Available online at www.sciencedirect.com
ScienceDirect
Blood Cells, Molecules, and Diseases 39 (2007) 119–123
BLOOD CELLS, MOLECULES, & DISEASES
www.elsevier.com/locate/ybcmd

Genetic and clinical features of patients with Gaucher disease in Hungary

Melinda Erdős [a,1], Katerina Hodanova [b,1], Szilvia Taskó [a], Anita Palicz [a], Larisa Stolnaja [b], Lenka Dvorakova [b], Martin Hrebicek [b], László Maródi [a,*]

[a] *Department of Infectious and Pediatric Immunology, Medical and Health Science Center, University of Debrecen, Debrecen, Hungary*
[b] *Institute of Metabolic Disorders, Charles University, First Faculty of Medicine, Prague, Czech Republic*

Submitted 28 January 2007; revised 15 February 2007
Available online 28 March 2007
(Communicated by E. Beutler, M.D., 16 February 2007)

Abstract

The aim of this study was to identify mutations in the gene encoding for lysosomal β-glucocerebrosidase (GBA; gene symbol, *GBA*) in Hungarian patients with Gaucher disease (GD), and to study genotype-phenotype relationships. Genotypes and allele variations in 27 patients with type I GD of 25 unrelated families were studied. Of the 54 mutant alleles, we detected 38 frequent (N370S, 22/54; Rec*Nci*I, 8/54; L444P, 8/54) and 9 rare (N188S, R257Q, R285C, G377S, R120W, T323I, 84GG, 1263–1317del and 1263–1317del/RecTL) mutations. In addition, we identified two novel mutations. The N370S/Rec*Nci*I genotype found in 8 patients and the N370S/L444P genotype found in 5 patients were the most frequent genotypes in this cohort. In 22 patients the mutations occurred in heterozygosity with the N370S sequence variant, and one patient was homozygous for the L444P mutation. These data suggest that N370S, Rec*Nci*I, and L444P are the most prevalent mutations in Hungarian patients with GD. This mutation profile is characteristic for a Caucasian (non-Jewish) population. The c.260G>A and c.999G>A missense mutations are described here for the first time in GD patients contributing to the panel of reported *GBA* mutations.

Keywords: Gaucher disease; β-glucocerebrosidase; *GBA* gene

Introduction

Gaucher disease (GD, MIM# 230800, 230900, 231000) is an autosomal recessive storage disorder caused by mutations in the gene coding for the acid β-glucocerebrosidase (GBA) [1]. *GBA*, the gene mutated in GD was mapped to 1q21 [1]; it consists of 11 exons encoding a protein of 497 amino acids [1–3]. More than 200 different mutations were described, and six of them (N370S, L444P, IVS2+1, 84GG, Rec*Nci*I, and RecTL) are especially common [3]. Whole gene deletions and gene-pseudogene fusions have also been identified in patients with GD [2–4]. Recombination events between the gene and its pseudogene lead to the formation of complex Rec alleles that carry two or more mutations normally present in the pseudogene. In patients with some genotypes, it is possible to draw predictions concerning phenotypes. Among the widely accepted genotype/phenotype correlations is the association of N370S heterozygosity or homozygosity with non-neuronopathic course of the disease, whereas the L444P homozygous mutation is associated with the neuronopathic type III GD. Homozygosity for the D409H is associated with a specific form of type III disease with valvular heart disease and aortic calcifications.

The mutation profiles causing GD are different in Jewish, Caucasian, Arabian, and Japanese populations [5–20]. N370S, 84GG, IVS2+1, and R496H are the predominant mutations in Ashkenazi Jews. The 84GG sequence variant was found exclusively in the Ashkenazi Jewish population, and together with the N370S mutation, they account for 83% of all mutant alleles [6]. Norrbottnian Swedes with type III GD are homozygous for the L444P mutation [9]. The N370S mutation has not been found in Japanese patients, in contrast to the L444P mutation that is the most common mutation in this ethnic group [14–16].

The N370S, L444P, Rec*Nci*I, RecTL, D409H, and IVS2+1 mutations altogether represent 60–70% of mutant alleles in

* Corresponding author. Fax: +36 52 430 323.
E-mail address: lmarodi@jaguar.dote.hu (L. Maródi).
[1] ME and KH contributed equally to this work. MH and LM share senior authorship.

1079-9796/$ - see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.bcmd.2007.02.005

the Caucasian population [12,13,20]. There are only a few reports on *GBA* mutations in East-European patients with GD [10,13,19,20]. In Polish patients the prevalence of the L444P mutation is relatively high explaining the high incidence of type III GD with severe visceral involvement [20]. We first report here the mutation profile of 27 Hungarian patients with GD, and describe genotype/phenotype associations. Mutation spectrum was compared with that found in other European populations.

Materials and methods

Patients

We studied genotypes and allele variations in 27 patients of 25 unrelated families diagnosed with type I GD. Based on history, only one patient was identified to have Jewish ancestry. The diagnosis of GD was made by analysis of clinical, hematological, and radiological findings, and by genomic DNA sequencing. Analysis of *GBA* was performed at both the Charles University in Prague and at the University of Debrecen in Debrecen. All studies were approved by the institutional review boards. Informed consent was obtained from the patients or parents.

DNA sequencing

EDTA blood from the patients and family members was obtained and genomic DNA was isolated by using QIAamp DNA Blood Mini kit (QIAGEN GmbH, Hilden, Germany). Long-range PCR was performed by using 3 primer pairs to selectively amplify products of the functional GBA gene. Exons 1–11 and the flanking intron regions were amplified by using long-range PCR products as templates. Primer sequences are available on request.

Amplified segments were purified using a MICROCON YM-100 Centrifugal Filter Devices (Millipore Co., Bedford, MA, USA). Mutational analysis was performed using the BigDye Terminator Cycle sequencing kit (Applied Biosystems, Foster City, CA) and an ABI PRISM 3130 Genetic Analyzer (Applied Biosystems, Foster City, CA). In Prague, the analysis of mutation was done as described previously [13]. Both novel mutations were confirmed from an independent sample of DNA and confirmed by direct sequencing.

GD research community uses preferentially trivial description of protein sequence variants—for this reason the trivial nomenclature was used in the body of the article with the amino acid sequence numbering of the mature protein after cleavage

Table 1
Demography, genotypes, and mutated exons

Pt no	Sex	Age (years)	cDNA sequence variant		Trivial allele name		Systematic allele name†		Exons affected	
			A1	A2	A1	A2	A1	A2	A1	A2
1	F	67	c.84G>GG	c.1226A>G	84GG	N370S	p.Leu29AlafsX18	p.Asn409Ser	2	9
2	M	30	**c.260G>A**	c.1085C>T	**R48Q**	T323I	**p.Arg87Gln**	p.Thr362Ile	3	8
3	M	40	c.475C>T	c.1226A>G	R120W	N370S	p.Arg159Trp	p.Asn409Ser	5	9
4	M	29	c.475C>T	c.1226A>G	R120W	N370S	p.Arg159Trp	p.Asn409Ser	5	9
5	F	16	c.680A>G	c.970C>T	N188S	R285C	p.Asn227Ser	p.Arg324Cys	6	7
6	F	5	c.680A>G	c.970C>T	N188S	R285C	p.Asn227Ser	p.Arg324Cys	6	7
7	M	23	c.887G>A	c.1226A>G	R257Q	N370S	p.Arg296Gln	p.Asn409Ser	7	9
8	M	67	**c.999G>A**	c.1226A>G	**Splicing defect**⌘	N370S	–	p.Asn409Ser	7	9
9	F	26	c.1226A>G	c.1263–1317del	N370S	1263del55	p.Asn409Ser	p.Leu422ProfsX4	9	9
10	M	26	c.1226A>G	c.1263–1317del	N370S	1263del55	p.Asn409Ser	p.Leu422ProfsX4	9	9
11	F	75	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
12	F	36	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
13	M	5	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
14	F	13	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
15	F	35	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
16	F	40	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
17	M	56	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
18	M	31	c.1226A>G	Rec*Nci*I[#]	N370S	Rec*Nci*I[@]	p.Asn409Ser	p.Leu483Pro; p.Ala495Pro	9	10
19	F	44	c.1226A>G	c.1448T>C	N370S	L444P	p.Asn409Ser	p.Leu483Pro	9	10
20	F	44	c.1226A>G	c.1448T>C	N370S	L444P	p.Asn409Ser	p.Leu483Pro	9	10
21	M	38	c.1226A>G	c.1448T>C	N370S	L444P	p.Asn409Ser	p.Leu483Pro	9	10
22	F	25	c.1226A>G	c.1448T>C	N370S	L444P	p.Asn409Ser	p.Leu483Pro	9	10
23	F	40	c.1226A>G	c.1448T>C	N370S	L444P	p.Asn409Ser	p.Leu483Pro	9	10
24	F	38	c.1226A>G	c.1263–1317del; RecTL[§]	N370S	1263del55; RecTL[◇]	p.Asn409Ser	p.Leu422ProfsX4	9	From 9
25	F	35	c.1226A>G	c.1263–1317del; RecTL[§]	N370S	1263del55; RecTL[◇]	p.Asn409Ser	p.Leu422ProfsX4	9	From 9
26	M	21	c.1246G>A	c.1448T>C	G377S	L444P	p.Gly416Ser	p.Leu483Pro	9	10
27	M	20	c.1448T>C	c.1448T>C	L444P	L444P	p.Leu483Pro	p.Leu483Pro	10	10

The amino acid sequence numbering is that of the mature protein, after cleavage of the leader sequence. Patients 5 and 6 and Patients 24 and 25 are siblings. Novel mutations are shown in bold. A1 and A2, alleles; [#]c.1448T>C+c.1483G>C+c.1497G>C; [@]L444P+A456P+V460V; [§]c.1342G>C, c.1448T>C+c.1483G>C+c.1497G>C; [◇]D409H+L444P+A456P+V460V; [⌘]RNA from the patient was not available for analysis and splicing defect was predicted (see text); [†]predicted effect on protein.