African descent, 375 individuals of East Indian descent, and 360 individuals of Chinese descent. Genotyping methods are shown in table 3. All genotyping was performed with blinding to affected status, and sequence-proven positive controls were run with each reaction. A random 5% of genotypes were repeated, and no disparities were found for any genotype call. All statistical analyses were performed using SAS version 8.1 (SAS Institute). Between-group comparisons of proportion of discrete variables and calculations of relative risk for carriers of the variants were made using χ^2 contingency analysis. The nominal P value for significance was .05.

Among the nine subjects with APL described in table 1, we found four heterozygous genomic sequence variants of *LMNB2* in five subjects (fig. 1*B*). These variants were exon 1 c.82G→A, intron 1 −6G→T, exon 5 c.643G→A (p.R215Q; in two patients), and exon 8 c.1218G→A (p.A407T). None of these *LMNB2* variants had been previously reported in any database.

To determine how specific these variants were to our sample of patients with APL, we genotyped our nonlipodystrophic control samples (table 4). Genotyping conditions are described in table 3. The c.82G→A variant had frequencies of 0.0091, 0.0, 0.008, and 0.067 in the white, African, East Indian, and Chinese samples, respectively, and an overall allele frequency of 0.027 in the multiethnic control sample. The intron 1 −6C→T and p.A407T variants were not seen in the multiethnic control sample. The p.R215Q variant was seen in 3 of the 330 white controls (allele frequency 0.0045) and in 1 of the 375 East Indian controls (allele frequency 0.0013) but was absent from the other control samples (overall allele frequency 0.0018). Allele frequencies were then compared between the APL and control samples. The observation of one *LMNB2* c.82G→A heterozygote among nine subjects with APL indicated an allele frequency (0.056) that was not significantly different from the allele frequency in the multiethnic control sample and the white subsample (0.027 and 0.0091, respectively; both $P > .05$). This suggested that, although the c.82G→A variant was relatively uncommon in normal subjects, it was not significantly associated with APL in this sample; the relative risk (RR) of APL in carriers of the variant was 2.1 (95% CI 0.35–12.9; $P > .05$).

However, the combined frequency of the *LMNB2* mutations intron 1 −6C→T, p.R215Q, and p.A407T in the nine subjects with APL was 0.222 (4 of 18 alleles) compared with 0.0018 (4 of 2,200 alleles) in all controls

Table 2. Amplification Primers for Reannotated *LMNB2* Gene

Exon(s)	Primer (5′→3′) Forward	Primer (5′→3′) Reverse	Product Size (bp)
1	GGCGCGGATTGAATGAG	GAAACCCCGCGGAAACC	413
2	TGCTGTAGTCCAAGGGGAAG	GTGCCAGGATCAGGGTGAC	238
3 and 4	CCTGAGGTGCTCCCTGTG	ACAGCCATGAGGGTGGACT	500
5 and 6	CTTGGGCCTGGTGCTTG	CAGGGCCCAGAAGTTGG	499
7	TCACCTGGGCCTTGTGAC	CCTCCTCCTCACTCTGTGCT	324
8	GGGGCTCCCACTACCAGT	CCTGGTCACCCCCATCAG	348
9	GTCACAGTCCTGGTGGCTGA	CAAGTCCTGTGCCTCCAGTC	235
10 and 11	GAAGGGGGCTTTGTGGTAG	TGTGTGCACGAGCTCACTCT	511
12	GTGGGGGCAGCCATCTAC	TGGCTCTGGGTAAAGAAAGG	163

Table 3. *LMNB2* Genotyping Procedures

Variant	Method	Conditions	Diagnostic Readout
c.82G→A	Restriction isotyping	Amplification with primers 5′-GGCGCGGATTGAATGAG-3′ and 5′-TCTCTGAGATCTTGAGCAGGAG-3′; digestion with *MslI*; electrophoresis in 2% agarose gel	−6G allele: single 253-bp fragment; −6A allele: 180- and 73-bp fragments
Intron 1 −6C→T	Restriction isotyping	Amplification with exon 2 primers; digestion with *BsaI*; electrophoresis in 2% agarose gel	−6C allele: 186- and 52-bp fragments; −6T allele: single 238-bp fragment
c.643G→A (p.R215Q)	Restriction isotyping	Amplification with exons 5 and 6 primers; digestion with *Msp*AI; electrophoresis in 2% agarose gel	R215: 318- and 181-bp fragments; Q215: 237-, 181-, and 81-bp fragments
c.1218G→A (p.A407T)	Allele-specific amplification	SNaPShot procedure (Applied Biosystems) with primers 5′-CCCACTACCAGTCCCACCT-3′ and 5′-CAAGGGCTCCTCCACCTC-3′ and allele-specific primer 5′-AGCAGCGGCAGGTTGTCC-3′	Fluorometric readout on Applied Biosystems 3730 Automated DNA Sequencer

($P = 2.1 \times 10^{-7}$) and 0.0045 (3 of 660 alleles) in white controls ($P = 1.2 \times 10^{-5}$). Thus, the RR of APL for carriers of these variants compared with all control subjects was 110.1 (95% CI 35.9–271.0); compared with white control subjects, it was 37.9 (95% CI 12.7–83.3). The clinical features of the four subjects with APL with these three *LMNB2* mutations are shown in table 5.

When the three mutations were considered individually, the presence of p.A407T in one of nine subjects with APL, together with its complete absence from 1,100 multiethnic and 330 white control subjects, was significant; the RRs of APL for carriers were 138.5 (95% CI 26.4–138.5) and 42.3 (95% CI 8.1–42.3) compared with all controls and the white controls, respectively. The intron 1 −6C→T mutation had the same RRs of APL for carriers compared with multiethnic and white control samples. The recurrence of p.R215Q in two of nine unrelated patients with APL compared with 3 of 330 reference white subjects was highly significant; the RR of APL for white carriers of the variant was 19.1 (95% CI 4.9–47.7). These two APL-affected patients who were heterozygous for p.R215Q had some similarities (e.g., both were women with long-term diabetes and dyslipidemia) but also some phenotypic differences (e.g., one had retinopathy and xanthomata, whereas the other had dermatomyositis, hepatosplenomegaly, osteoporosis, hirsutism, and alopecia). Interestingly, p.R215Q was also present, albeit at a very low frequency, in the control East Indian sample but not in the other ethnic groups; RR of APL for carriers compared with the overall multiethnic control group was 52.5 (95% CI 13.2–147.6). However, we recognize the limitations of RR calculations for rare alleles and a rare disease phenotype, especially for a small cohort of patients. Control subjects with these mutations had been randomly ascertained from healthy populations and did not have diabetes, hypertension, or dyslipidemia.

The findings indicate how resequencing of a candidate gene that has been reannotated at the genomic level can reveal new disease-associated mutations. The new *LMNB2* mutations include two coding-sequence variants—p.R215Q in two patients with APL and p.A407T in one patient with APL—and one variant near a newly defined intron-exon boundary (intron 1 −6C→T). The high prevalence of the mutations in the nine patients with APL and the very low frequency of the mutations in the control samples indicates an extremely strong, nonrandom association between the mutations and the APL phenotype.

Although it has been recognized as a clinical entity for almost a century, the etiology of APL has been unclear.[6] APL was first described as a form of progressive lipodystrophy.[6,12–14] Most reported APL cases have been sporadic, and the female:male affected ratio is ~4:1.[6] This is similar to the female bias (or at least clinical recognition) of the expression of FPLD2.[1–3] APL does not segregate as a classic simple Mendelian trait in families with an affected proband, nor does it typically cluster in families.[6,15–17] Discordance for APL in MZ twins has been noted.[18,19] In addition, the presence of dermatomyositis, systemic lupus erythematosis, C3 hypocomplementemia, and membranoproliferative glomerulonephritis in some patients with

Table 4. Allele Frequencies in Controls of New Mutations and Variants Found in the Reannotated *LMNB2* Gene among Patients with APL

	Allele Frequency				
Variant	Overall ($N = 1,100$)	White ($n = 330$)	African ($n = 35$)	East Indian ($n = 375$)	Chinese ($n = 360$)
c.82G→A	.027	.0091	.0	.0080	.067
Intron 1 −6C→T	.0	.0	.0	.0	.0
Exon 5 c.643G→A (p.R215Q)	.0018	.0045	.0	.0013	.0
Exon 8 c.1218G→A (p.A407T)	.0	.0	.0	.0	.0

Table 5. Clinical Features of Subjects with APL and Rare *LMNB2* Mutations

Characteristic	Finding in Subject with Diagnosed APL			
	APL02	APL03	APL07	APL09
LMNB2 mutation	p.R215Q	p.R215Q	Intron 1 −6C→T	p.A407T
Present age (years)	40	64	46	59
Age when fat loss began (years)	~5	~16	~17	~17
Age at APL diagnosis (years)	9	30	27	25
Diabetes, age at onset (years)	Yes, 19	Yes, 37	Yes, 27	No
Extent of fat loss	Symmetrical, upper body, to knees	Symmetrical, upper body, to upper thighs	Symmetrical, arms, legs, and gluteal region	Symmetrical, upper body, to upper thighs
Dyslipidemia	Type V	Type IV	Type V	Type IV
Hypertension	No	Yes	Yes	Yes
Polycystic ovarian syndrome	No	Yes	Yes	No
C3 deficiency	No	No	No	No
Membranoproliferative glomerulonephritis	No	No	No	No
C3 nephritogenic factor	No	No	No	No
Autoimmune disease	No	Yes	No	No
Dermatomyositis	No	Yes	No	No
Xanthomata	Eruptive on elbows and knees	No	No	No
Hirsutism	No	Yes	Yes	No
Retinal involvement	Yes	No	No	No
Other	Bilateral carpal tunnel syndrome	Hepatosplenomegaly, coronary artery disease, osteoporosis, alopecia	Peripheral neuropathy, gastroparesis, depression	Cerebral infarct at age 30 years

NOTE.—All subjects shown in the table are white women.

APL[6] suggests that such factors might trigger expression of the disease. These associations are consistent with the idea that APL represents a complex phenotype, possibly with a component of genetic susceptibility that requires the presence of environmental factors or acquired disorders to be expressed. The fact that not all subjects with APL had *LMNB2* mutations indicates possible molecular heterogeneity; if other associated genes are identified, it might be appropriate to divide APL into molecular subtypes, analogous to FPLD2 and FPLD3.[2] Furthermore, the fact that a few carriers of *LMNB2* mutations, particularly p.R215Q, were found among apparently healthy controls is consistent with a complex etiology for APL, in which a susceptibility allele may not be sufficient for expression of the phenotype, instead requiring the presence of additional factors.

LMNB2 was selected as a candidate gene for genomic sequence analysis in patients with APL because, like *LMNA,* which is the causative gene for FPLD2, it encodes a nuclear envelope constituent. The implication of mutant *LMNB2* in APL widens the range of disorders associated with mutations in intermediate filament proteins[20] and, in particular, increases to four the number of nuclear envelope proteins that are associated with human diseases.[21] Other nuclear envelope constituents in which mutations are associated with disease include lamin A/C (numerous laminopathies[22]), emerin (Emery-Dreifuss muscular dystrophy [MIM 310300]), and the lamin B receptor (Pelger-Huet anomaly [MIM 169400] and hydrops-ectopic calcification-moth-eaten skeletal dysplasia [MIM 215140]).

The p.R215Q mutation occurs within the rod domain of lamin B2 (fig. 1*C*) and alters an arginine residue that is conserved not only in human lamins A/C and B1 but also in mouse and zebrafish lamin B2 genes. Furthermore, certain missense mutations affecting this domain in *LMNA* result in a dilated cardiomyopathy phenotype—namely, p.E203K and p.L215Q—and in Emery-Dreifuss muscular dystrophy—namely, p.H222P.[22] In contrast, the p.A407T mutation occurs within the much less evolutionarily conserved tail domain of *LMNB2* (fig. 1*C*); the analogous residue to A407 is a threonine in both lamin A/C and lamin B1. Furthermore, there are no missense mutations affecting this residue in lamin A/C, although mutations in proximal residues p.R386K and p.R401C underlie Emery-Dreifuss muscular dystrophy.[22] The presence of the intron 1 −6C→T variant near an intron-exon boundary suggests that it might affect RNA splicing. However, functional assessment of any of these mutations would be difficult, given that the function of lamin B2 is not precisely defined,[23–26] particularly in comparison with lamin B1.[27] The range of potential roles of lamin B2 includes supporting the inner side of the nuclear envelope, stabilizing the nucleus and chromatin, and regulating gene expression.[28] Lamin B2 replicates in the 1st min of S-phase.[28] The *LMNB2* origin of replication is very well characterized, accommodates the defined start site of DNA synthesis, binds to origin-recognition complexes in living cells and in vitro, and supports the initiation of DNA synthesis at ectopic chromosomal locations.[28] It is thus possible that mutations that alter the function of lamin B2 might have an effect on the rate of DNA replication.

In summary, we have demonstrated human disease-associated mutations in newly defined coding sequences of a recently reannotated candidate gene, and, in doing so,

we found the first human mutations in *LMNB2* and the first mutations in APL. From these studies, APL appears to behave like a complex trait with a component of genetic susceptibility, which, in some patients, is mediated by *LMNB2,* with the further requirement for any one of several secondary associated illnesses or conditions. The findings are consistent with possible molecular genetic heterogeneity for APL, since five of nine patients did not have an *LMNB2* mutation. The findings extend the range of phenotypes associated with mutations in nuclear envelope components and increase the number of nuclear envelope components that are associated with disease. The findings also implicate a second nuclear lamin gene in a human lipodystrophy syndrome and suggest that other acquired lipodystrophy phenotypes, such as HIV-related or drug-induced lipodystrophies, may be associated with mutations affecting the nuclear lamina or interacting proteins. Finally, the findings suggest that reannotation of known genes will disclose new coding sequences that should be examined by genomic sequence analysis to search for new disease-associated mutations.

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Web Resources

Accession numbers and URLs for data presented herein are as follows:

GenBank, http://www.ncbi.nlm.nih.gov/Genbank/ (for *LMNB2* 2001 reference sequence [accession numbers M94362 and M94363] and 2005 reference sequence [accession numbers NT_011255 and NM_032737])

Online Mendelian Inheritance in Man (OMIM), http://www.ncbi.nlm.nih.gov/Omim/ (for *LMNA,* APL, FPLD2, *LMNB2,* CGL, FPLD3, Emery-Dreifuss muscular dystrophy, Pelger-Huet anomaly, and hydrops-ectopic calcification-moth-eaten skeletal dysplasia)

References

1. Garg A (2004) Acquired and inherited lipodystrophies. N Engl J Med 350:1220–1234
2. Hegele RA (2004) Phenomics, lipodystrophy, and the metabolic syndrome. Trends Cardiovasc Med 14:133–137
3. Capeau J, Magre J, Lascols O, Caron M, Bereziat V, Vigouroux C, Bastard JP (2005) Diseases of adipose tissue: genetic and acquired lipodystrophies. Biochem Soc Trans 33:1073–1077
4. Hegele RA (2005) Lessons from human mutations in PPARγ. Int J Obes (Lond) Suppl 1 29:S31–S35
5. Misra A, Garg A (2003) Clinical features and metabolic derangements in acquired generalized lipodystrophy: case reports and review of the literature. Medicine (Baltimore) 82: 129–146
6. Misra A, Peethambaram A, Garg A (2004) Clinical features and metabolic and autoimmune derangements in acquired partial lipodystrophy: report of 35 cases and review of the literature. Medicine (Baltimore) 83:18–34
7. Hegele RA, Yuen J, Cao H (2001) Single-nucleotide polymorphisms of the nuclear lamina proteome. J Hum Genet 46: 351–354
8. Frey BJ, Mohammad N, Morris QD, Zhang W, Robinson MD, Mnaimneh S, Chang R, Pan Q, Sat E, Rossant J, Bruneau BG, Aubin JE, Blencowe BJ, Hughes TR (2005) Genome-wide analysis of mouse transcripts using exon microarrays and factor graphs. Nat Genet 37:991–996
9. Cao H, Hegele RA (2000) Nuclear lamin A/C R482Q mutation in Canadian kindreds with Dunnigan-type familial partial lipodystrophy. Hum Mol Genet 9:109–112
10. Bhayana S, Siu VM, Joubert GI, Clarson CL, Cao H, Hegele RA (2002) Cardiomyopathy in congenital complete lipodystrophy. Clin Genet 61:283–287
11. Hegele RA, Cao H, Frankowski C, Mathews ST, Leff T (2002) PPARG F388L, a transactivation-deficient mutant, in familial partial lipodystrophy. Diabetes 51:3586–3590
12. Barraquer-Ferre L (1935) Lipodystrophie progressive: syndrome de Barraquer-Simons. Presse Med 86:1672–1674
13. Langhof H, Zabel R (1960) On lipodystrophia progressiva (follow-up study of the case published by A. Simons in 1911 for the first time in German literature) [in German]. Arch Klin Exp Dermatol 210:313–321
14. Jeune M, Freycon MT, Hermier M, Lamit HJ, Abboud N, Brunat N (1965) Nephropathies in progressive lipodystrophy or Barraquer-Simons disease [in French]. Ann Pediatr (Paris) 12:749–758
15. Biasi D, Caramaschi P, Carletto A, Bambara LM (1999) A case of acquired partial lipodystrophy associated with localized scleroderma and undifferentiated connective tissue disease. Rheumatol Int 19:75–76
16. Quecedo E, Febrer I, Serrano G, Martinez-Aparicio A, Aliaga A (1996) Partial lipodystrophy associated with juvenile dermatomyositis: report of two cases. Pediatr Dermatol 13:477–482
17. Ferrarini A, Milani D, Bottigelli M, Cagnoli G, Selicorni A (2004) Two new cases of Barraquer-Simons syndrome. Am J Med Genet A 126:427–429
18. Reichel W, Kobberling J, Fischbach H, Scheler F (1976) Membranoproliferative glomerulonephritis with partial lipodystrophy: discordant occurrence in identical twins. Klin Wochenschr 54:75–81
19. Bier DM, O'Donnell JJ, Kaplan SL (1978) Cephalothoracic lipodystrophy with hypocomplementemic renal disease: discordance in identical twin sisters. J Clin Endocrinol Metab 46:800–807
20. Omary MB, Coulombe PA, McLean WH (2004) Intermediate filament proteins and their associated diseases. N Engl J Med 351:2087–2100
21. Worman HJ (2005) Components of the nuclear envelope and their role in human disease. Novartis Found Symp 264:35–42
22. Hegele R (2005) LMNA mutation position predicts organ system involvement in laminopathies. Clin Genet 68:31–34
23. Tribioli C, Biamonti G, Giacca M, Colonna M, Riva S, Falaschi

A (1987) Characterization of human DNA sequences synthesized at the onset of S-phase. Nucleic Acids Res 15:10211–10232
24. Hoger TH, Zatloukal K, Waizenegger I, Krohne G (1990) Characterization of a second highly conserved B-type lamin present in cells previously thought to contain only a single B-type lamin. Chromosoma 99:379–390
25. Biamonti G, Giacca M, Perini G, Contreas G, Zentilin L, Weighardt F, Guerra M, Della Valle G, Saccone S, Riva S, Falaschi A (1992) The gene for a novel human lamin maps at a highly transcribed locus of chromosome 19 which replicates at the onset of S-phase. Mol Cell Biol 12:3499–3506
26. Giacca M, Zentilin L, Norio P, Diviacco S, Dimitrova D, Contreas G, Biamonti G, Perini G, Weighardt F, Riva S, Falaschi A (1994) Fine mapping of a replication origin of human DNA. Proc Natl Acad Sci USA 91:7119–7123
27. Vergnes L, Peterfy M, Bergo MO, Young SG, Reue K (2004) Lamin B1 is required for mouse development and nuclear integrity. Proc Natl Acad Sci USA 101:10428–10433
28. Kusic J, Kojic S, Divac A, Stefanovic D (2005) Noncanonical DNA elements in the lamin B2 origin of DNA replication. J Biol Chem 280:9848–9854