

Supplemental Data

Deleterious Mutations in *LRBA* Are Associated with a Syndrome of Immune Deficiency and Autoimmunity

Gabriela Lopez-Herrera, Giacomo Tampella, Qiang Pan-Hammarström, Peer Herholz, Claudia M Trujillo-Vargas, Kanchan Phadwal, Anna Katharina Simon, Michel Moutschen, Amos Etzioni, Adi Mory, Izhak Srugo, Doron Melamed, Kjell Hultenby, Chonghai Liu, Manuela Baronio, Massimiliano Vitali, Pierre Philippet, Vinciane Dideberg, Asghar Aghamohammadi, Nima Rezaei, Victoria Enright, Likun Du, Ulrich Salzer, Hermann Eibel, Dietmar Pfeifer, Hendrik Veelken, Hans Stauss, Vassilios Lougaris, Alessandro Plebani, E. Michael Gertz, Alejandro A. Schäffer, Lennart Hammarström, and Bodo Grimbacher

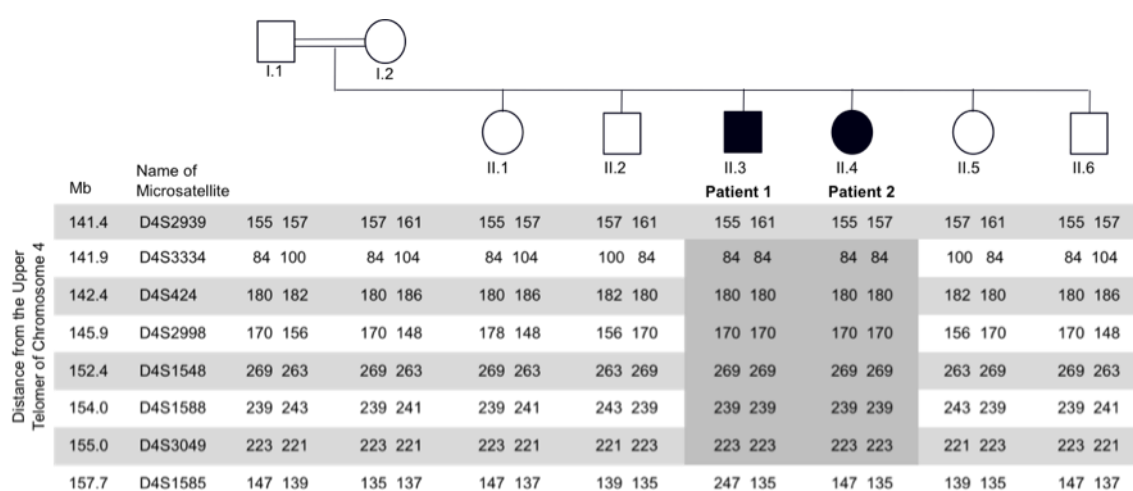


Figure S1. Genotyping in Family A

Microsatellite markers located on chromosome 4 between 141.4 and 157.7 Mb were genotyped, and a perfectly segregating interval with homozygous markers in the 2 affected siblings was detected from 141.9 to 155.0 Mb (shaded box). All positions are shown according to human genome build 36.

```

                                ↓
HUMAN004275  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETAAPRAILTGHDEVTCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
MOUSE020048  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG GETATPRAILTGHDEVTITCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
CANFA010295
RATNO012623
TEING015968  CGFWDKSFVYSDSGLKTIQVFGHRDVVSLARSESYIGGDCYII SGRSDATLLWYNGKYNISGESFGR EFTTPRAILTGHDEVTCAVCAELGLVLSGCK_EGPCLIHSMNGDLLRTLE  APEG
TAKRU016354  CGFWDKSFVYSDSGLKTIQVFGHRDVVSLARSESYIGGDCYII SGRSDATLLWYNGKHNSIGESFGTGETTPRAILTGHDEVTCAVCAELGLVLSGCK_EGPCLIHSMNGDLLRTLE  APED
BOVIN008350
FANTR003805  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETAAPRAILTGHDEVTCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
MACMU006577  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSDSYIGGNCYII SGRSDATLLWYNGKCSVIGDTQAVSETAAPRAILTGHDEVTCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
LOMAF002353  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
RABIT004921  GGFWKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTCAAVCAELGLVLSGSK_EAPCLHSMNGDLLRTLE  GFEN
ECHTE005340  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GPEK
GASAC005142  CGFWDKSFVYSDSGLKTIQVFGHRDVVSLARSESYIGGDCYII SGRSDATLLWYNGKLNISGESFGT EFTTPRAILTGHDEVTCAVCAELGLVLSGCK_EGPCLIHSMNGDLLRTLE  GPER
FELCA005301  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SEFTTPRAILTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
ERIEU010356
OTOGA012591  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
SPETR010492  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
TUEGB002455  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
MYOLU002717  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
MYOLU002717  XKSIGIGD PG_SGTTTPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
CAVFO002522  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
SORAR002358  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
MICMU009295  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETATPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
OCHFR013850  GGFWKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETAAPRAILTGHDEVTITCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
POMAB013499  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETAAPRAILTGHDEVTITCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN
HORSE010762
TURTR012072  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
PTEVA008686  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKTSIGIGDNFG XGPCLIHSMNGDLLRTLE  GFEN
LAMPF009214  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDA LLLWY NGKCSGIGDNFG SETAAPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
PROCA012703  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GPEH
DIFOR003498  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFGH
CHOR010522  XKLIVQVVFQHWVVTCLTRSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG CETTPPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GFEN
ANOCA016601  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKTNISIGDYA NGDSATPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  PFEN
TAEGU003356  CGFWDKSFVYSDSGLKTIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKTNISIGDYA NGDSATPRAVLTGHDEVTITCAAVCAELGLVLSGSK_EGPCLIHSMNGDLLRTLE  GPERLQSPES
CALJA023279  CGFWDKSFVYSDTGRLIQVVFQHWVVTCLARSESYIGGNCYII SGRSDATLLWYNGKCSGIGDNFG SETAAPRAVLTGHDEVTITCAAVCAELGLVLSGSQ_EGPCLIHSMNGDLLRTLE  GFEN

```

Figure S2. Multiple Alignment of LRBA from Several Species

Conserved sequence surrounding I2657 (black arrow) is shown in yellow. Multiple alignment was prepared using the tool <http://omabrowser.org/> (OMA group 316837).

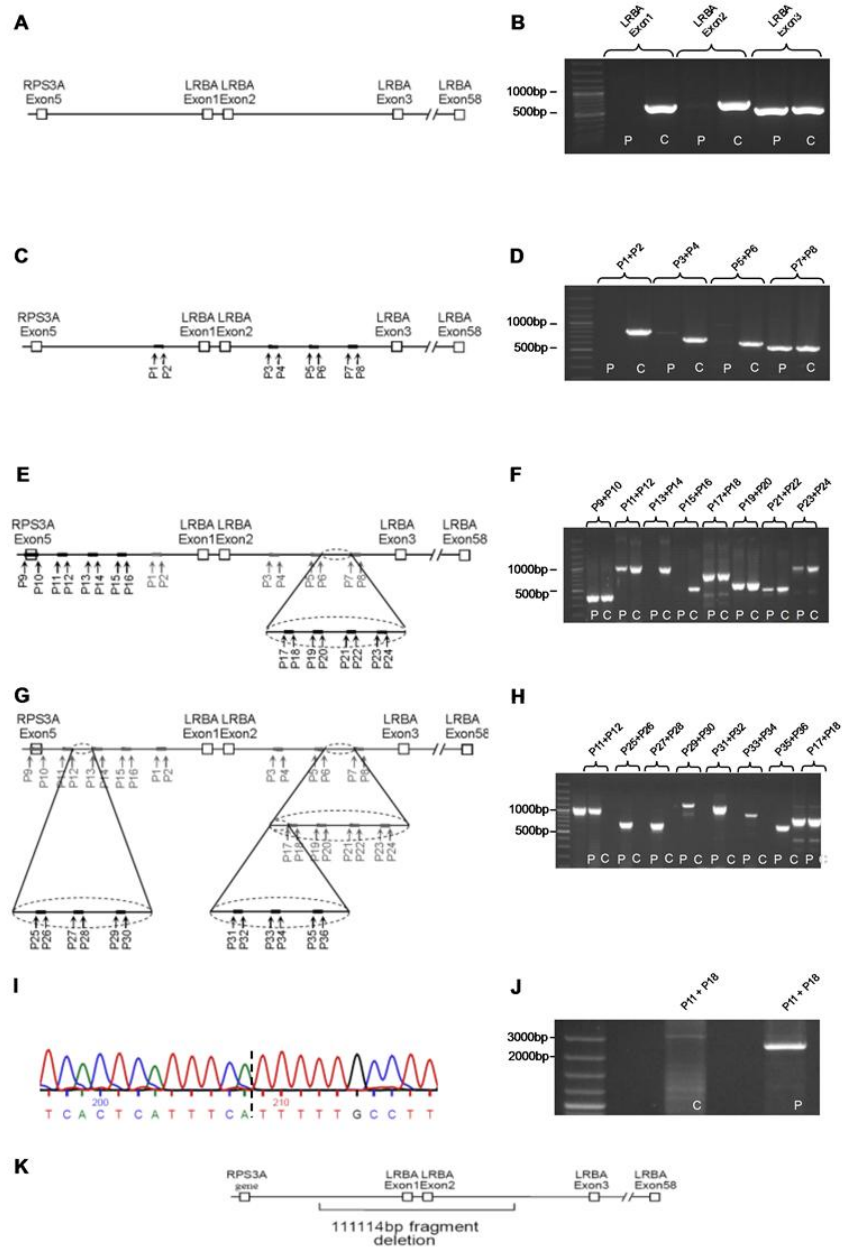


Figure S3. Large Homozygous Deletion in P5

Schematic representation and PCR products when the primers designed to map the deletion were used.

(A and B) PCR product from exons 1 and 2 in individual P5 were not detected.

(C and D) Amplification using primer pairs P1-P2 located about 20kb upstream of *LRBA* exon 1 and P3-P4, P5-P6 and P7-P8 which are located between exon 2 and exon 3.

(E and F) Primers pairs P9-P10 were designed to amplify the last exon (exon 5) of the upstream gene *RPS3A*, and P11-P12, P13-P14 and P15-P16 amplify between P10 and P1. P17-P18, P19-P20, P21-P22 and P23-P24 were used to amplify selected regions downstream of P6 and upstream of P7.

(G and H) Primers pairs P25-P26, P27-P28 and P29-P30 were subsequently used to amplify regions between P12 and P13 and primers P31-P32, P33-P34 and P35-P36 were distributed in the region between P6 and P17. (I, J) Direct sequencing of a 2.5 kb product amplified with P11 and P18 (the exact breakpoint of the deletion is indicated by dashed line). (K) Schematic representation of the region deleted (K).

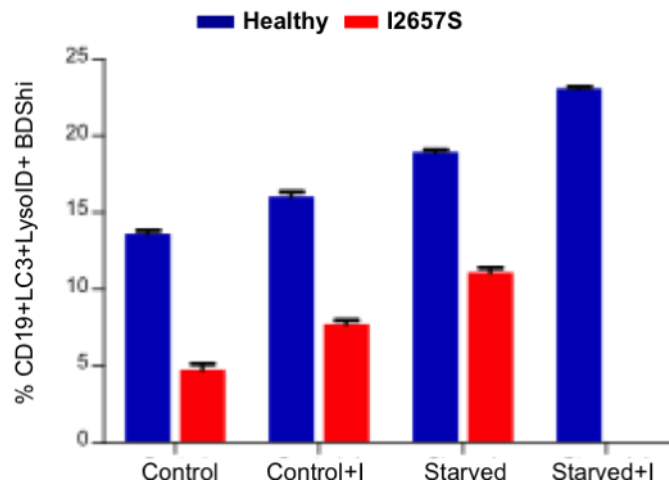


Figure S4. Defective Autophagy in an Individual with a Homozygous I2657S Mutation

Error bars are shown as computed by the Image Stream software based on 20,000 cells.

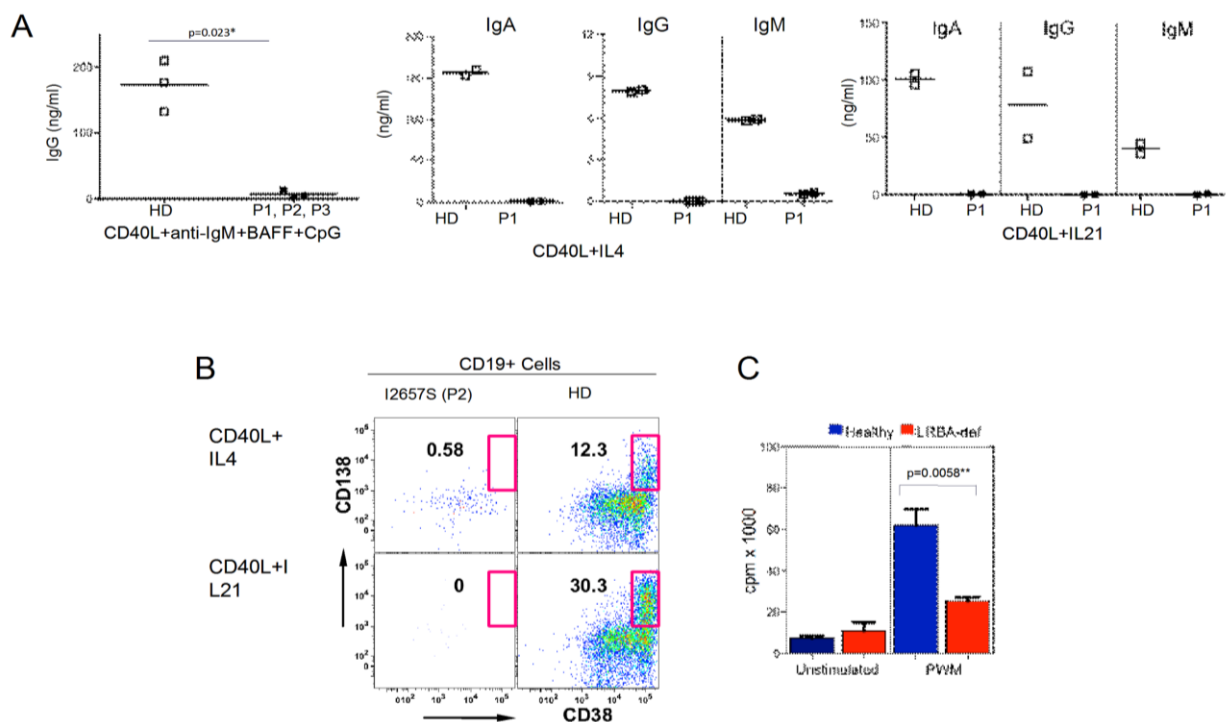


Figure S5. Defective Cell Activation in LRBA-Deficient B Cells

(A) Left, naïve B cells were isolated from total PBMCs and stimulated for 13 days with 2.5 μ g/ml anti-IgM, 1 μ g/ml rCD40L, 2.5 μ g/ml CpG ODN and 75ng/ml recombinant BAFF. After incubation, the concentration of IgG in the supernatant of stimulated and unstimulated cells was determined by ELISA. Data from individuals P1, P2 and P3, from two unrelated healthy controls (one of whom was a travel control whose blood was taken in Belgium at the same time as P3) and from one healthy heterozygous individual are shown. In a second approach, PBMCs were activated by adding soluble adiponectin-CD40L combined with IL21-Fc or IL4 as described previously⁴: IgA, IgG and IgM secretion after (middle) C40L+IL4, or (right) CD40L+IL21 stimulation in PBMCs cultures (duplicates from a healthy donor and P1 are shown).

(B) Plasmablast differentiation in Ile2657Ser cells. PBMCs were stimulated with CD40L+IL4 (top) or CD40L+IL21 (bottom) for 7 days. After incubation, plasmablast differentiation was determined by the expression of CD19+BAFFRloCD138hiCD38hi.

(C) Proliferative responses (obtained from P3 and P4) determined by 3H-Thymidine incorporation after 72 hours of stimulation with 10 μ g/ml PWM. The data are shown as the mean +/- for triplicate assays from two independent experiment for patient and control, the error bars were computed with Prism software.

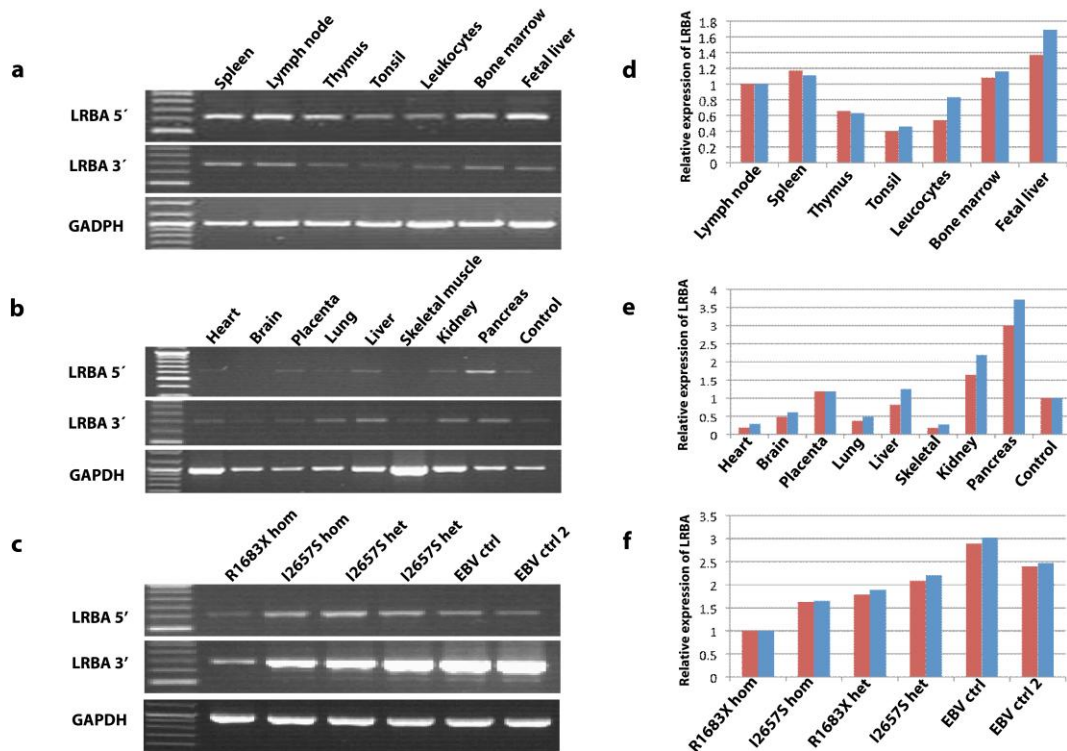


Figure S6. Gene Expression of *LRBA* in Different Tissues

RT-PCR was performed to detect either the 5' or the 3' of *LRBA* transcript.

(A–C) RT-PCR was performed to determine the expression of *LRBA* mRNA at the 5' (exons 1-3) or 3' (exons 54-58).

(D–F) Real-time RT PCR was performed to measure the relative expression level of gene transcript encompassing exon 4 and 5 border (red bars) or exon 26 and 27 border (blue bars) was determined.

(A and D) Immune system panel (human immune system MTC panel, Clontech), (B and E) non immune system panel (human MTC panel I, Clontech), (C and F) mRNA expression in EBV cell lines from *LRBA*-deficient individuals and healthy controls.

Table S1. Genes or Predicted Genes that Were Positional Candidates for Family A

Chr. 4:

1. *SCOC* short coiled-coil protein
2. *CLGN* calmegin
3. *LOC152586* similar to RIKEN cDNA 4933434I
4. *ELMOD2* ELMO/CED-12 domain containing 2
5. *UCP1* uncoupling protein 1 (mitochondrial, proton carrier)
6. *TBC1D9* TBC1 domain family, member 9 (with GRAM domain)
7. *RNF150* ring finger protein 150
8. *ZNF330* zinc finger protein 330
9. **IL15* interleukin 15
10. *INPP4B* inositol polyphosphate-4-phosphatase, type II, 105kDa
11. *FLJ44477* FLJ44477 protein
12. **USP38* ubiquitin specific peptidase 38
13. **GAB1* GRB2-associated binding protein 1
14. *LOC100128055* hypothetical protein LOC100128055
15. *SMARCA5* SWI/SNF related, matrix associated, actin dependent regulator of chromatin, subfamily a, member 5
16. *LOC100129551* hypothetical LOC100129551
17. *FREM3* FRAS1 related extracellular matrix 3
18. *GYPE* glycophorin E
19. *GYPB* glycophorin B
20. *GYPB* glycophorin A
21. *LOC646576* hypothetical LOC646576
22. *HHIP* hedgehog interacting protein
23. *ANAPC10* anaphase promoting complex subunit 10
24. **ABCE1* ATP-binding cassette, sub-family E (OABP), member 1
25. *OTUD4* OTU domain containing 4
26. *LOC100131639* hypothetical LOC100131639
27. **SMAD1* SMAD family member 1
28. *MMAA* methylmalonic aciduria (cobalamin deficiency) cblA type
29. *C4orf51* chromosome 4 open reading frame 51
30. *ZNF827* zinc finger protein 827
31. *LSM6* LSM6 homolog, U6 small nuclear RNA associated (*S. cerevisiae*)
32. *hCG_38984* hCG38984
33. *SLC10A7* solute carrier family 10 (sodium/bile acid cotransporter family), member 7
34. *POU4F2* POU class 4 homeobox 2
35. *TTC29* tetratricopeptide repeat domain 29
36. *EDNRA* endothelin receptor type A
37. *TMEM184C* transmembrane protein 184C
38. *PRMT10* protein arginine methyltransferase 10 (putative)
39. *LOC100129572* hypothetical protein LOC100129572
40. *ARHGAP10* Rho GTPase activating protein 10
41. *NR3C2* nuclear receptor subfamily 3, group C, member 2
42. *LOC285423* hypothetical LOC285423
43. *DCLK2* doublecortin-like kinase 2
44. *LOC729558* hypothetical protein LOC729558

45. *MAB21L2* mab-21-like 2 (*C. elegans*)
46. *LOC729566* similar to hCG1645309
47. **LRBA* LPS-responsive vesicle trafficking, beach and anchor containing
48. *RPS3A* ribosomal protein S3A
49. *SNORD73A* small nucleolar RNA, C/D box 73A
50. *SH3D19* SH3 domain containing 19
51. *ESSPL* epidermis-specific serine protease-like protein
52. *FAM160A1* family with sequence similarity 160, member A1
53. *PET112L* PET112-like (yeast)
54. *FBXW7* F-box and WD repeat domain containing 7
55. *DKFZP434I0714* hypothetical protein DKFZP434I0714
56. *TMEM154* transmembrane protein 154
57. *TIGD4* tigger transposable element derived 4
58. *ARFIP1* ADP-ribosylation factor interacting protein 1
59. *LOC729870* hypothetical LOC729870
60. *FHDC1* FH2 domain containing 1
61. *TRIM2* tripartite motif-containing 2
62. *MND1* meiotic nuclear divisions 1 homolog (*S. cerevisiae*)
63. *KIAA0922* KIAA0922
64. **TLR2* toll-like receptor 2
65. *RNF175* ring finger protein 175
66. *SFRP2* secreted frizzled-related protein 2
67. *DCHS2* dachsous 2 (*Drosophila*)
68. *PLRG1* pleiotropic regulator 1 (PRL1 homolog, *Arabidopsis*)
69. *FGB* fibrinogen beta chain
70. *LOC100129857* hypothetical LOC1001298
71. *FGA* fibrinogen alpha chain
72. *FGG* fibrinogen gamma chain
73. *LRAT* lecithin retinol acyltransferase (phosphatidylcholine--retinol O-acyltransferase)
74. *RBM46* RNA binding motif protein 46
75. *NPY2R* neuropeptide Y receptor Y2
76. *MAP9* microtubule-associated protein 9
77. *GUCY1A3* guanylate cyclase 1, soluble, alpha 3
78. *GUCY1B3* guanylate cyclase 1, soluble, beta 3
79. *ACCN5* amiloride-sensitive cation channel 5, intestinal
80. *TDO2* tryptophan 2,3-dioxygenase
81. **CTSO* cathepsin O

Chr. 5

1. *STK10* serine/threonine kinase 10
2. *LOC285588* hypothetical LOC285588
3. **UBTD2* ubiquitin domain containing 2
4. *FLJ40453* hypothetical LOC401217
5. *SH3PXD2B* SH3 and PX domains 2B
6. *DUSP1* dual specificity phosphatase 1
7. *ERGIC1* endoplasmic reticulum-golgi intermediate compartment (ERGIC) 1
8. *RPL26L1* ribosomal protein L26-like 1

9. *ATP6V0E1* ATPase, H⁺ transporting, lysosomal 9kDa, V0 subunit e1
10. *SNORA74B* small nucleolar RNA, H/ACA box 74B
11. *C5orf41* chromosome 5 open reading frame 41
12. *BNIP1* BCL2/adenovirus E1B 19kDa interacting protein 1
13. *NKX2-5* NK2 transcription factor related, locus 5 (Drosophila)
14. *STC2* stanniocalcin 2
15. *FAM44B* family with sequence similarity 44, member B
16. *LOC729170* hypothetical protein LOC729170
17. *CPEB4* cytoplasmic polyadenylation element binding protein 4
18. *LOC133491* hypothetical protein LOC133491
19. *HMP19* HMP19 protein
20. *GAPDHL16* glyceraldehyde-3-phosphate dehydrogenase-like 16
21. *MSX2* msh homeobox 2
22. *FLJ16171* FLJ16171 protein
23. *DRD1* dopamine receptor D1
24. *SFXN1* sideroflexin 1
25. **HRH2* histamine receptor H2
26. *CPLX2* complexin 2
27. *THOC3* THO complex 3
28. *LOC728334* hypothetical protein LOC728334
29. *LOC202134* hypothetical protein LOC202134
30. *LOC643201* hypothetical protein LOC643201
31. *C5orf25* chromosome 5 open reading frame 25
32. *KIAA1191* KIAA1191
33. *ARL10* ADP-ribosylation factor-like 10
34. *HSPC111* hypothetical protein HSPC111
35. *HIGD2A* HIG1 domain family, member 2A
36. *CLTB* clathrin, light chain (Lcb
37. *UBXD8* UBX domain containing 8
38. *RNF44* ring finger protein 44
39. *PCLKC* protocadherin LKC
40. *GPRIN1* G protein regulated inducer of neurite outgrowth 1
41. *LOC729370* hypothetical protein LOC729370
42. *SNCB* synuclein, beta
43. *EIF4E1B* eukaryotic translation initiation factor 4E family member 1B
44. *TSPAN17* tetraspanin 17
45. *LOC729378* hypothetical protein LOC729378
46. *UNC5A* unc-5 homolog A (C. elegans)
47. *HK3* hexokinase 3 (white cell)
48. **UIMC1* ubiquitin interaction motif containing 1
49. *ZNF346* zinc finger protein 346
50. *FGFR4* fibroblast growth factor receptor 4
51. *NSD1* nuclear receptor binding SET domain protein 1
52. *RAB24* RAB24, member RAS oncogene family
53. **PRELID1* PRELI domain containing 1 (formerly PX19)
54. *MXD3* MAX dimerization protein 3
55. *LMAN2* lectin, mannose-binding 2
56. *RGS14* regulator of G-protein signaling 14
57. *SLC34A1* solute carrier family 34 (sodium phosphate), member 1
58. *PFN3* profilin 3

59. *F12* coagulation factor XII (Hageman factor)
60. *GRK6* G protein-coupled receptor kinase 6
61. *PRR7* proline rich 7 (synaptic)
62. *DBN1* drebrin 1
63. *PDLIM7* PDZ and LIM domain 7 (enigma)
64. *DOK3* docking protein 3
65. *DDX41* DEAD (Asp-Glu-Ala-Asp) box polypeptide 41
66. *FLJ10404* hypothetical protein FLJ10404
67. *TMED9* transmembrane emp24 protein transport domain containing 9
68. *B4GALT7* xylosylprotein beta 1,4-galactosyltransferase, polypeptide 7 (galactosyltransferase I)
69. *LOC202181* hypothetical protein LOC202181
70. *NY-REN-7* NY-REN-7 antigen
71. *LOC401222* hypothetical gene supported by BC036933
72. *LOC441119* hypothetical LOC441119
73. *PROP1* PROP paired-like homeobox 1
74. *LOC401220* hypothetical gene supported by BC036933
75. *LOC653316* hypothetical protein LOC653316
76. *LOC653314* similar to ribosomal protein L19
77. *LOC729503* hypothetical protein LOC729503
78. *N4BP3* Nedd4 binding protein 3
79. *LOC645848* hypothetical LOC645848
80. *RMND5B* required for meiotic nuclear division 5 homolog B (*S. cerevisiae*)

Chr. 12

1. *STAB2* stabilin 2
2. *NT5DC3* 5'-nucleotidase domain containing 3
3. *LOC253724* hypothetical LOC253724
4. *HSP90B1* heat shock protein 90kDa beta (Grp94), member 1
5. *C12orf73* chromosome 12 open reading frame 73
6. *TDG* thymine-DNA glycosylase
7. *GLT8D2* glycosyltransferase 8 domain containing 2
8. *LOC100131054* hypothetical protein LOC100131054
9. *HCFC2* host cell factor C2
10. *NFYB* nuclear transcription factor Y, beta
11. *TXNRD1* thioredoxin reductase 1
12. *LOC100130902* hypothetical protein LOC100130902
13. *EID3* EP300 interacting inhibitor of differentiation 3
14. *CHST11* carbohydrate (chondroitin 4) sulfotransferase 11
15. *SLC41A2* solute carrier family 41, member 2
16. *C12orf45* chromosome 12 open reading frame 45
17. *ALDH1L2* aldehyde dehydrogenase 1 family, member L2
18. *KIAA1033*
19. *APPL2* adaptor protein, phosphotyrosine interaction, PH domain and leucine zipper containing 2
20. *OCC-1* overexpressed in colon carcinoma-1
21. *NUAK1* NUAK family, SNF1-like kinase, 1
22. *LOC341378* similar to Golgi-associated microtubule-binding protein
23. *CKAP4* cytoskeleton-associated protein 4

24. *LOC100130905* hypothetical protein LOC100130905
25. *TCP11L2* t-complex 11 (mouse)-like 2
26. *POLR3B* polymerase (RNA) III (DNA directed) polypeptide B
27. *RFX4* regulatory factor X, 4 (influences HLA class II expression)
28. *RIC8B* resistance to inhibitors of cholinesterase 8 homolog B (*C. elegans*)
29. *C12orf23* chromosome 12 open reading frame 23
30. *MTERFD3* MTERF domain containing 3
31. *CRY1* cryptochrome 1 (photolyase-like)
32. *BTBD11* BTB (POZ) domain containing 11
33. *PWP1* PWP1 homolog (*S. cerevisiae*)
34. *PRDM4* PR domain containing 4
35. *ASCL4* achaete-scute complex homolog 4 (*Drosophila*)
36. *LOC100131660* hypothetical protein LOC100131660
37. *LOC100129448* hypothetical protein LOC100129448
38. *WSCD2* WSC domain containing 2
39. **CMKLR1* chemokine-like receptor 1
40. *FICD* FIC domain contains (was HYPE Huntingtin interacting protein E)
41. *SART3* squamous cell carcinoma antigen recognized by T cells 3
42. *ISCU* iron-sulfur cluster scaffold homolog (*E. coli*)
43. *TMEM119* transmembrane protein 119
44. *SELPLG* selectin P ligand
45. *CORO1C* coronin, actin binding protein, 1C
46. *SSH1* slingshot homolog 1 (*Drosophila*)
47. *MIRN619* microRNA 619
48. *DAO* D-amino-acid oxidase
49. *SVOP* SV2 related protein homolog (rat)
50. *LOC100131733* hypothetical protein LOC100131733
51. *USP30* ubiquitin specific peptidase 30
52. *ALKBH2* alkB, alkylation repair homolog 2 (*E. coli*)
53. *UNG* uracil-DNA glycosylase (mutated in Hyper-IgM type 5)
54. *ACACB* acetyl-Coenzyme A carboxylase beta
55. *FOXP4* forkhead box N4
56. *MYO1H* myosin 1H
57. *KCTD10* potassium channel tetramerisation domain containing 10
58. *UBE3B* ubiquitin protein ligase E3B
59. *MMAB* methylmalonic aciduria (cobalamin deficiency) cblB type
60. *MVK* mevalonate kinase (mevalonic aciduria)
61. *C12orf34* chromosome 12 open reading frame 34
62. *TRPV4* transient receptor potential cation channel, subfamily V, member 4
63. *GLTP* glycolipid transfer protein
64. *TCHP* trichoplein, keratin filament binding
65. *GIT2* G protein-coupled receptor kinase interactor 2
66. *ANKRD13A* ankyrin repeat domain 13A
67. *FLJ40142* FLJ40142 protein
68. *IFT81* intraflagellar transport 81 homolog (*Chlamydomonas*)
69. *ATP2A2* ATPase, Ca⁺⁺ transporting, cardiac muscle, slow twitch 2
70. *ANAPC7* anaphase promoting complex subunit 7
71. *ARPC3* actin related protein 2/3 complex, subunit 3, 21kDa
72. *ATPBD1C* ATP binding domain 1 family, member C
73. *LOC100129882* similar to mCG49427

74. *C12orf24* chromosome 12 open reading frame 24
75. *VPS29* vacuolar protein sorting 29 homolog (S. cerevisiae)
76. *RAD9B* RAD9 homolog B (S. cerevisiae)
77. **PPTC7* PTC7 protein phosphatase homolog (S. cerevisiae) (formerly T cell activation protein phosphatase 2C)
78. *TCTN1* tectonic family member 1
79. *HVCN1* hydrogen voltage-gated channel 1
80. *PPP1CC* protein phosphatase 1, catalytic subunit, gamma isoform
81. *CCDC63* coiled-coil domain containing 63
82. *MYL2* myosin, light chain 2, regulatory, cardiac, slow
83. *LOC100131138* similar to hCG2040918
84. *CUX2* cut-like homeobox 2 (Drosophila)
85. *FAM109A* family with sequence similarity 109, member A
86. *LOC642580* hypothetical LOC642580
87. **SH2B3* SH2B adaptor protein 3
88. *ATXN2* ataxin 2
89. *BRAP* BRCA1 associated protein
90. *ACAD10* acyl-Coenzyme A dehydrogenase family, member 10
91. *ALDH2* aldehyde dehydrogenase 2 family (mitochondrial)
92. *MAPKAPK5* mitogen-activated protein kinase-activated protein kinase 5
93. *TMEM116* transmembrane protein 116
94. *LOC728543* hypothetical protein LOC728543
95. *ERP29* endoplasmic reticulum protein 29
96. *C12orf30* chromosome 12 open reading frame 30
97. **TRAFD1* TRAF-type zinc finger domain containing 1
98. *C12orf51* chromosome 12 open reading frame 51
99. *LOC100133013* similar to PRO1477
100. *RPL6* ribosomal protein L6
101. *LOC728585* hypothetical LOC728585
102. *PTPN11* protein tyrosin phosphatase, non-receptor type 11
103. *RPH3A* rabphilin 3A homolog (mouse)
104. *OAS1* 2',5' oligoadenylate synthetase 1, 40/46 kDa
105. *OAS3* 2',5' oligoadenylate synthetase 3, 100 kDa
106. *OAS2* 2',5' oligoadenylate synthetase 2, 69/71 kDa
107. **DTX1* deltex homolog 1

The maximal interval on chromosome 4 contains 81 genes or predicted genes in human genome build 36. Genes numbered 6 through 66 are in the minimal interval consistent with linkage. Functional candidate genes sequenced in family A are indicated with a *. Genes in the perfect interval on chromosome 5 and in the near perfect interval on chromosome 12 are also listed.

Table S2. Primers and PCR Conditions Used for Mapping the Boundary of Deletion in P5

Primer ID	Sequence	Size (bp)	Ta (°C)
P1	GCAAACCTCCACAAAGAGATTTTCGTA	841	58
P2	GGTAATCTGGTTAGCCTTTGGAATG		
P3	CCTGAAAGGCTTGTCTGCTG	692	58
P4	TGAAGAAGAGGGTCATACAGAGAACT		
P5	ACTTTTCTGGTTTGGGTCTGTCA	630	58
P6	TGACACCTTGAGACAATGAAACC		
P7	GGGATGTGTTCTCTCCTCCTCAG	575	58
P8	CGCATAGTAAACCTAAAGCAAGCA		
P9	CAGACCAAGATGCCTGCTACAAT	420	58
P10	TGTGTAGTGGATGGACCGATGA		
P11	GAGACAGGACAGCAACTATGACTAAG	1087	58
P12	TACTCTCTTGTCCCTTGTCCCTTGC		
P13	GAAAGCCCATCTCCATTGAA	1006	58
P14	TTTCATCACCCCTAAAAATAATCCTA		
P15	TGAAACTACACTCAGAGGCAAAGA	509	58
P16	GATTATCTGAGGTTGGCAGTTTG58		
P17	GA CTCAGGTCTGTCAGTTCTTGG58	809	58
P18	TAACAGACTAAAGTAGGCTGGGTG58		
P19	GCAATGGACAAGTTTTATCTGGA58	591	58
P20	CTCAGTGTCGTGTCATCAGGAAT58		
P21	ATTATCCATTTGTATCCTTTGCTCA58	480	58
P22	GGAGAGGTCGGATTGTTATTGTG		
P23	TGTTCTTGTGTCCTCCGTATTTTC	912	58
P24	TTGCCATAAAAATAGCAGAAACAT		
P25	TGTTCTCAAATGTGGTAATAGGG	697	60
P26	TGGGATTTAGTGATACTCCTACCTT		
P27	GCTCACAATCATTTTTACAACCC	673	60
P28	AGTGATGCGTGCTTGTAGTATGA		
P29	ACCAAGACAGATGAGCAAAGAATA	1262	62
P30	AAATACAGGATGAGTTGAAGAAAGC		
P31	CTCATTGTTATTTTGCCTGCTG	1170	60
P32	GTGTCACATTCTCTTGGTTTCAG		
P33	TAACAGTTTTGGATTTTGTCTTCTAT	895	60
P34	TCCTCAACTCAACACAACAGAAAT		
P35	ATTTCTGTTGTGTTGAGTTGAGGA	633	60
P36	TGTGCTGGGCATTATTTTCAA		