**SAUER et al. 2012**

Defective B-cell tolerance in ADA-deficiency is corrected by gene therapy

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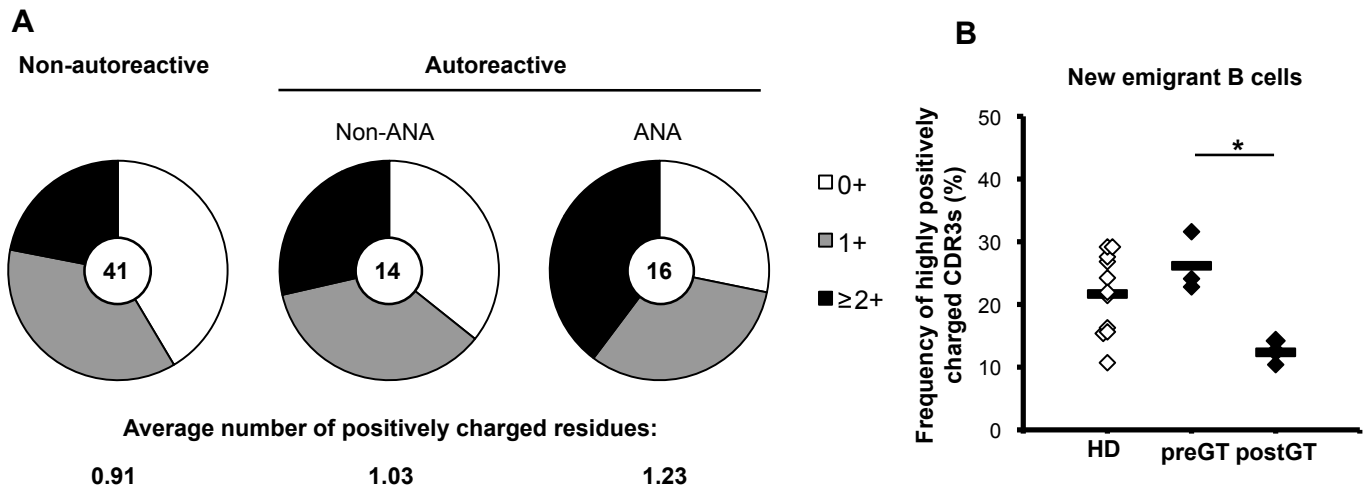
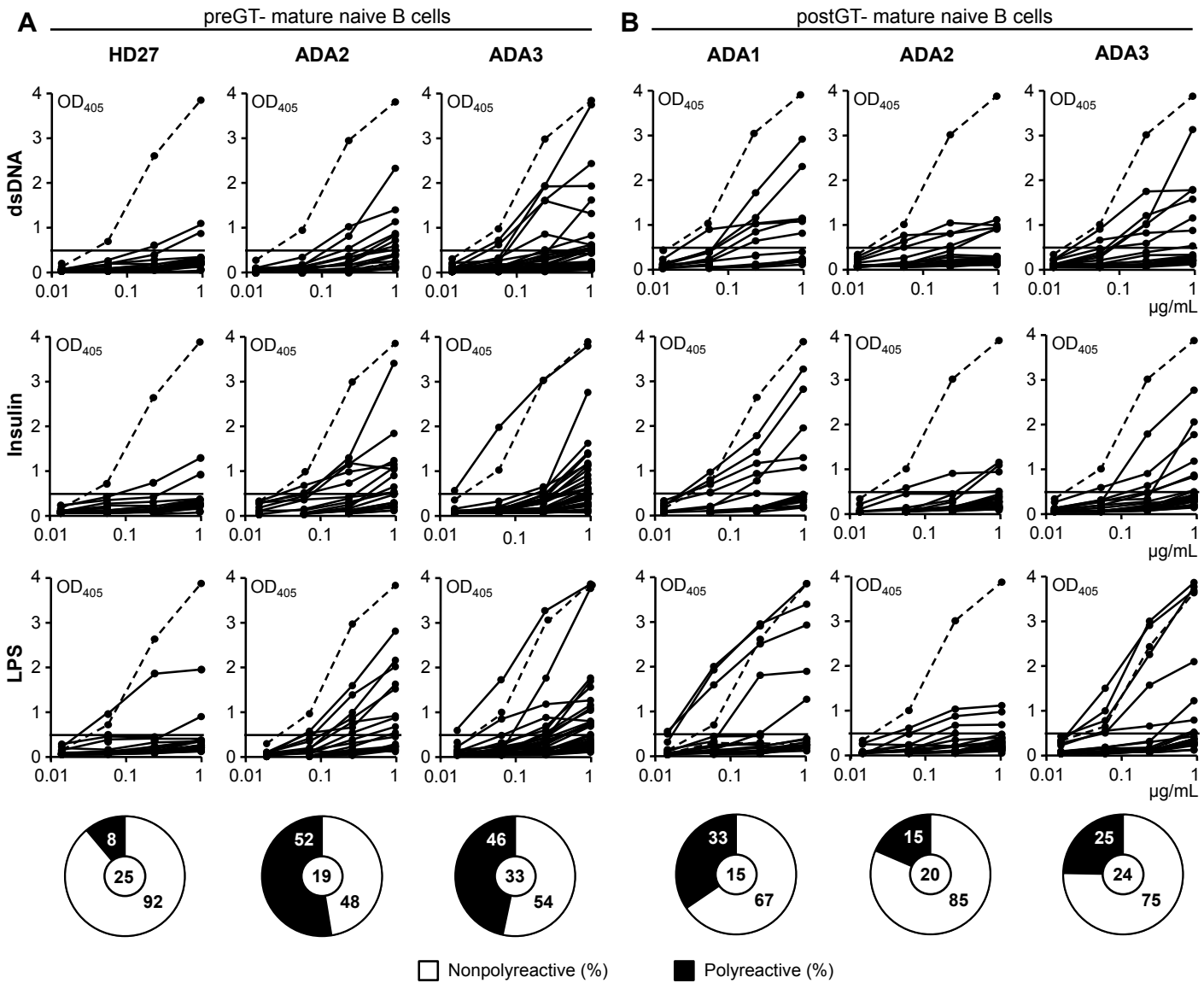
**Table S17:** Serum autoantibodies in ADA-SCID patients 6yrs postGT



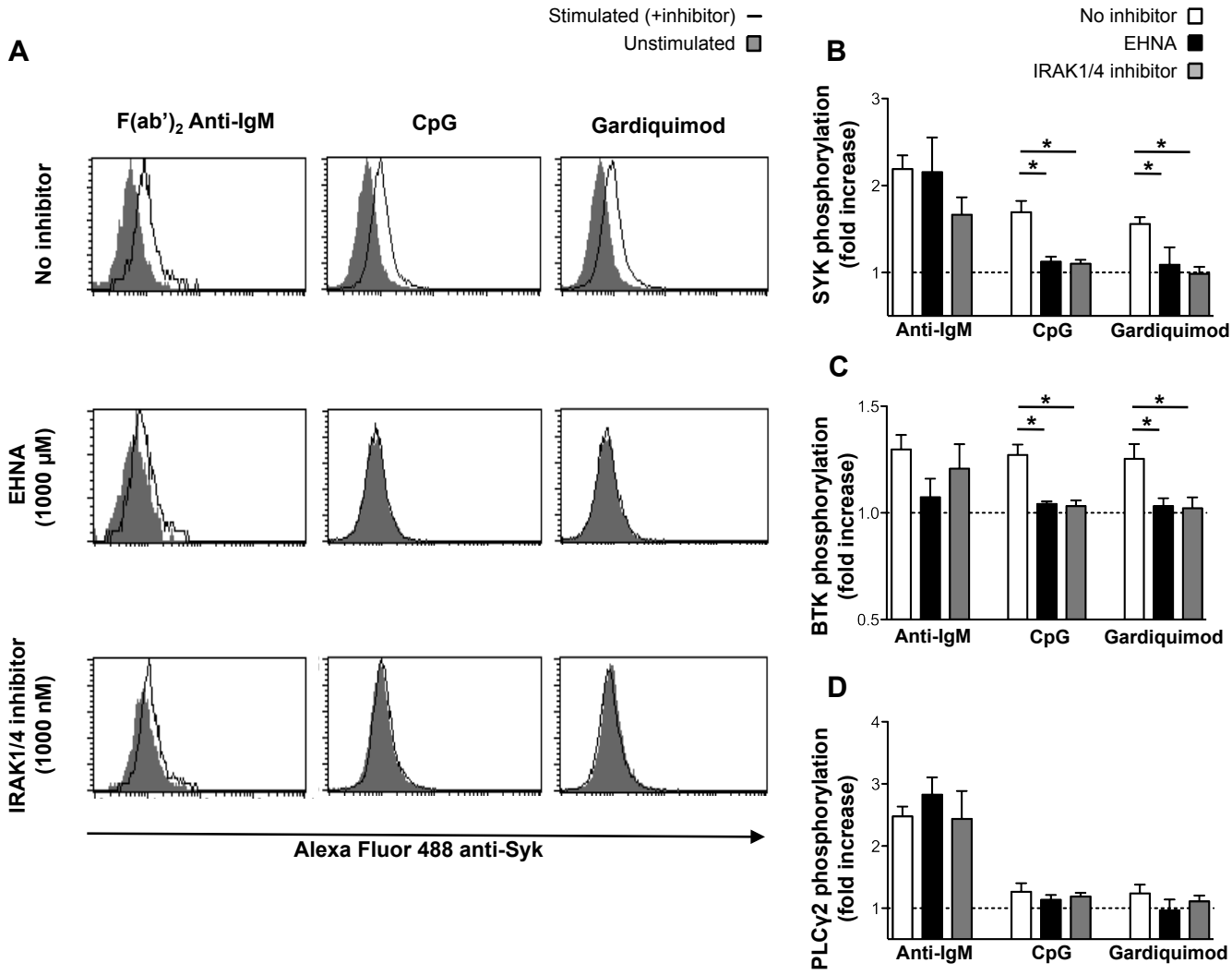

**Supplemental Figure 1**

ADA deficient ANA-expressing B cells display the highest frequency of positively charged residues in their heavy-chain CDR3s. **(A)** New emigrant B cells from ADA deficient patients were grouped into non-autoreactive, autoreactive non-ANA (Hep-2-reactive and/or polyreactive) and ANA clones. The numbers of positively charged residues in immunoglobulin heavy-chain CDR3s were calculated excluding the arginine or lysine found at position 94 in most germline VHs. Pie charts show the proportion of heavy-chain CDR3s with zero, one, two or more positive charges. The absolute number of clones analyzed in each B cell fraction is indicated in the center. A one-tailed, two-proportion z-test was performed considering the non-autoreactive and ANA-reactive groups as two independent samples. Zero positive charges, $p$= n.s.; one positive charge, $p$= n.s.; two or more positive charges, $p$< 0.05*. The average number of positively charged residues per CDR3 is indicated below each fraction. **(B)** The frequency of positively charged residues (≥2) per heavy-chain CDR3 of the new emigrant compartment of ADA deficient patients significantly decreases postGT.



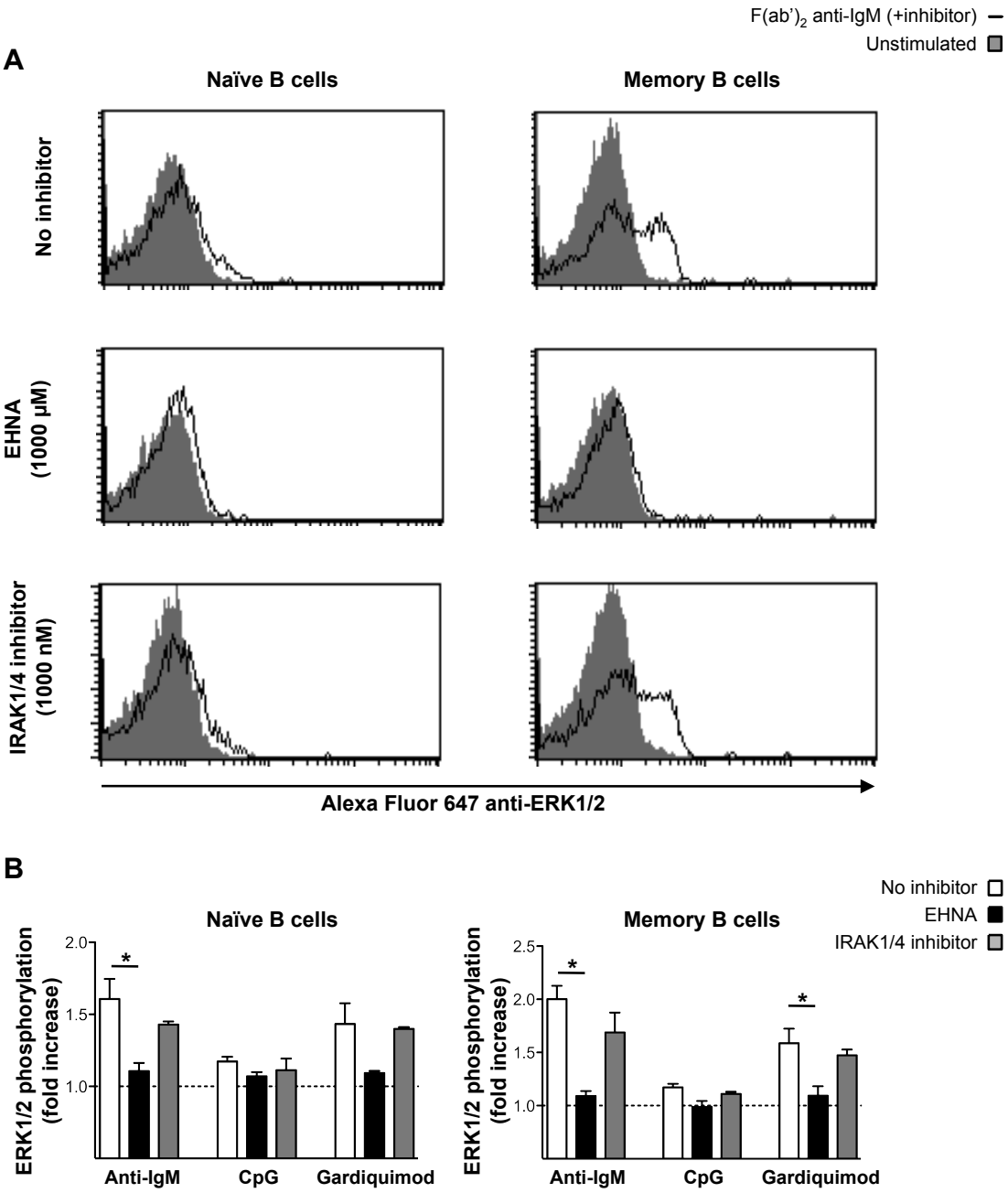

**Supplemental Figure 2**

ADA-deficient mature naive B cells express polyreactive antibodies before and after gene therapy. Antibodies from the mature naïve B cell fraction of **(A)** two ADA-SCID patients before and **(B)** three patients after gene therapy were tested in ELISA for reactivity with double-stranded DNA (dsDNA), insulin and lipopolysaccharide (LPS). Young adult HD27 is shown as representative control. Dotted lines show ED38-positive control. Horizontal lines show cut-off OD405 for positive reactivity. For each individual, the frequency of polyreactive (in black) and non polyreactive (in white) clones is summarized in pie charts with the number of antibodies tested indicated in the centers.



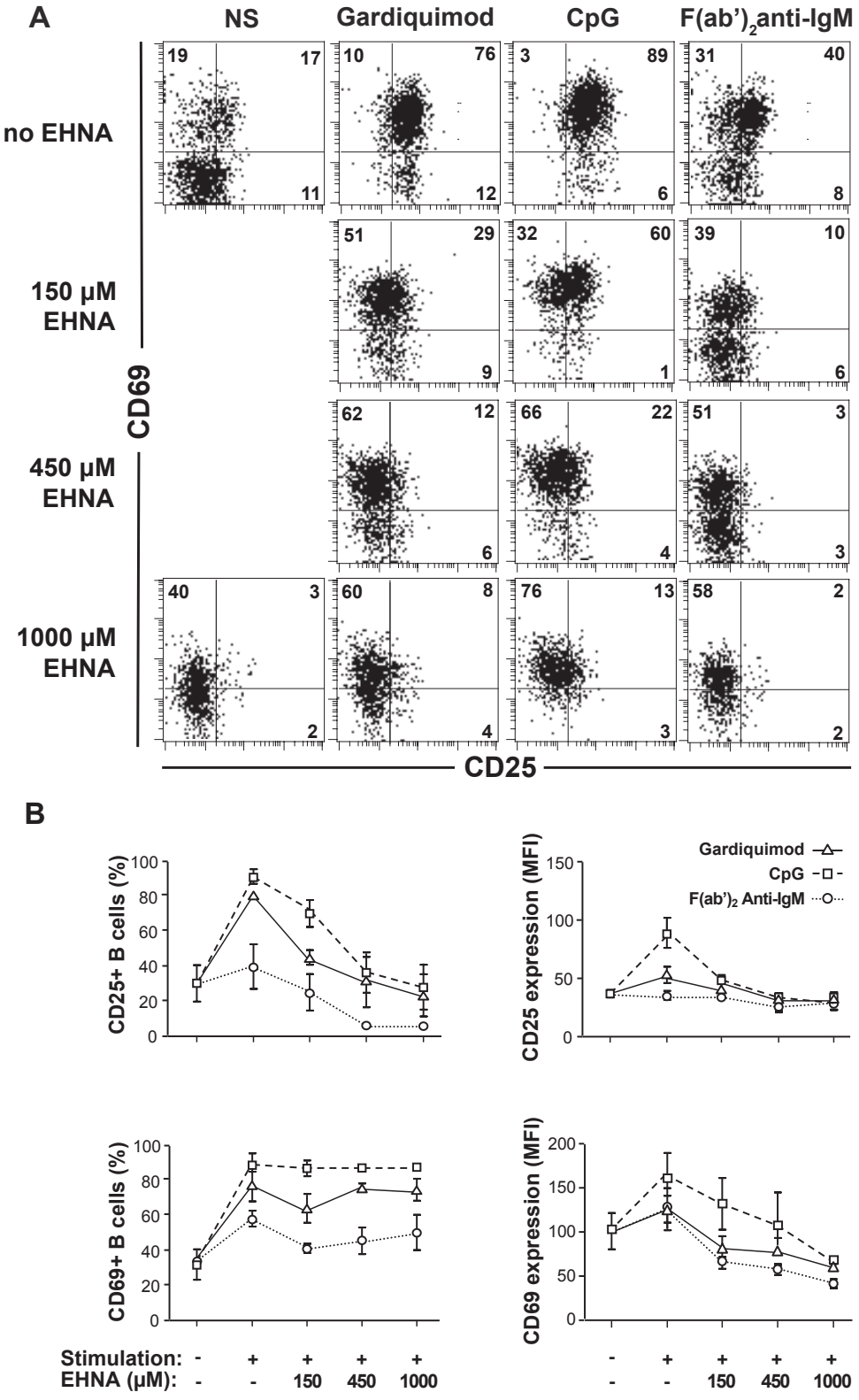

**Supplemental Figure 3**

Defective phosphorylation of Syk and BTK upon TLR stimulation in naïve B cells after ADA and IRAK1/4 inhibition. Total PBMCs from healthy donors after ADA or IRAK1/4 inhibition were stimulated *in vitro* with F(ab'2) anti-IgM, CpG or Gmod for 15min. **(A)** Representative histogram blots for Syk phosporylation. **(B-D)** Fold increase of phosphorylation as compared to non-stimulated controls for **(B)** Syk, **(C)** BTK and **(D)** PLCγ2. Data are representative of at least four independent experiments; **p*≤0.05.



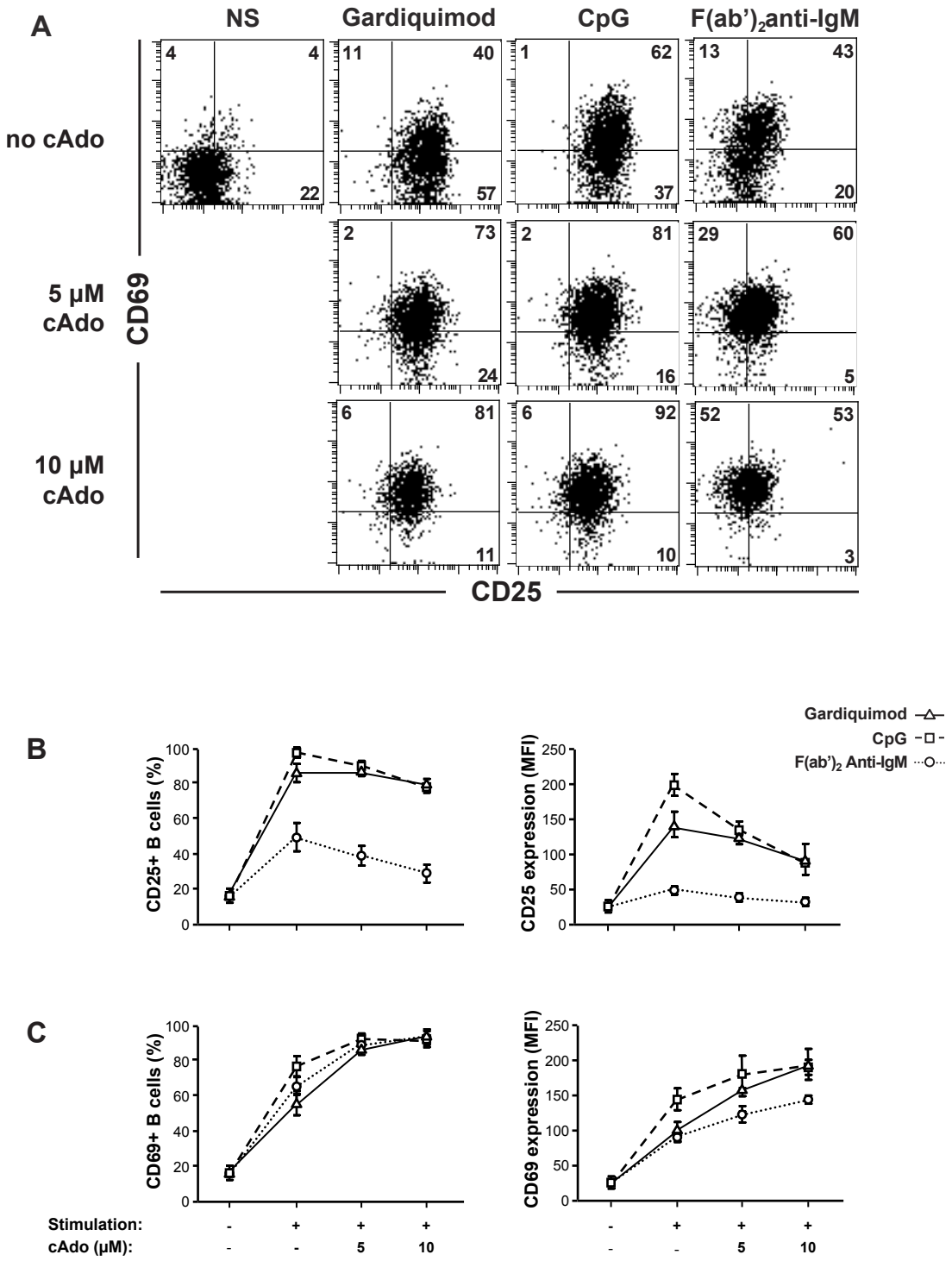

**Supplemental Figure 4**

Defective phosphorylation of ERK1/2 upon BCR stimulation in B cells after ADA inhibition. Total PBMCs from healthy donors after ADA or IRAK1/4 inhibition were stimulated *in vitro* with F(ab'2) anti-IgM, CpG or Gardiquimod for 15min. **(A)** Representative histogram blots for ERK1/2 phosporylation in naïve B cells (left) and memory B cells (right) after F(ab'2) anti-IgM stimulation. **(B)** Fold increase of phosphorylation as compared to non-stimulated controls for ERK1/2. Data are representative of three independent experiments; *$p \leq 0.05$.



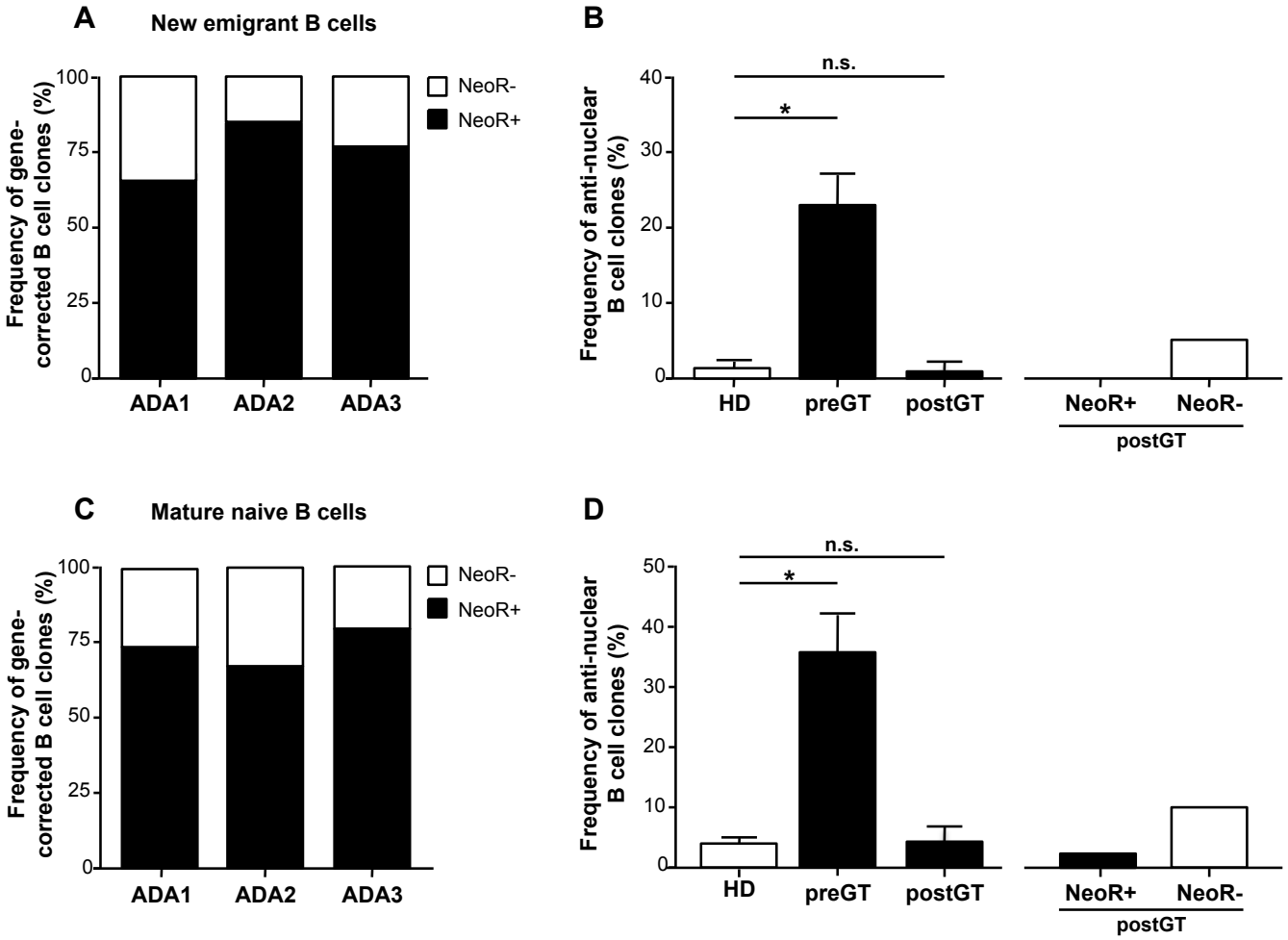

**Supplemental Figure 5**

Defective TLR-induced upregulation of B cell activation markers after ADA inhibition. Naïve B cells from healthy donors after ADA inhibition with increasing concentrations of EHNA were stimulated in vitro with Gardiquimod (TLR7 agonist), CpG (TLR9 agonist) or F(ab')2 anti-IgM for 2 days. **(A)** Representative dot plots, **(B)** percentages and mean fluorescence intensity (MFI) for CD25 and CD69. Data are representative of at least three independent experiments.




**Supplemental Figure 6**

Defective TLR-induced upregulation of B-cell activation markers after stimulation in the presence of 2-chloroadenosine (cAdo). **(A)** Dot plots show CD25 and CD69 expression on naïve B cells from healthy donors exposed to increasing concentrations of cAdo and stimulated or not (NS) *in vitro* with Gardiquimod (TLR7 agonist), CpG (TLR9 agonist) or F(ab')2 anti-IgM for 2 days. Percentages and mean fluorescence intensity (MFI) for CD25 and CD69 are shown in **(B)**. Data are representative of at least six independent experiments ±SEM.




**Supplemental Figure 7**

The majority of ANA expressing B cell clones found in the postGT fractions of ADA-SCID patients are not gene-corrected. **(A)** Overall frequency of gene-corrected (NeoR+) and not gene-corrected (NeoR-) B cells in the new emigrant B-cell compartment of ADA-SCID patients 1-3. **(B)** The frequency of anti-nuclear new emigrant B cells in ADA-deficient patients is completely corrected after gene therapy and is comparable to controls. Average is shown as bar +SEM; **p*=0.05- 0.005; n.s.= not significant. **(C)** Overall frequency of gene-corrected (NeoR+) and not gene-corrected (NeoR-) B cells in the mature naïve B-cell compartment of ADA-SCID patients 1-3. **(D)** The frequency of anti-nuclear mature naive B cells in ADA-deficient patients after gene therapy is comparable to controls. Average is shown as bar +SEM; **p*=0.05- 0.005; n.s.= not significant. When separating gene-corrected (NeoR+) from non-corrected (NeoR-) within the new emigrant and mature naïve B cells postGT the NeoR- fraction contains most ANA expressing clones.

**Supplemental Table 1:** Repertoire and reactivity of antibodies from new emigrant B cells of HD27

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Staining |
| neHD27 01 | 3-23 | / | / | 5 | DSSFDWFDP | 9 | 1-8 | 1 | QQYYSYPRT | 9 | - | - |
| neHD27 05 | 4-39 | 4-17 | 2 | 4 | DPNYGFSFDY | 10 | 1-6 | 4 | LQDYNYPPT | 9 | - | - |
| neHD27 06 | 3-23 | 3-22 | 3 | 1 | GAPSGIVVVITLYFQH | 16 | 4-1 | 4 | QQYYSTPLT | 9 | - | - |
| neHD27 07 | 3-23 | 3-3 | 3 | 6 | LGEIFGVVIIGGMDV | 15 | 1-9 | 4 | QQLNSYFLT | 9 | - | - |
| neHD27 08 | 3-7 | 4-17 | 2 | 3 | DRITYGRPDAFDI | 13 | 1-5 | 4 | QQYNSYSRLT | 10 | - | - |
| neHD27 09 | 4-39 | 1-26 | 2 | 6 | VGGSSLHYYYYYMDV | 15 | 1-39 | 4 | QQSYSTLALT | 10 | - | + (C) |
| neHD27 10 | 4-39 | 6-13 | 3 | 4 | RPRIAAAGGGYFDY | 14 | 3-15 | 2 | QQYNNWPQD | 9 | - | - |
| neHD27 11 | 4-31 | 3-10 | 1 | 3 | ELGFGEVPDAFDI | 13 | 1D-8 | 1 | QQYYSFPRT | 9 | - | + (C) |
| neHD27 13 | 3-21 | 6-13 | 3 | 4 | GTTLGIAAAGSTVLGYFDY | 19 | 3-15 | 1 | QQYNNWPRT | 9 | - | + (C) |
| neHD27 15# | 3-30 | 2-2 | 3 | 6 | DRGDIVVVPAAIRYYYYGMDV | 21 | 2-29 | 5 | MQSIQPPIT | 9 | | |
| neHD27 16 | 3-30 | 6-19 | 3 | 5 | DPSIAVAGTWWFDP | 14 | 1-12 | 3 | QQANSFPPS | 9 | - | - |
| neHD27 18 | 4-59 | / | / | 4 | DSDS | 4 | 1-39 | 2 | QQSYSTPRT | 9 | - | - |
| neHD27 19 | 4-34 | 1-26 | 3 | 6 | GGATRPAPMDV | 11 | 1-8 | 2 | QQYYSYPHT | 9 | - | - |
| neHD27 20 | 4-59 | 2-2 | 3 | 6 | VAYGFQVVPAAIRYYYMDV | 19 | 1-39 | 4 | QQSYSTPLT | 9 | + | - |
| neHD27 23 | 3-64 | 2-2 | 3 | 4 | DHHIVVVPAAPI | 12 | 2-28 | 1 | MQALQTPRT | 9 | - | - |
| neHD27 24 | 4-39 | 3-16 | 2 | 4 | ATWGGSGGSYRAYYFDY | 17 | 3-20 | 3 | QQYGSSPHT | 9 | - | - |
| neHD27 25 | 4-4 | / | / | 4 | RNGAFHY | 7 | 1-5 | 2 | QQYNSYSYT | 9 | - | - |
| neHD27 26 | 3-7 | 6-19 | 3 | 4 | DKVPIAVAPNDY | 12 | 1-9 | 1 | QQLNSYPPRT | 10 | - | - |
| neHD27 27# | 4-4 | 3-3 | 3 | 6 | GPHPTIFGVPDRDYYYYYYMDV | 22 | 3-11 | 4 | QQRSNWPLPT | 10 | | |
| neHD27 28 | 6-1 | 6-19 | 2 | 4 | APEYSSGYDY | 10 | 4-1 | 1 | QQYYSTPPWT | 10 | - | - |
| neHD27 32 | 1-24 | 6-6 | 2 | 4 | DPPSSSGYDY | 10 | 3-11 | 1 | QQRSNWPPT | 9 | - | - |
| neHD27 33# | | | | | see lambda | | 3-20 | 2 | QQYGSSPPDT | 10 | | |
| neHD27 35 | 3-7 | 6-19 | 3 | 3 | GAAVAGHNGAFDI | 13 | 1-8 | 1 | QQYYSYPQT | 9 | - | - |
| neHD27 36 | 4-4 | 3-22 | 2 | 4 | LRTYYYDSSGLPNQHYYFDY | 20 | 3-15 | 1 | QQYNNWPRT | 9 | - | - |
| neHD27 39 | 3-30 | 1-7 | 3 | 4 | DGRTGTTLGPFDY | 13 | 3-20 | 1 | QQYGSSPRT | 9 | - | - |
| neHD27 45# | 3-15 | 3-22 | 2 | 2 | DRGIEVSGYPPPHAFDI | 17 | 3-11 | 5 | QQRSNWPPIT | 10 | | |
| neHD27 46 | 3-3- | 3-3 | 2 | 4 | GALPDFWSGYSLDYFDY | 17 | 3-15 | 1 | QQYNNWPPWT | 10 | - | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Staining |
| neHD27 02 | 4-34 | 3-9 | 2 | 5 | GGYDILTGYGNWFDP | 15 | 1-40 | 3 | QSYDSSLSGSRV | 12 | - | - |
| neHD27 04 | 4-59 | 3-10 | 2 | 6 | MGHYYGSGSYYNTGDMDV | 18 | 2-14 | 2 | SSYTSSSTVV | 10 | - | - |
| neHD27 09 | | | | | see kappa | | 1-51 | 3 | GTWDSSLSAWV | 11 | - | - |
| neHD27 12 | 4-34 | 3-22 | 3 | 5 | ALTMIVVGNWFDP | 13 | 1-40 | 2 | QSYDSSLSGSV | 11 | - | - |
| neHD27 14 | 1-69 | 5-24 | 3 | 4 | AVEMATIRYFDY | 12 | 8-61 | 3 | VLYMGSGIWV | 10 | + | - |
| neHD27 17 | 3-23 | / | / | 4 | DRGIVTFRTFDY | 12 | 2-14 | 1 | SSYTSSSTPGSV | 12 | - | + (C) |
| neHD27 21 | 3-43 | 6-13 | 3 | 4 | AGGIAASAAGGFDY | 14 | 3-21 | 1 | QVWDSSSDHYV | 11 | - | - |
| neHD27 22 | 4-61 | 6-13 | 3 | 6 | DLGTAAADYYYYYGMDV | 17 | 2-14 | 1 | SSYTSSFYV | 9 | - | - |
| neHD27 28 | | | | | see kappa | | 2-11 | 3 | CSYAGSYWV | 9 | - | - |
| neHD27 29 | 3-13 | 1-26 | 2 | 2 | GREYSGSYYWYFDL | 14 | 6-57 | 2 | QSYDSSNHVV | 10 | - | - |
| neHD27 30 | 3-15 | 5-24 | 2 | 2 | GRDGYNGGLTGHWYFDL | 17 | 2-23 | 2 | CSYAGSSTSVV | 11 | - | - |
| neHD27 31 | 3-21 | 4-17 | 3 | 5 | DYTVTTGGWFDP | 12 | 3-1 | 2 | QAWDSSTAWDVV | 12 | - | - |
| neHD27 33# | 4-61 | 1-26 | 2 | 1 | YSGSYFEYFQH | 11 | 3-1 | 2 | QAWDSSTVV | 9 | | |
| neHD27 38 | 3-30 | 6-13 | 3 | 4 | GGGIAAAGKRGVLWY | 15 | 2-14 | 2 | SSYTSSSIVV | 10 | - | - |
| neHD27 41 | 3-30 | 6-13 | 3 | 6 | DQTSYIAAAHYYYYYGMDV | 19 | 1-44 | 3 | AAWDDSLNGLV | 11 | - | + (F) |
| neHD27 42 | 3-30 | 5-5 | 3 | 6 | APSRTAMVVGMDV | 13 | 8-61 | 3 | VLYMGSGIWV | 10 | - | - |
| neHD27 43 | 3-23 | 3-10 | 1 | 4 | AQGFGELLSLSYFDY | 15 | 1-40 | 3 | QSYDSSLSGSWV | 12 | - | - |
| neHD27 44 | 3-33 | 3-10 | 2 | 6 | DHLYGSGSYYNENYYYYYGMDV | 22 | 1-44 | 1 | AAWDDSLNGYV | 11 | - | - |
| neHD27 47 | 3-33 | 6-19 | 2 | 4 | DLRHSGWYLDSY | 12 | 2-14 | 2 | SSYTSSSTVV | 10 | - | - |
| neHD27 48 | 4-39 | 3-22 | 2 | 4 | RGYYDSSGYKFDY | 13 | 2-14 | 3 | SSYTSSSPWV | 10 | - | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | |
| neHD27 40 | 4-59 | 3-22 | 2 | 6 | GVSLDSSGYLDPYYYMDV | 18 | | | | | | |
| neHD27 37 | 1-69 | 3-22 | 2 | 4 | DQPYYYDSSGRTNLFDY | 17 | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 2:** Repertoire and reactivity of antibodies from new emigrant B cells of HD28

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Staining |
| neHD28 1# | 1-69 | 2-15 | 3 | 5 | DRVVAATRGEFDP | 14 | 1-39 | 2 | QQSYSTPCS | 9 | | |
| neHD28 2 | 1-24 | 3-16 | 2 | 3 | ANPWGSYRSHAFDI | 14 | 3-11 | 4 | QQRSNWPLT | 9 | - | - |
| neHD28 3# | 1-69 | 3-22 | 2 | 6 | VLYYDSSGYSFYYYGMDV | 18 | 1-27 | 1 | QKYNSAPWT | 9 | | |
| neHD28 4 | 3-30 | 3-22 | 2 | 4 | ELGDHYYDSSGYQRTPFDY | 19 | 3-15 | 3 | QQYNNWPPLFT | 11 | - | - |
| neHD28 6 | 3-30-3 | 1-26 | 2 | 3 | GVASYYGGAFDI | 12 | 1-8 | 2 | QQYYSYPYT | 9 | - | + (F) |
| neHD28 7# | 3-23 | 3-22 | 2 | 4 | GAYYYDSSGYYRWPFDY | 17 | 1-8 | 2 | QQYYSYLYT | 9 | | |
| neHD28 8 | 3-21 | 5-12 | 2 | 1 | VFDHTNPTEGYSGYDEYFQH | 20 | 1-5 | 2 | QQYNSYPCS | 9 | - | - |
| neHD28 11 | 4-59 | 6-13 | 1 | 6 | DRGIVKQQLVRVGYYYYGMDV | 21 | 3-11 | 2 | QQRSNWLYT | 9 | + | + (F) |
| neHD28 17 | 1-2 | 2-2 | 2 | 5 | DFWDRGDGPGYCSSTSCYTSSPPRMIFDP | 29 | 1-39 | 1 | QQSYSTPWT | 9 | - | - |
| neHD28 18 | 3-33 | 3-22 | 2 | 1 | GRYYYDSSGYYYGEYFQH | 18 | 4-1 | 1 | QQYYSTPPT | 9 | - | - |
| neHD28 20# | 1-8 | / | / | 6 | KRDYYYGMDV | 10 | 1-39 | 2 | QQSYSTPYT | 9 | | |
| neHD28 24# | 4-34 | 6-13 | 3 | 1 | GLGAAGPVHQH | 11 | 4-1 | 2 | QQYYSTPPG | 9 | | |
| neHD28 25 | 4-59 | 6-19 | 3 | 5 | HGREAGTP | 8 | 4-1 | 5 | QQYYSTLIT | 9 | - | + (C) |
| neHD28 26 | 4-31 | 3-10 | 2 | 4 | GLWYYGSGSYYKQFDY | 16 | 1-12 | 5 | QQANSFPIT | 9 | + | - |
| neHD28 28 | 4-39 | 3-22 | 2 | 3 | RYYYDSSGYLRDAFDI | 16 | 1-6 | 1 | LQDYNYPWT | 9 | - | - |
| neHD28 31 | 3-33 | / | / | 6 | VRVPNYYYYGMDV | 13 | 2-28 | 1 | MQALQTPWT | 9 | - | - |
| neHD28 33 | 3-15 | 4-17 | 3 | 3 | DRGLTTVTTDAFDI | 14 | 2-28 | 2 | MQALQTPPS | 9 | - | - |
| neHD28 34 | 1-18 | 3-10 | 2 | 6 | DRPYGSSYDLYYYYYGMDV | 19 | 3-20 | 1 | QQYGSSPQT | 9 | - | + (C) |
| neHD28 36 | 3-21 | / | / | 4 | GPGSFQVDY | 9 | 1-39 | 4 | QQSYSTPALT | 10 | + | + (C) |
| neHD28 41 | 4-59 | 5-24 | 2 | 3 | LMATGKDAFDI | 11 | 1-8 | 2 | QQYYSFPYT | 9 | - | - |
| neHD28 44 | 1-2 | 6-19 | 2 | 6 | ASSSGWYNYYGMDV | 14 | 1-16 | 2 | QQYNSYPYT | 9 | - | - |
| neHD28 45# | 1-18 | 3-3 | 2 | 3 | GAGSMSYDFWSGY | 13 | 4-1 | 1 | QQYYSTPQT | 9 | | |
| neHD28 46 | 4-31 | 2-15 | 3 | 5 | EVVVAAMKNWFDP | 13 | 1-39 | 1 | QQSYSTPRT | 9 | - | - |
| neHD28 5# | | | | | | | 4-1 | 2 | QQYYSTPCS | 9 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Staining |
| neHD28 12 | 3-73 | 3-22 | 2 | 6 | RVDDSSGYTNYYYGMDV | 17 | 3-1 | 2 | QAWDSSIVV | 9 | - | - |
| neHD28 14 | 4-34 | 4-17 | 3 | 4 | GRSYLPTVTTYYFDY | 15 | 3-21 | 1 | QVWDSSSDHYV | 11 | - | - |
| neHD28 15# | 5-51 | / | / | 2 | HRYFDL | 6 | 2-8 | 3 | SSYAGSNK | 8 | | |
| neHD28 19# | 1-18 | 1-1 | 2 | 4 | DRATAVTNWSDY | 12 | 2-14 | 3 | SSYTSSSTRV | 10 | | |
| neHD28 29 | 3-33 | 5-5 | 3 | 4 | VDTAMGTKGKEKTFDY | 16 | 1-44 | 3 | AAWDDSLNGWV | 11 | - | - |
| neHD28 30 | 3-23 | 2-21 | 2 | 4 | EGTYCGGDCYSGHFDY | 16 | 3-21 | 2 | QVWDSSSDHVV | 11 | - | - |
| neHD28 32# | 1-18 | 3-10 | | 4 | HLSIGFGSDYFDY | 13 | 1-40 | 3 | QSYDSSLSEGV | 11 | | |
| neHD28 34 | | | | | see kappa | | 3-21 | 3 | QVWDSSSDHWV | 11 | - | + (C) |
| neHD28 35 | 1-2 | 3-3 | 1 | 5 | VLEGAAPWGTLE | 12 | 1-47 | 1 | AAWDDSLSGYV | 11 | - | - |
| neHD28 36 | | | | | see kappa | | 2-14 | 1 | SSYTSSSTPYV | 11 | - | - |
| neHD28 39 | 3-21 | 6-19 | 2 | 4 | DLWDAGYSSGWYQVGYGYYFDY | 22 | 1-44 | 1 | AAWDDSLNGYV | 11 | - | - |
| neHD28 40 | 1-2 | 2-8 | 2 | 6 | GYKCTNGVCTYDGMDV | 16 | 3-21 | 1 | QVWDSSSDHYV | 11 | - | - |
| neHD28 41 | | | | | see kappa | | 2-11 | 1 | CSYAGSYTVYV | 11 | - | - |
| neHD28 43 | 4-4 | 1-20 | 3 | 6 | VWGTGTTYGMDV | 12 | 1-44 | 1 | AAWDDSLNGLYV | 12 | - | - |
| neHD28 47 | 4-31 | 3-22 | 2 | 4 | AAGNYYDSSGYYHFDY | 16 | 2-14 | 2 | SSYTSSSTVV | 10 | - | - |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 3:** Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 1 before gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Staining |
| neADA1-pre 01 | 3-7 | 7-27 | 3 | 3 | DPSTGGHDAFDI | 12 | 3-11 | 2 | QQRSNWSGPLCS | 12 | - | - |
| neADA1-pre 02 | 1-2 | 3-10 | 2 | 4 | PLGSGSYNY | 9 | 3-20 | 1 | QQYGSSPWT | 9 | - | - |
| neADA1-pre 03 | 1-3 | 6-19 | 3 | 4 | LAVAGTPFDY | 10 | 1-39 | 2 | QQSYSTPYT | 9 | - | - |
| neADA1-pre 04 | 3-7 | 2-15 | 2 | 4 | GYCNGGSCPYYFDY | 14 | 1-16 | 1 | QQYNSYPRQSLMT | 13 | + | + (C) |
| neADA1-pre 06 | 3-9 | 6-6 | 3 | 3 | DRAAARPLRYAFDI | 14 | 1-5 | 4 | QQYNSYPT | 8 | - | - |
| neADA1-pre 08# | 3-21 | 2-15 | 3 | 6 | EDIVVVVAATPFRDMDV | 17 | 1-39 | 5 | QQSYSTLT | 8 | | |
| neADA1-pre 09 | 3-15 | 3-3 | 1 | 4 | HSPKLQFLEWFCDY | 14 | 3-20 | 2 | QQYGSSPCS | 9 | + | + (N) |
| neADA1-pre 10 | 4-59 | 6-13 | 3 | 4 | HGIAAVWSPFDY | 12 | 1-8 | 1 | QQYYSYPWT | 9 | + | + (C) |
| neADA1-pre 12 | 3-30-3 | / | / | 3 | AQRGDAFDI | 9 | 1-39 | 2 | QQSYSTPYT | 9 | - | - |
| neADA1-pre 13# | 3-15 | 3-10 | 3 | 4 | EPLKKTGKGVIIKGDY | 16 | 3-11 | 4 | QQRSNWPSLT | 10 | | |
| neADA1-pre 14# | 4-34 | 6-6 | 3 | 4 | RYAARPYYFDY | 11 | 1-33 | 5 | QQYDNLPIT | 9 | | |
| neADA1-pre 15 | 3-66 | 6-19 | 2 | 4 | DLVSGSGWDY | 10 | 3-20 | 1 | QQYGSSLWT | 9 | - | - |
| neADA1-pre 16 | 3-11 | 3-3 | 2 | 4 | VYDFWSFDY | 9 | 2-30 | 4 | MQGTHWPPLT | 10 | - | - |
| neADA1-pre 18 | 3-15 | 3-22 | 2 | 4 | DSDDSSGYYGYFDY | 14 | 2-28 | 3 | MQALQTFT | 8 | - | - |
| neADA1-pre 19# | 3-11 | 3-3 | 1 | 4 | GNLRFLEWLSYFDY | 14 | 3-20 | 1 | QQYGSSLWT | 9 | | |
| neADA1-pre 22 | 3-43 | 1-7 | 3 | 4 | QTGTGLDY | 8 | 1-39 | 4 | QQSYSTPLT | 9 | - | - |
| neADA1-pre 23 | 3-15 | 6-13 | 1 | 4 | DLQQQLPRRYFDY | 13 | 1-6 | 3 | LQDYNYPFT | 9 | + | - |
| neADA1-pre 24 | 4-39 | 2-21 | 2 | 4 | YCGGDCVYYFDY | 12 | 3-20 | 2 | QQYGSSPYT | 9 | + | + (C) |
| neADA1-pre 26 | 3-30 | 6-19 | 3 | 3 | DIAVADHDAFDI | 12 | 1-8 | 3 | QQYYSYPFT | 9 | - | + (C) |
| neADA1-pre 27 | 3-9 | 3-9 | 2 | 4 | GRHRYDILTGYRDY | 14 | 1-33 | 3 | QQHDNLPFT | 9 | + | - |
| neADA1-pre 28# | 3-11 | 3-10 | 3 | 4 | GPITMVQG | 8 | 3-15 | 4 | QQYNNWPPNLT | 11 | | |
| neADA1-pre 31# | 1-3 | 6-13 | 3 | 4 | DPLVGIAAAGSGY | 13 | 3-20 | 4 | QQYGSSPV | 8 | | |
| neADA1-pre 33 | 3-23 | 2-15 | 2 | 3 | FYCSGGSCYRDAFDI | 15 | 2-30 | 4 | MQGTHWPT | 8 | - | + (S) |
| neADA1-pre 34 | 3-23 | 1-20 | 2 | 4 | RRYNWNDEEQVY | 12 | 3-20 | 2 | QQYGSSPCS | 9 | + | + (N) |
| neADA1-pre 35 | 5-51 | / | / | 4 | FRTYYFDY | 8 | 1-12 | 4 | QQANSFPLT | 9 | - | - |
| neADA1-pre 39# | 4-34 | 4-23 | 3 | 4 | TVEHYYFDY | 9 | 3-15 | 2 | QQYNNWPRFRFRYT | 14 | | |
| neADA1-pre 40# | 4-34 | 6-13 | 2 | 3 | PCRYSSIQDAFDI | 13 | 1-17 | 2 | LQHNSYPRS | 9 | | |
| neADA1-pre 41 | 3-30-3 | 6-13 | 1 | 5 | ASLRQQLVGGWFDP | 14 | 3-15 | 4 | QQYNNWPPLT | 10 | - | - |
| neADA1-pre 45 | 3-30-3 | 6-13 | 2 | 1 | VQGSSWYLGYFQH | 13 | 1-17 | 4 | LQHNSYPPT | 9 | + | + (N) |
| neADA1-pre 46 | 3-30-3 | 6-19 | 3 | 2 | DARIAVADPYWYFDL | 15 | 1-8 | 4 | QQYYSYPLLT | 10 | + | + (N) |
| neADA1-pre 48 | 3-9 | / | / | 4 | VGRERSGFDY | 10 | 1-39 | 2 | QQSYSTLMCS | 10 | - | - |
| neADA1-pre 05 | | | | | | | 3-20 | 2 | QQYGSSPFSCS | 11 | | |
| neADA1-pre 07 | | | | | | | 1-27 | 3 | QKYNSALPIT | 10 | | |
| neADA1-pre 11 | | | | | | | 1-39 | 4 | QQSYSTPLT | 9 | | |
| neADA1-pre 17 | | | | | | | 1-33 | 3 | QQHDNLPLT | 9 | | |
| neADA1-pre 21 | | | | | | | 3-15 | 1 | QQYNNWPPWT | 10 | | |
| neADA1-pre 25 | | | | | | | 1-9 | 4 | QQLNSYPLT | 9 | | |
| neADA1-pre 29 | | | | | | | 4-1 | 5 | QQYYSTIT | 8 | | |
| neADA1-pre 32 | | | | | | | 3-11 | 1 | QQRSNWPWT | 9 | | |
| neADA1-pre 36 | | | | | | | 1-27 | 2 | QKYNSAPHT | 9 | | |
| neADA1-pre 37 | | | | | | | 3-15 | 1 | QQYNNWPPWT | 10 | | |
| neADA1-pre 38 | | | | | | | 2-29 | 3 | MQGIHLPFT | 9 | | |
| neADA1-pre 43 | | | | | | | 2-30 | 1 | MQGTHWPRT | 9 | | |
| neADA1-pre 44 | | | | | | | 3-11 | 5 | QQRSNWPIT | 9 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Staining |
| neADA1-pre 30 | 3-15 | / | / | 2 | DPNRWYFDL | 9 | 2-23 | 1 | CSYAGSSTFV | 10 | - | - |
| neADA1-pre 32# | 1-2 | / | / | 4 | SPEDY | 5 | 2-23 | 1 | CSYAGSSTYV | 10 | | |
| neADA1-pre 42# | 4-59 | 3-22 | 2 | 4 | RVYDSSGYVYFDY | 13 | 2-23 | 1 | CSYAGSSTYV | 10 | | |
| neADA1-pre 47 | 4-39 | 3-9 | 1 | 4 | DLRYFDWLLYYFDY | 14 | 2-14 | 2 | SSYTSSSTLVV | 11 | + | + (C) |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 4: Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 2 before gene therapy**

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Staining |
| neADA2-pre 02 | 3-7 | 6-19 | 2 | 4 | AGYSSAGY | 8 | 1-5 | 2 | QQYNSYSMYT | 10 | - | + (N) |
| neADA2-pre 03# | 3-21 | 2-2 | 3 | 6 | DVLVVPAAMDV | 11 | 1-8 | 2 | QQYYSYPRT | 9 | | |
| neADA2-pre 07 | 1-8 | 6-13 | 3 | 2 | RAAARSYWYFDL | 12 | 1-5 | 1 | QQYNSYPWT | 9 | + | + (C) |
| neADA2-pre 09 | 3-48 | 1-26 | 3 | 5 | LVGAMGTYWFDP | 12 | 3-20 | 2 | QQYGSSPPYT | 10 | - | - |
| neADA2-pre 11 | 3-15 | 1-26 | 3 | 4 | EVGASQY | 7 | 3-11 | 2 | QQRNNT | 6 | - | - |
| neADA2-pre 24 | 1-18 | 6-19 | 3 | 5 | VLGIAVAGTSWFDP | 14 | 1-39 | 3 | QQSYSTLT | 8 | - | - |
| neADA2-pre 28 | 3-53 | 2-15 | 2 | 4 | MSGYCSGGSCLLM | 13 | 1-5 | 2 | QQYNSYLYT | 9 | + | - |
| neADA2-pre 30 | 3-30 | 3-10 | 2 | 4 | STKSQGSGSYYNVRRYFDP | 19 | 3-15 | 2 | QQYNNWPPYT | 10 | + | + (N) |
| neADA2-pre 43# | 1-2 | 2-2 | 3 | 5 | LKGIVVVPAAIPNNWFDP | 18 | 1D-17 | 3 | LQHNSYPFT | 9 | | |
| neADA2-pre 103# | 3-23 | 3-10 | 1 | 4 | VLGENWLDY | 9 | 1-8 | 2 | QQYYSYPRT | 9 | | |
| neADA2-pre 202 | 3-30-3 | 3-16 | 2 | 4 | EGYYDYVWGSYRPYYFDY | 18 | 1-5 | 4 | QQYNSYPLT | 9 | - | - |
| neADA2-pre 204 | 4-4 | / | / | 5 | AEVDWGYIEVGWFDP | 15 | 3-15 | 2 | QQYNNWPRT | 9 | - | - |
| neADA2-pre 205 | 4-59 | 5-5 | 2 | 6 | SGYSYGYRLHGMDV | 14 | 1-39 | 2 | QQSYSTPHT | 9 | + | + (C) |
| neADA2-pre 207 | 3-21 | 6-19 | 3 | 4 | DRTTGIAVASDY | 12 | 3-11 | 2 | QQRSNWPQT | 9 | - | - |
| neADA2-pre 209 | 4-4 | 2-8 | 2 | 4 | TVSTNGELNFDY | 12 | 1-8 | 1 | QQYYSYPRT | 9 | - | + (N) |
| neADA2-pre 211# | 3-15 | 6-6 | 3 | 4 | DLAARRCFDY | 10 | 1D-12 | 1 | QQANS | 5 | | |
| neADA2-pre 215 | 4-59 | 6-19 | 3 | 4 | VNGGAGKGLDY | 11 | 2-28 | 1 | MQALQTPWT | 9 | - | - |
| neADA2-pre 216 | 3-30 | 6-6 | 2 | 4 | EPFLPRYSSHVLGYFDY | 17 | 4-1 | 2 | QQYYSMYT | 8 | - | - |
| neADA2-pre 219 | 5-51 | 2-2 | 2 | 4 | HGGYCSSTSCQYSPFDY | 17 | 3-20 | 1 | QQYGSSPRT | 9 | - | - |
| neADA2-pre 221 | 4-34 | 2-2 | 3 | 4 | RRIVVVPAAKENFDY | 15 | 3-15 | 1 | QQYNNWPRT | 9 | - | - |
| neADA2-pre 222 | 1-18 | 6-19 | 3 | 4 | GRIAVAGTFDY | 11 | 1-5 | 4 | QQYNSYPLT | 9 | - | - |
| neADA2-pre 225 | 3-30-3 | 1-26 | 1 | 1 | GVIRWELLILAGEYFQH | 17 | 3-15 | 1 | QQYNNWPPWT | 10 | + | + (N) |
| neADA2-pre 227 | 3-9 | 6-6 | 2 | 4 | RYSSSSEFDY | 10 | 3-15 | 3 | QQYNNWPG | 8 | - | - |
| neADA2-pre 231 | 3-21 | 3-22 | 2 | 4 | DHLNRDYYDSSGYKHYFDY | 19 | 1-39 | 2 | QQSYSTPYT | 9 | - | - |
| neADA2-pre 232 | 1-2 | 5-5 | 3 | 4 | GSDTAMVPGTYYFDY | 15 | 3-11 | 1 | QQRSNWT | 7 | - | - |
| neADA2-pre 233# | 4-34 | 6-13 | 3 | 4 | GIIAAAGTRILIPKNRRHVLVRLDY | 25 | 3-20 | 1 | QQYGSSPPWT | 10 | | |
| neADA2-pre 234# | 3-15 | 2-2 | 2 | 6 | RCSSTSCYLNYYYGMDV | 17 | 1-33 | 2 | QQYDNLPMYT | 10 | + | - |
| neADA2-pre 236# | 3-9 | 1-26 | 3 | 4 | DRGLGANLDYFDY | 13 | 3-20 | 2 | QQYGSSPYT | 9 | | |
| neADA2-pre 246 | 3-30 | 4-17 | 2 | 4 | DYGTMAYYFDY | 11 | 3D-20 | 5 | QQYGSSPIT | 9 | - | - |
| neADA2-pre 05 | | | | | | | 1-39 | 2 | QQSYSTPYT | 9 | | |
| neADA2-pre 06 | | | | | | | 2-28 | 1 | MQALQTPPWT | 10 | | |
| neADA2-pre 14 | | | | | | | 1-27 | 4 | QKYNSAPLT | 9 | | |
| neADA2-pre 20 | | | | | | | 3-15 | 2 | QQYNNWPPTYT | 11 | | |
| neADA2-pre 26 | | | | | | | 4-1 | 2 | QQYYSTPYT | 9 | | |
| neADA2-pre 34 | | | | | | | 3-15 | 1 | QQYNNWPRT | 9 | | |
| neADA2-pre 37 | | | | | | | 1-39 | 3 | QQSYSTPFT | 9 | | |
| neADA2-pre 40 | | | | | | | 1-5 | 2 | QQYNSYPYT | 9 | | |
| neADA2-pre 210 | | | | | | | 3-20 | 1 | QQYGSSPRT | 9 | | |
| neADA2-pre 213 | | | | | | | 1-5 | 5 | QQYNSYSKT | 9 | | |
| neADA2-pre 218 | | | | | | | 1-39 | 3 | QQSYSTPLFT | 10 | | |
| neADA2-pre 220 | | | | | | | 2-28 | 5 | MQALQTPIT | 9 | | |
| neADA2-pre 224 | | | | | | | 2-28 | 4 | MQALQTLPHT | 10 | | |
| neADA2-pre 239 | | | | | | | 4-1 | 2 | QQYYSTPMYT | 10 | | |
| neADA2-pre 241 | | | | | | | 3-20 | 3 | QQYGSSRFT | 9 | | |
| neADA2-pre 242 | | | | | | | 1-39 | 3 | QQSYSTPFT | 9 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Staining |
| neADA2-pre 131 | 3-43 | 6-13 | 3 | 4 | DILQGAGSCFDY | 12 | 2-23 | 2 | CSYAGSSTFVV | 11 | - | - |
| neADA2-pre 217 | 3-30 | 6-13 | 3 | 4 | GGVAAAGRDSSGSPWPFYYFDY | 22 | 2-23 | 1 | CSYAGSSTYV | 10 | - | - |
| | | 3-22 | 2 | | | | | | | | | |
| neADA2-pre 226# | 3-30 | / | / | 4 | GRLALRGIDY | 10 | 3-21 | 3 | QVWDSSSDHWV | 11 | | |
| neADA2-pre 228 | 3-23 | 3-22 | 2 | 6 | AGLDSSGSKLRYYYYYGMDV | 20 | 3-1 | 2 | QAWDSSTVV | 9 | - | + (N) |
| neADA2-pre 230# | 1-8 | 3-22 | 3 | 3 | GAGAVEVVVITFDI | 14 | 3-21 | 2 | QVWDSSSDHVV | 11 | | |
| neADA2-pre 237 | 3-43 | 6-6 | 2 | 6 | DMRGREDSSSLRDGYYYGMDV | 21 | 1-47 | 2 | AAWDDSLSGPV | 11 | - | - |
| neADA2-pre 241 | 3-30 | 3-22 | 3 | 6 | GFFLSMIVVVMGGMDV | 16 | 2-14 | 2 | SSYTSSSTLVV | 11 | + | + (C) |
| neADA2-pre 245 | | | | | | | 1-51 | 1 | GTWDSSLSAFYV | 12 | | |
| neADA2-pre 233 | | | | | | | 2-14 | 2 | SSYTSSSTHVV | 11 | | |
| neADA2-pre 240 | | | | | | | 2-23 | 1 | CSYAGSSTSHYV | 12 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | |
| neADA2-pre 41 | 1-8 | 6-6 | 2 | 4 | GPYSSLDY | 8 | | | | | | |
| neADA2-pre 47 | 1-2 | 6-19 | 2 | 4 | YEDSSGLGY | 9 | | | | | | |
| neADA2-pre 122 | 1-2 | 2-15 | 2 | 5 | GDCSGGKRVMNLTNWFDP | 18 | | | | | | |
| neADA2-pre 203 | 3-30 | 5-24 | 3 | 4 | IKGGATILADYFDY | 14 | | | | | | |
| neADA2-pre 235 | 3-7 | 4-17 | 2 | 6 | DSDGDYLDSHYYYGMDV | 17 | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 5:** Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 3 before gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Staining |
| neADA3-pre 01 | 3-30 | 2-2 | 2 | 6 | APYCSSTSCLRRLDYYYGMDV | 21 | 3-11 | 4 | QQRSNWPPLT | 10 | - | - |
| neADA3-pre 02 | 3-48 | 6-6 | 2 | 4 | DRDRYSSSSGDY | 12 | 1-39 | 2 | QQSYSTPYS | 9 | - | + (N) |
| neADA3-pre 03# | 4-59 | 6-19 | 3 | 5 | DRGAVAGTVENWFDP | 15 | 4-1 | 1 | QQYYSTPPT | 9 | | |
| neADA3-pre 05 | 1-69 | / | / | 6 | DRGLGGYYYGMDV | 13 | 1-NL1 | 2 | QQYYSTPAS | 9 | + | - |
| neADA3-pre 06 | 3-49 | 5-12 | 3 | 4 | EGGGRVVALRY | 11 | 1-5 | 1 | QQYNSYSWT | 9 | + | + (C+N) |
| neADA3-pre 16 | 3-9 | 3-3 | 1 | 4 | QPAFLEWSFFDY | 12 | 3-20 | 4 | QQYGSSPST | 9 | - | + (C+N) |
| neADA3-pre 18 | 3-7 | 3-22 | 2 | 4 | SIECFHYYDSSGYYRHFDY | 19 | 2-29 | 3 | MQGIHLPPT | 9 | + | - |
| neADA3-pre 20 | 3-11 | 2-2 | 2 | 6 | SSLWRGYCSSTSCYASYGMDV | 21 | 2-28 | 2 | MQALQTPYT | 9 | - | - |
| neADA3-pre 21 | 1-18 | 6-13 | 2 | 3 | ANRGPYSSSWPGDAFDI | 17 | 1-8 | 1 | QQYYSYPPT | 9 | - | - |
| neADA3-pre 22 | 3-48 | 5-5 | 1 | 4 | DLVQLWPFDY | 10 | 3-15 | 1 | QQYNNWPPWT | 10 | - | - |
| neADA3-pre 23 | 4-39 | 6-19 | 3 | 6 | VCIAVAGHYYYYYGMDV | 17 | 3-11 | 2 | QQRSNWPPYT | 10 | + | + (C) |
| neADA3-pre 25 | 3-66 | 4-23 | 2 | 6 | DGGNSEYYYYGMDV | 14 | 1-8 | 1 | QQYYSYPWT | 9 | - | + (C) |
| neADA3-pre 26 | 3-23 | 6-6 | 1 | 4 | RTQQLVTDY | 9 | 4-1 | 4 | QQYYSTPVT | 9 | - | + (C+N) |
| neADA3-pre 30 | 1-18 | 6-19 | 2 | 4 | VSGSGWYDGY | 10 | 3-20 | 1 | QQYGSSPVT | 9 | - | - |
| neADA3-pre 38 | 3-53 | 6-19 | 3 | 4 | GGVIAVAGSFDY | 12 | 1D-8 | 1 | QQYYSFPWT | 9 | - | + (C+N) |
| neADA3-pre 43# | 1-69 | 3-10 | 2 | 5 | DRYYGSGTLYNWFDP | 15 | 1D-43 | 4 | QQYYSTPRLT | 10 | | |
| neADA3-pre 44 | 3-15 | / | / | 3 | DPYAPYDI | 8 | 1-8 | 2 | QQYYSYPYS | 9 | - | - |
| neADA3-pre 46 | 4-b | 3-22 | 2 | 4 | HTYYYDSSGYPFDY | 14 | 1-39 | 1 | QQSYSTLTWT | 10 | + | + (C+N) |
| neADA3-pre 13 | | | | | | | 1-5 | 1 | QQYNSYPWT | 9 | | |
| neADA3-pre 14 | | | | | | | 3-11 | 2 | QQRSNWPPYT | 10 | | |
| neADA3-pre 17 | | | | | | | 1-39 | 2 | QQSYSTPYT | 9 | | |
| neADA3-pre 42 | | | | | | | 3-20 | 1 | QQYGSSLWT | 9 | | |
| neADA3-pre 45 | | | | | | | 1-5 | 2 | QQYNSYSSYT | 10 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Staining |
| neADA3-pre 04 | 1-2 | 6-19 | 1 | 2 | DKREWLAWYFDL | 12 | 2-14 | 2 | SSYTSSSTLVV | 11 | - | + (C) |
| neADA3-pre 15 | 3-15 | 3-9 | 2 | 4 | DDILTGYYGGDY | 12 | 1-36 | 3 | AAWDDSLNGWV | 11 | + | + (C) |
| neADA3-pre 17 | 3-21 | 3-9 | 3 | 4 | GLVITNGGY | 9 | 6-57 | 2 | QSYDSSNVV | 9 | - | + (C) |
| neADA3-pre 25 | | | | | see kappa | | 2-23 | 3 | CSYAGSSRV | 9 | - | + (C) |
| neADA3-pre 28 | 3-11 | 2-21 | 3 | 4 | DRVTAMLFDY | 10 | 3-21 | 2 | QVWDSSSDHVV | 11 | - | - |
| neADA3-pre 35# | 3-15 | 3-16 | 2 | 6 | DTLPDYDYVWGSYKDYGMDV | 20 | 1-47 | 1 | AAWDDSLSGYV | 11 | | |
| neADA3-pre 39 | 3-23 | 5-5 | 2 | 4 | PGYSYAGDFDY | 11 | 7-46 | 2 | LLSYSGAVV | 9 | + | + (C+N) |
| neADA3-pre 19 | | | | | | | 3-1 | 3 | QAWDSSTAV | 9 | | |
| neADA3-pre 34 | | | | | | | 2-8 | 1 | SSYAGSNIGV | 10 | | |
| neADA3-pre 36 | | | | | | | 1-47 | 2 | AAWDDSLSGPV | 11 | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | |
| neADA3-pre 07 | 4-61 | 1-26 | 3 | 4 | SIVGATTGVFDY | 12 | | | | | | |
| neADA3-pre 08 | 4-61 | / | / | 2 | QWGPVDNWYFDL | 12 | | | | | | |
| neADA3-pre 11 | 3-23 | 1-26 | 2 | 4 | LNHRSGSYDYLDPQFDY | 17 | | | | | | |
| neADA3-pre 33 | 4-39 | 3-10 | 1 | 4 | HPLAWFDRSFDY | 12 | | | | | | |
| neADA3-pre 37 | 3-15 | 1-26 | 2 | 4 | GYSGSYYKY | 9 | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 6:** Repertoire and reactivity of ANA-expressing B cell clones from ADA-SCID patients 1-3 before and after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Hep-2 staining | Chromatin | Crithidia staining |
| neADA1-pre 09 | 3-15 | 3-3 | 1 | 4 | HSPKLQFLEWFCDY | 14 | 3-20 | 2 | QQYGSSPCS | 9 | + (N) | - | - |
| neADA1-pre 33 | 3-23 | 2-15 | 2 | 3 | FYCSGGSCYRDAFDI | 15 | 2-30 | 4 | MQGTHWPT | 8 | + (S) | - | - |
| neADA1-pre 34 | 3-23 | 1-20 | 2 | 4 | RRYNWNDEEQVY | 12 | 3-20 | 2 | QQYGSSPCS | 9 | + (N) | - | - |
| neADA1-pre 45 | 3-30-3 | 6-13 | 2 | 1 | VQGSSWYLGYFQH | 13 | 1-17 | 4 | LQHNSYPPT | 9 | + (N) | + | + (K) |
| neADA1-pre 46 | 3-30-3 | 6-19 | 3 | 2 | DARIAVADPYWYFDL | 15 | 1-8 | 4 | QQYYSYPLLT | 10 | + (N) | - | - |
| neADA2-pre 02 | 3-7 | 6-19 | 2 | 4 | AGYSSAGY | 8 | 1-5 | 2 | QQYNSYSMYT | 10 | + (N) | - | - |
| neADA2-pre 30 | 3-30 | 3-10 | 2 | 4 | STKSQGSGSYYNVRRYFDP | 19 | 3-15 | 2 | QQYNNWPPYT | 10 | + (N) | + | + (K+N) |
| neADA2-pre 209 | 4-4 | 2-8 | 2 | 4 | TVSTNGELNFDY | 12 | 1-8 | 1 | QQYYSYPRT | 9 | + (N) | - | - |
| neADA2-pre 225 | 3-30-3 | 1-26 | 1 | 1 | GVIRWELLILAGEYFQH | 17 | 3-15 | 1 | QQYNNWPPWT | 10 | + (N) | - | - |
| neADA3-pre 02 | 3-48 | 6-6 | 2 | 4 | DRDRYSSSSGDY | 12 | 1-39 | 2 | QQSYSTPYS | 9 | + (N) | - | - |
| neADA3-pre 06 | 3-49 | 5-12 | 3 | 4 | EGGGRVVALRY | 11 | 1-5 | 1 | QQYNSYSWT | 9 | + (C+N) | - | - |
| neADA3-pre 16 | 3-9 | 3-3 | 1 | 4 | QPAFLEWSFFDY | 12 | 3-20 | 4 | QQYGSSPST | 9 | + (C/N) | - | - |
| neADA3-pre 26 | 3-23 | 6-6 | 1 | 4 | RTQQLVTDY | 9 | 4-1 | 4 | QQYYSTPVT | 9 | + (C/N) | - | - |
| neADA3-pre 38 | 3-53 | 6-19 | 3 | 4 | GGVIAVAGSFDY | 12 | 1D-8 | 1 | QQYYSFPWT | 9 | + (C/N) | + | + (K) |
| neADA3-pre 46 | 4-b | 3-22 | 2 | 4 | HTYYYDSSGYPFDY | 14 | 1-39 | 1 | QQSYSTLTWT | 10 | + (C/N) | + | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Hep-2 staining | Chromatin | Crithidia staining |
| neADA2-pre 228 | 3-23 | 3-22 | 2 | 6 | AGLDSSGSKLRYYYYYGMDV | 20 | 3-1 | 2 | QAWDSSTVV | 9 | + (N) | - | - |
| neADA3-pre 39 | 3-23 | 5-5 | 2 | 4 | PGYSYAGDFDY | 11 | 7-46 | 2 | LLSYSGAVV | 9 | + (N+C) | + | + (K) |

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Hep-2 staining | Chromatin | Crithidia staining |
| mADA2-pre 57 | 3-30 | 3-3 | 3 | 4 | GEGRITIFGVVIIKGLIDY | 19 | 2D-29 | 1 | MQSIQRWT | 8 | + (N) | - | - |
| mADA2-pre 71 | 3-30 | 2-2 | 2 | 4 | GRYCSSTSCYLDY | 13 | 1-5 | 1 | QQYNSYPWT | 9 | + (N) | - | - |
| mADA2-pre 85 | 3-30 | 6-19 | 3 | 4 | GEVRAVAGSSC | 11 | 1-5 | 1 | QQYNSYSWT | 9 | + (N) | - | - |
| mADA2-pre 252 | 3-64 | 3-3 | 2 | 4 | GRYDFWSGYQY | 11 | 1-16 | 2 | QQYNSYPYT | 9 | + (N) | - | - |
| mADA2-pre 259 | 4-34 | 6-6 | 3 | 4 | GKGTIAARRSKETGGYYFDY | 20 | 1-39 | 1 | QQSYSIPRT | 9 | + (N) | + | + (K) |
| | | 7-27 | 3 | | | | | | | | | | |
| mADA2-pre 274 | 4-31 | 3-10 | 2 | 4 | EAYYGSGSKQQYSNFDY | 17 | 4-1 | 1 | QQYYSTPRT | 9 | + (N) | - | - |
| mADA2-pre 278 | 3-21 | 3-10 | 3 | 4 | DLWLVRGVIDW | 11 | 3-15 | 1 | QQYNNWPWT | 9 | + (N) | - | - |
| mADA3-pre 67 | 4-31 | / | / | 5 | DRTRNNWFDP | 10 | 1-33 | 4 | QQYDNLPLT | 9 | + (C/N) | - | - |
| mADA3-pre 71 | 3-33 | 2-2 | 3 | 4 | DTGVVPAAYFDY | 12 | 1-27 | 3 | QKYNSAPFT | 9 | + (C/N) | - | - |
| mADA3-pre 79 | 3-53 | 3-22 | 2 | 4 | LYDSSGYYPFDY | 12 | 1-39 | 2 | QQSYSTPPYT | 10 | + (N) | - | - |
| mADA3-pre 87 | 3-33 | 3-3 | 1 | 5 | GGVQWFHP | 8 | 2-30 | 4 | MQGTHWPLT | 9 | + (C/N) | - | - |
| mADA3-pre 92 | 1-2 | 1-26 | 3 | 4 | DHGWGIVGAMVY | 12 | 3-15 | 1 | QQYNNWPPWT | 10 | + (N) | - | - |
| mADA3-pre 95 | 3-30 | 1-26 | 1 | 4 | GELLGY | 6 | 3-15 | 4 | QQYNNWPPLT | 10 | + (C/N) | + | + (K) |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Hep-2 staining | Chromatin | Crithidia staining |
| mADA2-pre 293 | 3-7 | 6-6 | 3 | 4 | GGARRCDY | 8 | 2-8 | 2 | SSYAGSNNFVV | 11 | + (N) | + | + (N+K) |
| mADA3-pre 52 | 5-51 | 3-10 | 3 | 6 | QDRITMVRGVITYYYYYGMDV | 21 | 1-51 | 3 | GTWDSSLSAGV | 11 | + (C/N) | + | + (K) |
| mADA3-pre 96 | 3-21 | 2-21 | 2 | 6 | CGGDCYSDYYGMDV | 14 | 3-25 | 1 | QSADSSGTYV | 10 | + (N) | - | - |

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Hep-2 staining | Chromatin | Crithidia staining |
| neADA2-post 215 | 4-4 | 6-19 | 3 | 6 | DLTAVAGTYYGMDV | 14 | 3-11 | 2 | QQRSNWPPYT | 10 | - | + (N) | + | - |

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Hep-2 staining | Chromatin | Crithidia staining |
| mADA2-post 287 | 3-33 | 2-2 | 2 | 6 | DMGYCSSTSCLGVLKLTYYYGMDV | 24 | 1-39 | 3 | QQSYSTPFT | 9 | + | + (C/N) | - | - |
| mADA3-post 61 | 6-1 | 6-19 | 1 | 4 | EPKPKSQWLVHGAIDY | 16 | 1-39 | 1 | QQSYSTPWT | 9 | - | + (N) | + | + (N+K) |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Hep-2 staining | Chromatin | Crithidia staining |
| mADA3-post 51 | 1-2 | 6-13 | 3 | 4 | IHLKISAAGSKSFYFDY | 17 | 3-21 | 2 | QVWDSSSDHHVV | 12 | - | + (C/N) | - | - |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; K, kinetoplast

**Supplemental Table 7:** Repertoire and reactivity of antibodies from mature naive B cells of HD27

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mHD27 04 | 3-33 | 6-13 | 2 | 4 | VGGWSSSWYGADY | 13 | 2-28 | 4 | MQALQTPLT | 9 | - | - | - |
| mHD27 06# | 3-48 | 2-21 | 3 | 4 | DHIVVVTSYYFDY | 13 | 1-9 | 1 | QQLNSYPRT | 9 | | | |
| mHD27 11# | 1-46 | 5-5 | 2 | 6 | GEEYSYGYRNYYYYYMDV | 18 | 1-39 | 4 | QQSYSNPLT | 9 | | | |
| mHD27 19# | 3-23 | 3-22 | 2 | 6 | DRGFRYDSSPKRDYYYMDV | 19 | 3-15 | 4 | QQYNNWPLT | 9 | | | |
| mHD27 23 | 4-39 | 3-3 | 3 | 5 | ITSTIFGVRDRFDP | 14 | 1-39 | 2 | QQSYSTPS | 8 | - | + | + (F) |
| mHD27 26 | 3-66 | 1-7 | 3 | 6 | GTRSDYYYGMDV | 12 | 2-40 | 2 | MQRIEFPYT | 9 | - | - | - |
| mHD27 28 | 3-48 | 2-2 | 1 | 4 | RGDQLLDWTSAFDY | 14 | 1D-39 | 5 | QQSYSTPSIT | 10 | - | - | - |
| mHD27 29 | 1-18 | 4-4 | 2 | 5 | TPYYSNPDGWFDP | 13 | 4-1 | 1 | QQYYSTPGT | 9 | - | - | - |
| mHD27 30 | 3-64 | 5-24 | 2 | 4 | GPSRDGYNPDY | 11 | 3-15 | 4 | QQYNNWPPLT | 10 | - | - | - |
| mHD27 31# | 3-30 | 3-22 | 3 | 3 | DRVRIVVVIRWAFDI | 15 | 1-33 | 4 | QQYDNLPF | 8 | | | |
| mHD27 33 | 4-34 | 1-7 | 2 | 5 | AFTVIYNWNYGWFDP | 15 | 3-20 | 4 | QQYGSSPT | 8 | - | - | - |
| mHD27 39# | 3-48 | 2-15 | 2 | 3 | DSPSGVGLCSGGSCYLDAFDI | 21 | 3-20 | 2 | QQYGT | 5 | | | |
| mHD27 42 | 3-23 | 2-2 | 2 | 4 | GLGYCSSTSCPLH | 13 | 1-27 | 4 | QKYNSAPLT | 9 | - | - | + (F) |
| mHD27 44 | 3-30 | 3-10 | 1 | 3 | GRTNRPGFGDPDAFDI | 16 | 1-33 | 2 | QQYDNLPVT | 9 | - | - | - |
| mHD27 46 | 3-30 | 6-13 | 1 | 6 | DRAAAGRLGYYYGMDV | 16 | 1-12 | 3 | QQANSFPRT | 9 | - | - | + (N) |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mHD27 02 | 4-39 | 6-13 | 2 | 4 | MPYSSRSLGY | 10 | 2-14 | 1 | SSYTSSSTLHV | 11 | - | - | - |
| mHD27 03 | 3-30-3 | 4-4 | 2 | 6 | ALQDNYYYGMDV | 12 | 2-14 | 3 | SSYTSSSTWV | 10 | - | - | - |
| mHD27 05 | 3-7 | 4-17 | 2 | 3 | GGLYGDYGNDAFDI | 14 | 1-40 | 2 | QSYDSSLSGVV | 11 | - | - | - |
| mHD27 07 | 4-4 | 2-15 | 3 | 6 | GVVVAATHWSMDV | 13 | 6-57 | 3 | QSYDSSNHWV | 10 | - | - | - |
| mHD27 08 | 4-4 | 6-19 | 3 | 4 | SRGIAVAEYYFDY | 13 | 1-40 | 1 | QSYDSSLSGYV | 11 | - | - | - |
| mHD27 09 | 4-59 | 2-2 | 3 | 2 | HVDIVVWYFDL | 11 | 3-1 | 2 | QAWDSSTVV | 9 | - | - | - |
| mHD27 12 | 3-21 | 3-10 | 3 | 5 | IRGAPRGWFDP | 11 | 2-23 | 3 | CSYAGSSV | 8 | + | + | - |
| mHD27 13 | 4-59 | 6-19 | 3 | 2 | SPEIAVAGHYFDY | 13 | 3-25 | 3 | QSADSSGTYEV | 11 | - | - | - |
| mHD27 14 | 1-69 | 1-26 | 2 | 5 | TSGSPVYWFDP | 11 | 1-40 | 3 | QSYDSSLSGSV | 11 | + | - | - |
| mHD27 15 | 3-33 | 5-5 | 2 | 3 | PRRGGYSYGDAFDI | 14 | 2-8 | 2 | SSYAGSNNLV | 10 | - | - | + (C) |
| mHD27 16 | 1-3 | / | / | 4 | DYRRGESRKTGLAFGY | 16 | 1-40 | 3 | QSYDSSLSGWV | 11 | - | + | + (C) |
| mHD27 17 | 3-23 | 3-22 | 3 | 3 | DRSWGRVVVITYAFDI | 16 | 1-51 | 3 | GTWDSSLSAEV | 11 | - | + | + (C) |
| mHD27 18# | 5-a | 3-10 | 1 | 4 | YRVGGGFGELLSDY | 14 | 1-44 | 3 | AAWDDSLNGWV | 11 | | | |
| mHD27 20# | 3-30-3 | 2-21 | 2 | 4 | VDDGSHGAEYCGGDCYQFDY | 20 | 3-9 | 7 | QVWDSSTAV | 9 | | | |
| mHD27 21 | 3-23 | 2-15 | 3 | 4 | DLKDIVVVVAVGDYIDY | 17 | 2-8 | 2 | SSYAGSNKG | 9 | - | - | - |
| mHD27 22 | 5-51 | 3-22 | 2 | 4 | INYYDSNYFDY | 11 | 3-21 | 2 | QVWDSSSDHPV | 11 | - | - | - |
| mHD27 24 | 3-30-3 | 6-19 | 3 | 6 | DLHPRAVAGPYYYYYGMDV | 19 | 3-1 | 2 | QAWDSSNVV | 9 | - | - | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | | |
| mHD27 25# | 1-46 | 3-22 | 2 | 6 | SGVGSYYYDKGMDV | 14 | | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 8:** Repertoire and reactivity of antibodies from mature naïve B cells of HD28

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mHD28 1# | 3-66 | 3-10 | 1 | 4 | FGFGEFLYYFDY | 12 | 3-20 | 1 | QQYGSSLWT | 9 | | | |
| mHD28 4# | 4-30-4 | 3-22 | 2 | 4 | ERGYYYDSSGYYYYFDY | 17 | 1-17 | 1 | LQHNSYPRT | 9 | | | |
| mHD28 8# | 3-48 | 3-22 | 2 | 4 | DAVYYDSSGSCDY | 13 | 3-15 | 1 | QQYNNWPWT | 9 | | | |
| mHD28 10 | 3-49 | 3-22 | 3 | 3 | GLTMIVGEDAFDI | 13 | 1-39 | 4 | QQSYSTPLT | 9 | - | - | - |
| mHD28 11 | 3-30-3 | 6-6 | 3 | 6 | VHWLIAEPRGNGMDV | 15 | 2-28 | 5 | MQALQTPT | 8 | - | + | + (C) |
| mHD28 12 | 4-39 | 1-26 | 3 | 4 | PFLVGATPPEYYFDY | 15 | 3-11 | 4 | QQRSNWPLLT | 10 | - | - | - |
| mHD28 14# | 4-34 | 4-23 | 2 | 4 | ITTAWGGGGYFDY | 13 | 3-20 | 2 | QQYGSSPPMCS | 11 | | | |
| mHD28 16# | 5-51 | / | / | 6 | REGMRGMDV | 9 | 2-28 | 1 | MQALQTPPWT | 10 | | | |
| mHD28 23# | 1-46 | 3-10 | 2 | 4 | VWENYYGSGSLNY | 13 | 2-28 | 5 | MQALQTPT | 8 | | | |
| mHD28 24# | 4-39 | 5-5 | 2 | 4 | LHSGYSYGYGVFDY | 14 | 2-29 | 4 | MQSIQLPLT | 9 | | | |
| mHD28 26 | 3-23 | 5-24 | 1 | 4 | DLSRWLPERGYYFDY | 15 | 3-15 | 1 | QQYNNWPWT | 9 | - | - | - |
| mHD28 28# | 3-33 | 2-2 | 2 | 3 | DTVGGYCSSTSCYRGAFDI | 19 | 1-5 | 1 | QQYNSYPTWT | 10 | | | |
| mHD28 29 | 4-39 | 3-3 | 2 | 4 | ASGSGYPTFDY | 11 | 1-39 | 5 | QQSYSTL | 7 | - | + | - |
| mHD28 33 | 3-30-3 | 4-17 | 2 | 4 | GATYGDYVDY | 10 | 3-11 | 4 | QQRSNWPALT | 10 | - | - | - |
| mHD28 34 | 1-2 | 5-12 | 2 | 4 | RGYSGYDAFDY | 11 | 2-28 | 3 | MQALQTPFT | 9 | - | - | - |
| mHD28 35 | 3-21 | / | / | 4 | DGDDFTGEVY | 10 | 4-1 | 1 | QQYYSTPWT | 9 | - | - | - |
| mHD28 36 | 4-4 | 6-13 | 3 | 5 | GRAAGHNWFDP | 11 | 1-5 | 2 | QQYNSYSPYT | 10 | - | - | - |
| mHD28 39 | 1-69 | 5-24 | 3 | 4 | DREVAKGGFDY | 11 | 4-1 | 4 | QQYYSTPPT | 9 | + | + | - |
| mHD28 41# | 3-73 | 4-4 | 2 | 4 | HAAIYSNYVDY | 11 | 4-1 | 4 | QQYYSTPLT | 9 | | | |
| mHD28 43# | 3-15 | 3-3 | 1 | 4 | DLATELRFLETIDY | 14 | 1D-43 | 5 | QQYYSTL | 7 | | | |
| mHD28 47 | 5-a | 3-22 | 2 | 4 | LREYYYDSSGYYGDY | 15 | 3-15 | 1 | QQYNNWPET | 9 | - | + | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mHD28 5 | 1-2 | / | / | 4 | DRREYGIDY | 9 | 7-43 | 1 | LLYYGGAYV | 9 | - | + | - |
| mHD28 6 | 3-33 | 3-10 | 1 | 5 | GESSWFGEL | 9 | 6-57 | 3 | QSYDSSSWV | 9 | - | - | - |
| mHD28 13 | 4-31 | / | / | 2 | RGVVWYFDL | 9 | 2-14 | 2 | SSYTSSSTLDVV | 12 | - | - | - |
| mHD28 15# | 3-30-3 | 2-2 | 2 | 6 | NHCISTSCYRHYYYYYGMDV | 20 | 1-51 | 3 | GWDSSLSAGGV | 11 | | | |
| mHD28 17 | 3-30 | 3-10 | 2 | 4 | DAYGSGRVGYFDY | 13 | 1-44 | 2 | AAWDDSLNGVV | 11 | - | - | - |
| mHD28 18# | 5-a | 2-15 | 2 | 3 | VDCSGGSCYSPNAFDI | 16 | 2-23 | 1 | CSYAGSSFYV | 10 | | | |
| mHD28 21 | 4-59 | / | / | 3 | HNPIDAFDI | 9 | 1-51 | 1 | GTWDSSLSAYV | 11 | - | - | - |
| mHD28 27 | 3-43 | 5-5 | 3 | 4 | DIWDTAMVTGFDY | 13 | 2-23 | 2 | CSYAGSSPVV | 10 | - | - | - |
| mHD28 35 | | | | | see kappa | | 3-21 | 3 | QVWDSSSDHWV | 11 | - | - | - |
| mHD28 37 | 1-46 | 6-19 | 2 | 4 | DIPGSSGWYYFDY | 13 | 2-14 | 1 | SSYTSSSTLYV | 11 | - | - | - |
| mHD28 38 | 4-59 | 3-22 | 2 | 5 | RSEYYDSWFDP | 11 | 1-47 | 2 | AAWDDSLSGRVV | 12 | - | - | - |
| mHD28 43 | | | | | see kappa | | 3-1 | 2 | QAWDSSTAV | 9 | - | - | - |
| mHD28 46 | 1-69 | 6-6 | 3 | 6 | DLKAQSGVAARPAYYYGMDV | 20 | 3-1 | 2 | QAWDSSTGV | 9 | + | - | - |
| mHD28 48 | 6-1 | 1-7 | 2 | 6 | EPYNWIPVNYYGMDV | 15 | 1-44 | 1 | AAWDDSLNGYV | 11 | - | - | - |
| mHD28 25# | | | | | | | 3-25 | 2 | QSADSSGTVV | 10 | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | | |
| mHD28 30# | 3-30-3 | 6-19 | 3 | 6 | VVAVAGYQDYYYGMDV | 16 | | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 9:** Repertoire and reactivity of antibodies from mature naive B cells of ADA-SCID patient 2 before gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mADA2-pre 57 | 3-30 | 3-3 | 3 | 4 | GEGRITIFGVVIIKGLIDY | 19 | 2D-29 | 1 | MQSIQRWT | 8 | + | + | + (N) |
| mADA2-pre 71 | 3-30 | 2-2 | 2 | 4 | GRYCSSTSCYLDY | 13 | 1-5 | 1 | QQYNSYPWT | 9 | - | + | + (N) |
| mADA2-pre 75 | 3-11 | 4-17 | 2 | 4 | DRPGDYGDYFKDYFDY | 16 | 3-15 | 4 | QQYNNWPLT | 9 | - | - | - |
| mADA2-pre 76 | 3-48 | 6-13 | 2 | 4 | ALSSSGVYFDY | 11 | 3-11 | 4 | QQRSNWPPPLT | 11 | - | - | - |
| mADA2-pre 83 | 3-21 | 5-5 | 3 | 4 | VLVPTDTAMTVGYYFDY | 17 | 3-15 | 1 | QQYNNWPPWT | 10 | - | + | + (C) |
| mADA2-pre 85 | 3-30 | 6-19 | 3 | 4 | GEVRAVAGSSC | 11 | 1-5 | 1 | QQYNSYSWT | 9 | + | + | + (N) |
| mADA2-pre 91# | 3-23 | / | / | 4 | SGV | 3 | 2-24 | 1 | MQATQFPRT | 9 | | | |
| mADA2-pre 252 | 3-64 | 3-3 | 2 | 4 | GRYDFWSGYQY | 11 | 1-16 | 2 | QQYNSYPYT | 9 | + | + | + (N) |
| mADA2-pre 253 | 4-59 | 3-10 | 2 | 4 | SLPRFDTYYYGSGSYSYFDY | 20 | 3-15 | 5 | QQYNNWPPIT | 10 | + | + | - |
| mADA2-pre 258# | 3-23 | / | / | 4 | VDYGDY | 6 | 3-11 | 1 | QQRSNWPSGT | 10 | | | |
| mADA2-pre 259 | 4-34 | 6-6 | 3 | 4 | GKGTIAARRSKETGGYYFDY | 20 | 1-39 | 1 | QQSYSIPRT | 9 | + | + | + (N) |
| | | 7-27 | 3 | | | | | | | | | | |
| mADA2-pre 267 | 3-15 | 3-22 | 2 | 4 | FYYDSSGYNRYYFDY | 15 | 1-39 | 2 | QQSYSTPNT | 9 | + | - | - |
| mADA2-pre 274 | 4-31 | 3-10 | 2 | 4 | EAYYGSGSKQQYSNFDY | 17 | 4-1 | 1 | QQYYSTPRT | 9 | + | + | + (N) |
| mADA2-pre 277# | 1-46 | 2-21 | 2 | 4 | DKGGLDDCGGECYHGAPNDY | 20 | 3-11 | 4 | QQRSNWPLT | 9 | | | |
| mADA2-pre 278 | 3-21 | 3-10 | 3 | 4 | DLWLVRGVIDW | 11 | 3-15 | 1 | QQYNNWPWT | 9 | + | + | + (N) |
| mADA2-pre 287# | 4-34 | 3-10 | 3 | 5 | GTRIYRIMVRGVGGWFDP | 18 | 3-20 | 1 | QQYGSSPT | 8 | | | |
| mADA2-pre 290 | 4-59 | 5-5 | 2 | 2 | GGRFGYSYGSNWYFDL | 16 | 3-20 | 3 | QQYGSSPPFT | 10 | + | + | + (C) |
| mADA2-pre 50 | | | | | | | 1-39 | 1 | QQSYSTPQT | 9 | | | |
| mADA2-pre 51 | | | | | | | 1-16 | 4 | QQYNSYPLT | 9 | | | |
| mADA2-pre 56 | | | | | | | 3-11 | 5 | QQRSNWIT | 8 | | | |
| mADA2-pre 66 | | | | | | | 2-28 | 1 | MQALQTWT | 8 | | | |
| mADA2-pre 73 | | | | | | | 2-28 | 4 | MQALQTSFSLT | 11 | | | |
| mADA2-pre 77 | | | | | | | 1-39 | 2 | QQSYSTYT | 8 | | | |
| mADA2-pre 78 | | | | | | | 1-6 | 1 | LQDYNYPRT | 9 | | | |
| mADA2-pre 89 | | | | | | | 3-20 | 1 | QQYGSSPYVT | 10 | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mADA2-pre 250 | 3-23 | / | / | 4 | EGWRPLFDY | 9 | 2-14 | 3 | SSYTSSSTWV | 10 | - | - | - |
| mADA2-pre 251 | 4-39 | 2-2 | 3 | 5 | QNIVVVPAAIAYWFDP | 16 | 1-51 | 3 | GTWDSSLSAGV | 11 | - | - | - |
| mADA2-pre 260 | 3-23 | 1-26 | 3 | 4 | RKVGATKPIDY | 11 | 3-21 | 7 | QVWDSSSAV | 9 | - | - | - |
| mADA2-pre 269# | 3-7 | 1-26 | 2 | 4 | DWVERDSGSYGY | 12 | 2-14 | 2 | SSYTSSSTLV | 10 | | | |
| mADA2-pre 276 | 4-30-4 | 6-6 | 1 | 4 | ALYEQQLVCFDY | 12 | 1-51 | 3 | GTWDSSLSALL | 11 | - | - | - |
| mADA2-pre 288 | 1-18 | 1-26 | 1 | 4 | DEGWELFAGDY | 11 | 2-18 | 3 | SSYTSSSTWV | 10 | - | - | - |
| mADA2-pre 293 | 3-7 | 6-6 | 3 | 4 | GGARRCDY | 8 | 2-8 | 2 | SSYAGSNNFVV | 11 | + | + | + (N) |
| mADA2-pre 296# | 4-34 | 7-27 | LG | 5 | GLGAVIAADKARFDP | 15 | 1-47 | 3 | AAWDDSLSGRV | 11 | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | | |
| mADA2-pre 257 | 3-66 | 3-10 | 1 | 6 | AKPDERFGELLETFYYGMDV | 20 | | | | | | | |
| mADA2-pre 280 | 3-30 | 1-26 | 1 | 4 | AKTWELLDY | 9 | | | | | | | |
| mADA2-pre 282 | 3-21 | 6-6 | 1 | 4 | VITFSARQLEFDY | 13 | | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 10:** Repertoire and reactivity of antibodies from mature naive B cells of ADA-SCID patient 3 before gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mADA3-pre 49# | 3-66 | / | / | 4 | VLLTVYYYFDY | 11 | 3-15 | 5 | QQYNNWPIT | 9 | | | |
| mADA3-pre 51 | 4-34 | 3-22 | 2 | 4 | GQNYYDSSGSFYYFDY | 16 | 3-20 | 1 | QQYGSSPRT | 9 | - | - | - |
| mADA3-pre 55 | 4-34 | 6-19 | 1 | 4 | EGLVRSYFDY | 10 | 1-39 | 1 | QQSYSTPRT | 9 | - | - | - |
| mADA3-pre 61 | 3-23 | 6-13 | 2 | 4 | DISSSGNGYYFDY | 13 | 1-39 | 1 | QQSYSTPRT | 9 | + | + | - |
| mADA3-pre 62 | 4-b | 2-2 | 2 | 5 | DAPEYCSSTSCSNSNWFDP | 19 | 3-11 | 4 | QQRSNWPLT | 9 | - | - | - |
| mADA3-pre 66 | 3-48 | 5-12 | 2 | 6 | DRRGYSGYDFRFGGMDV | 17 | 1-33 | 4 | QQYDNLPLT | 9 | + | + | + (C) |
| mADA3-pre 67 | 4-31 | / | / | 5 | DRTRNNWFDP | 10 | 1-33 | 4 | QQYDNLPLT | 9 | - | + | + (C+N) |
| mADA3-pre 71 | 3-33 | 2-2 | 3 | 4 | DTGVVPAAYFDY | 12 | 1-27 | 3 | QKYNSAPFT | 9 | - | + | + (C+N) |
| mADA3-pre 74 | 3-21 | 4-23 | 2 | 4 | GIGNSFDY | 8 | 1-8 | 1 | QQYYSYPRT | 9 | + | + | + (C) |
| mADA3-pre 75 | 3-11 | 3-22 | 2 | 4 | EDPFDSSGYLDY | 13 | 1-39 | 3 | QQSYSTPFT | 9 | - | - | - |
| mADA3-pre 77 | 3-49 | 3-16 | 2 | 4 | FDYLVDY | 7 | 3-20 | 2 | QQYGSSPYS | 9 | - | - | - |
| mADA3-pre 78 | 4-31 | 3-22 | 2 | 4 | ESRTGYYDSSPYFDY | 15 | 3-20 | 1 | QQYGSSPRT | 9 | - | + | + (C) |
| mADA3-pre 79 | 3-53 | 3-22 | 2 | 4 | LYDSSGYYPFDY | 12 | 1-39 | 2 | QQSYSTPPYT | 10 | - | + | + (N) |
| mADA3-pre 80# | 3-33 | 6-13 | 3 | 4 | PSRPRSEGIAAAGAYYFDY | 19 | 1-33 | 5 | QQYDNLPFT | 9 | | | |
| mADA3-pre 81# | 3-30 | 1-26 | 2 | 4 | GGSLDY | 6 | 1D-17 | 4 | LQHNSYPLT | 9 | | | |
| mADA3-pre 84 | 1-69 | 1-26 | 3 | 5 | REVVGGWFDP | 10 | 1-33 | 4 | QQYDNLPLT | 9 | + | - | - |
| mADA3-pre 85 | 1-8 | 2-2 | 3 | 6 | GPVPAAITDYYYYYGMDV | 18 | 3-20 | 2 | QQYGSSPPYT | 10 | - | - | - |
| mADA3-pre 87 | 3-33 | 3-3 | 1 | 5 | GGVQWFHP | 8 | 2-30 | 4 | MQGTHWPLT | 9 | + | + | + (C+N) |
| mADA3-pre 88 | 3-66 | / | / | 6 | DRDYYYYYGMDV | 12 | 3-20 | 4 | QQYGSSPRLT | 10 | + | + | + (C) |
| mADA3-pre 91 | 1-69 | 7-27 | 3 | 6 | GYYYGMDV | 8 | 3-15 | 1 | QQYNNWPWT | 9 | + | + | + (C) |
| mADA3-pre 92 | 1-2 | 1-26 | 3 | 4 | DHGWGIVGAMVY | 12 | 3-15 | 1 | QQYNNWPPWT | 10 | + | + | + (N) |
| mADA3-pre 94 | 3-15 | 6-19 | 2 | 4 | VGSSGWLSFDY | 11 | 1-39 | 2 | QQSYSTPYT | 9 | + | + | + (C) |
| mADA3-pre 95 | 3-30 | 1-26 | 1 | 4 | GELLGY | 6 | 3-15 | 4 | QQYNNWPPLT | 10 | + | + | + (C+N) |
| mADA3-pre 50 | | | | | | | 1-5 | 1 | QQYNSYPWT | 9 | | | |
| mADA3-pre 53 | | | | | | | 1-5 | 2 | QQYNSYPYT | 9 | | | |
| mADA3-pre 60 | | | | | | | 3-20 | 4 | QQYGSSPPLT | 10 | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | Poly | Hep2 | Staining |
| mADA3-pre 52 | 5-51 | 3-10 | 3 | 6 | QDRITMVRGVITYYYYYGMDV | 21 | 1-51 | 3 | GTWDSSLSAGV | 11 | + | - | + (C+N) |
| mADA3-pre 54 | 4-31 | 2-21 | 2 | 3 | IYCGGDCYRRDDAFDI | 16 | 1-51 | 2 | GTWDSSLSAVV | 11 | + | + | + (C) |
| mADA3-pre 55 | | | | | see kappa | | 3-1 | 2 | QAWDSSTVV | 9 | - | + | + (C) |
| mADA3-pre 56 | 3-15 | 5-12 | 2 | 4 | HGYSGTRDY | 9 | 4-60 | 3 | ETWDSNTQV | 9 | - | - | - |
| mADA3-pre 63 | 4-39 | 2-2 | 2 | 4 | HCSSTSCYSGDDY | 13 | 2-11 | 3 | CSYAGSYTLRV | 11 | - | - | - |
| mADA3-pre 68 | 3-21 | / | / | 6 | DWSDFYGMDV | 10 | 1-47 | 3 | AAWDDSLSGWV | 11 | - | - | - |
| mADA3-pre 69 | 3-30 | 4-17 | 2 | 3 | RESDYPWEDAFDI | 13 | 8-61 | 3 | VLYMGSGIWV | 10 | - | - | - |
| mADA3-pre 72 | 3-74 | 2-15 | 2 | 4 | VAGYCSGGSCYYFDY | 15 | 3-21 | 2 | QVWDSSSDHVV | 11 | - | - | - |
| mADA3-pre 86 | 4-61 | 3-22 | 2 | 3 | VGNYDSPFDI | 10 | 2-14 | 1 | SSYTSSSTRYV | 11 | + | + | + (C) |
| mADA3-pre 89 | 1-2 | / | / | 3 | TFGRDGFDAFDI | 12 | 1-44 | 3 | AAWDDSLNGPV | 11 | - | - | - |
| mADA3-pre 90 | 1-2 | 3-22 | 2 | 3 | PYDSSGYYYHAFDI | 14 | 2-11 | 1 | CSYAGSYTYV | 10 | + | + | + (C) |
| mADA3-pre 93 | 3-15 | / | / | 4 | RRVGWKVDY | 9 | 3-25 | 2 | QSADSSGTYVV | 11 | + | + | + (F) |
| mADA3-pre 96 | 3-21 | 2-21 | 2 | 6 | CGGDCYSDYYGMDV | 14 | 3-25 | 1 | QSADSSGTYV | 10 | - | + | + (N) |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | | | |
| mADA3-pre 57 | 3-66 | 6-6 | 2 | 2 | ESGSSSYWYFDL | 12 | | | | | | | |
| mADA3-pre 59 | 3-49 | 3-22 | 2 | 3 | EEYYDSSGYPDAFDI | 15 | | | | | | | |
| mADA3-pre 65 | 3-30 | 7-27 | 3 | 4 | DHSAGDGGY | 9 | | | | | | | |
| mADA3-pre 70 | 4-61 | 3-10 | 3 | 5 | VRGARVDP | 8 | | | | | | | |
| mADA3-pre 73 | 3-33 | 6-19 | 2 | 4 | DRDPYSSGWYLDY | 13 | | | | | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 11:** Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 1 after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA1-post 01# | 3-48 | 3-3 | 2 | 3 | ADYDFWSGYYLGAFDI | 16 | 3-20 | 1 | QQYGSSTRT | 9 | + | | |
| neADA1-post 03 | 4-39 | 2-15 | 3 | 4 | RGVGDIVVVVAATYDY | 15 | 1-39 | 3 | QQSYSTPFT | 9 | + | + | + (C) |
| neADA1-post 05 | 3-20 | 2-2 | 2 | 4 | GRDCSSTSCYYFDY | 14 | 3-11 | 4 | QQRSNWPPT | 9 | + | - | - |
| neADA1-post 13 | 4-b | 3-3 | 3 | 4 | LPTIFGVADDY | 11 | 1D-17 | 4 | LQHNSYPLT | 9 | + | - | - |
| neADA1-post 14 | 4-34 | 6-19 | 2 | 4 | GSGRGSGWYYFDY | 13 | 3-20 | 2 | QQYGSSMCS | 9 | - | - | - |
| neADA1-post 16# | 3-21 | 2-8 | 2 | 6 | ATYCTGGVCYNGVEKGYYYYYMDV | 24 | 1D-12 | 1 | QQANSFPWT | 9 | + | | |
| neADA1-post 17# | 1-2 | 3-3 | 2 | 4 | NRFWSGYQIDY | 11 | 2-40 | 1 | MQRIEFQT | 8 | - | | |
| neADA1-post 18 | 1-2 | 6-13 | 3 | 2 | DMKGASAAGLNWYFDL | 16 | 1-5 | 5 | QQYNSYSIT | 9 | + | - | - |
| neADA1-post 19# | 3-11 | 3-3 | 2 | 5 | YDFWSGYYTPNWFDP | 15 | 1-5 | 1 | QQYNSYPWT | 9 | - | | |
| neADA1-post 21# | 4-b | 3-3 | 3 | 6 | DSAVTIFGVVDSYYMDV | 17 | 1-5 | 1 | QQYNSYPT | 8 | - | | |
| neADA1-post 23 | 4-59 | 3-10 | 2 | 6 | HNYYGSGSYYNLYYYYYGMDV | 21 | 4-1 | 4 | QQYYSTPLT | 9 | + | - | - |
| neADA1-post 24 | 4-59 | 6-13 | 2 | 6 | SNSSSSWYDERYGMDV | 16 | 4-1 | 1 | QQYYSTPWT | 9 | + | - | - |
| neADA1-post 25 | 4-39 | 6-13 | 2 | 6 | DGRQDSSSWYYYYGMDV | 17 | 3-11 | 5 | QQRSNWPPVT | 10 | + | + | + (C) |
| neADA1-post 26 | 3-23 | 6-19 | 1 | 4 | GQWLVPYFDY | 10 | 3-11 | 4 | QQRSNWPLT | 9 | + | - | - |
| neADA1-post 29# | 3-23 | 3-16 | 3 | 4 | DSPMITFGGVIVPFDY | 16 | 3-11 | 5 | QQRSNWPPIT | 10 | + | | |
| neADA1-post 35 | 4-59 | 3-10 | 2 | 4 | SPPWRYYYGSGSYVFDY | 17 | 3-15 | 4 | QQYNNWPPLT | 10 | + | + | + (C) |
| neADA1-post 38 | 3-48 | 6-19 | 1 | 4 | DIGQWLVPYYFDY | 13 | 3-15 | 2 | QQYNNWSYT | 9 | + | - | - |
| neADA1-post 42# | 3-30 | 5-12 | 2 | 4 | DRVYSGYDLIDY | 12 | 2-29 | 2 | MQGIHLPT | 8 | + | | |
| neADA1-post 48 | 4-b | 3-16 | 3 | 5 | DPEFGQVRFDP | 11 | 1-9 | 2 | QQLNSYPLCS | 10 | + | + | - |
| neADA1-post 09 | | | | | | | 3-15 | 3 | QQYNNWPT | 8 | - | | |
| neADA1-post 45 | | | | | | | 1-39 | 2 | QQSYSTLRS | 9 | - | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA1-post 02 | 3-53 | / | / | 6 | GYGMDV | 6 | 6-57 | 2 | QSYDSSTHVV | 10 | + | - | - |
| neADA1-post 04 | 3-21 | 3-16 | 1 | 4 | DASRHLGELSSPMGIAVAGVGTFDY | 25 | 2-14 | 2 | SSYTSSSTPVV | 11 | - | - | - |
| | | 6-19 | 3 | | | | | | | | | | |
| neADA1-post 06 | 4-39 | 1-26 | 3 | 4 | ETDLKRVVGATFYFDY | 16 | 1-51 | 2 | GTWDSSLSAGV | 11 | - | - | - |
| neADA1-post 08 | 1-18 | 1-7 | 1 | 3 | VPTTSLELLDAFDI | 14 | 2-23 | 3 | CSYAGSSTWV | 10 | + | - | - |
| neADA1-post 10 | 4-30-4 | 6-19 | 3 | 4 | VAGRHFDY | 8 | 1-51 | 2 | GTWDSSLSAGPYVV | 14 | + | - | - |
| neADA1-post 17 | 1-2 | 3-3 | 2 | 4 | NRFWSGYQIDY | 11 | 2-14 | 1 | SSYTSSSTYV | 10 | - | + | + (C) |
| neADA1-post 22 | 1-46 | 6-6 | 2 | 4 | GYDKEYSSSSECGY | 14 | 1-44 | 1 | AAWDDSLNGYV | 11 | - | - | - |
| neADA1-post 27# | 3-30-3 | 6-6 | 2 | 3 | IEYSSSSRAFDI | 12 | 3-10 | 2 | YSTDSSGNHRV | 11 | + | | |
| neADA1-post 28 | 3-11 | 6-19 | 1 | 3 | DRSATSPWLVRSESHDAFDI | 20 | 2-14 | 3 | SSYTSSSTWV | 10 | - | - | - |
| neADA1-post 30# | 1-2 | 3-9 | 2 | 3 | NMTAYYDILTGYYFSDQEFDAFDI | 24 | 2-14 | 7 | AAWDDSLNGAV | 11 | + | | |
| neADA1-post 32 | 4-30-4 | 2-2 | 2 | 4 | AGYCSSTSCYRGKNYFDY | 18 | 2-14 | 1 | SSYTSSSTYV | 10 | + | - | - |
| neADA1-post 33 | 1-2 | 3-3 | 2 | 4 | LYYDFLFDY | 9 | 1-51 | 1 | GTWDSSLSAGPYV | 13 | - | - | - |
| neADA1-post 36 | 3-13 | 3-16 | 1 | 4 | GLHLGELSLVDY | 12 | 2-14 | 2 | SSYTSSSTLVV | 11 | + | - | - |
| neADA1-post 40 | 3-23 | 4-17 | 3 | 4 | NDPISTVTVDY | 10 | 3-21 | 2 | QVWDSSSDHVV | 11 | - | - | - |
| neADA1-post 47 | 5-51 | 6-13 | 3 | 4 | QPLIAVAGSDY | 11 | 2-8 | 2 | SSYAGTHVV | 9 | + | - | - |
| neADA1-post 29 | | | | | | | 3-25 | 3 | QSADSSGTYWV | 11 | + | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | |
| neADA1-post 44 | 3-7 | 4-17 | 3 | 6 | EPLTTWGYYYYYGMDV | 17 | | | | | + | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 12:** Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 2 after gene therapy

| | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA2-post 02# | 4-4 | / | / | 4 | RETYFGY | 7 | 3-20 | 4 | QQYGSSPLT | 9 | + | | |
| neADA2-post 05# | 1-2 | 3-22 | 2 | 4 | ASSGYPYYFDY | 11 | 3-15 | 1 | QQYNNWPQT | 9 | + | | |
| neADA2-post 22# | 3-33 | 1-7 | 1 | 4 | DELELDFIDY | 10 | 2-28 | 4 | MQALQIPLT | 9 | + | | |
| neADA2-post 38# | 3-9 | 6-13 | 2 | 4 | GSYSSSWYLPFDY | 13 | 1-5 | 2 | QQYNSYPYT | 9 | + | | |
| neADA2-post 41# | 3-30 | 5-5 | 2 | 1 | WGGYSYGTAYFQH | 13 | 3-20 | 1 | QQYGSSLSWT | 10 | + | | |
| neADA2-post 46# | 3-23 | 3-22 | 2 | 4 | DQGYYDSSGYYPLDY | 15 | 1-39 | 1 | QQSYSTPRT | 9 | + | | |
| neADA2-post 48# | 4-59 | / | / | 4 | VHSGRIFDY | 9 | 1-33 | 3 | QQYDNLPFT | 9 | - | | |
| neADA2-post 104# | 1-2 | 3-22 | 2 | 4 | YYDSSGTPDY | 10 | 1-5 | 4 | QQYNSYPLT | 9 | + | | |
| neADA2-post 116 | 3-23 | 1-26 | 3 | 4 | SEAIVGAGVYFDY | 13 | 1-39 | 1 | QQSYSTPWT | 9 | - | - | - |
| neADA2-post 119# | 4-61 | 1-7 | 3 | 4 | DGDITGTTMGLDY | 13 | 1D-17 | 1 | LQHNSYPRT | 9 | + | | |
| neADA2-post 127# | 3-7 | 3-10 | 1 | 4 | DQVFGGLWFGEEDY | 14 | 1-27 | 2 | QKYNSAPYT | 9 | + | | |
| neADA2-post 133 | 3-30 | 7-27 | 3 | 4 | GGAGDESPYYFDY | 13 | 1-8 | 4 | QQYYSYPLT | 9 | + | - | - |
| neADA2-post 135 | 3-43 | 6-19 | 3 | 4 | DPTVAGTRLDY | 11 | 3-15 | 1 | QQYNNWPPWT | 10 | + | - | - |
| neADA2-post 139# | 1-69 | 6-13 | 3 | 4 | AVAAAGRDPGYFDY | 14 | 3-20 | 4 | QQYGSSPLT | 9 | + | | |
| neADA2-post 143 | 3-48 | 6-19 | 2 | 4 | DGSPHSGWAPPYFDY | 15 | 1-39 | 2 | QQSYSTLMYT | 10 | - | - | - |
| neADA2-post 201 | 3-30 | 2-2 | 3 | 6 | DPLQVPAAESDYYYGMDV | 18 | 2-30 | 4 | MQGTHWPLT | 9 | + | - | - |
| neADA2-post 202 | 1-18 | 5-5 | 2 | 4 | THSSYSYGRRGAGAFDY | 17 | 3-15 | 2 | QQYNNWPYT | 9 | - | + | - |
| neADA2-post 203 | 3-48 | 4-17 | 2 | 4 | DGDYGDYQHSSGNY | 14 | 2-28 | 2 | MQALQTPYT | 9 | + | - | - |
| neADA2-post 206# | 1-3 | 4-4 | 2 | 4 | MADDDYSNRYFDY | 13 | 4-1 | 1 | QQYYSTPWT | 9 | - | | |
| neADA2-post 207# | 1-69 | 3-22 | 2 | 6 | DPPGYYDSSGYPDV | 14 | 1-17 | 4 | LQHNSYPPLT | 10 | + | | |
| neADA2-post 212 | 3-66 | 3-22 | 2 | 4 | VGDYYDSSGYSVLD | 14 | 1-8 | 4 | QQYYSYPLT | 9 | + | - | - |
| neADA2-post 214 | 3-43 | 3-22 | 2 | 4 | DSWYYDSSGYFDY | 13 | 2-30 | 2 | MQGTHWPPYT | 10 | + | - | - |
| neADA2-post 215 | 4-4 | 6-19 | 3 | 6 | DLTAVAGTYYGMDV | 14 | 3-11 | 2 | QQRSNWPPYT | 10 | - | - | + (N) |
| neADA2-post 217 | 4-59 | 4-23 | 3 | 3 | DQTTVANAFDI | 11 | 3-15 | 4 | QQYNNWPL | 8 | + | - | - |
| neADA2-post 218 | 1-46 | 2-8 | 2 | 6 | DNRGYCTNGVCNYYYGMDV | 19 | 1-39 | 1 | QQSYSTPWT | 9 | + | - | - |
| neADA2-post 220 | 3-43 | 6-13 | 3 | 6 | DQSVAAAGTLGYYYYYGMDV | 19 | 2D-29 | 1 | MQSIQLPRT | 9 | + | - | - |
| neADA2-post 226 | 3-15 | 2-2 | 3 | 6 | RGNEVPAAIPGWDGMDV | 17 | 2D-28 | 2 | MQALQTPMYT | 10 | - | - | - |
| neADA2-post 228 | 1-58 | 6-13 | 3 | 4 | DGIAAAGNQLDY | 12 | 2-28 | 4 | MQALQTLT | 8 | + | - | + (C) |
| neADA2-post 231 | 3-53 | 6-19 | 1 | 4 | SSEQWLARGYYFDY | 14 | 1-5 | 2 | QQYNSLYT | 8 | + | + | + (C) |
| neADA2-post 235 | 3-30 | 1-26 | 2 | 6 | DGRDAYSGSYGGGMDV | 16 | 2-28 | 4 | MQALQTPQLT | 10 | + | - | - |
| neADA2-post 238 | 4-39 | 6-13 | 2 | 4 | RYSSSWIDY | 9 | 1-5 | 1 | QQYNSYSWT | 9 | + | + | + (C) |
| neADA2-post 240 | 4-34 | 2-15 | 3 | 4 | GRILVVVVAATPDYFDY | 17 | 1-17 | 4 | LQHNSYPLT | 9 | - | - | - |
| neADA2-post 241 | 4-59 | 7-27 | 3 | 4 | ETTGGPTDY | 9 | 1-5 | 4 | QQYNSYPLT | 9 | + | - | - |
| neADA2-post 244# | 4-34 | 3-9 | 2 | 4 | GRVHYDILTGYLYYFEY | 17 | 3-11 | 3 | QQRSNWPPGGLFT | 13 | + | | |
| neADA2-post 245 | 4-59 | 3-9 | 2 | 4 | GSWVDYDILTGYYLD | 15 | 3-11 | 3 | QQRSNWVIT | 9 | + | + | + (C) |
| neADA2-post 248 | 4-4 | 5-12 | 2 | 4 | GDYSGYDPNGYFDY | 14 | 3-11 | 4 | QQRSNWPLT | 9 | - | - | - |
| neADA2-post 12 | | | | | | | 1-6 | 1 | LQDYNYPRT | 9 | + | | |
| neADA2-post 16 | | | | | | | 1-39 | 1 | QQSYSTPWT | 9 | + | | |
| neADA2-post 31 | | | | | | | 1-33 | 3 | QQYDNLPF | 8 | + | | |
| neADA2-post 40 | | | | | | | 1-39 | 4 | QQSYSTPLT | 9 | + | | |
| neADA2-post 224 | | | | | | | 3-20 | 3 | QQYGSSPFT | 9 | + | | |
| neADA2-post 232 | | | | | | | 1-5 | 2 | QQYNSYSPYT | 10 | + | | |
| neADA2-post 234 | | | | | | | 1-27 | 1 | QKYNSAPLT | 9 | + | | |
| neADA2-post 242 | | | | | | | 1-39 | 2 | QQSYSTPLT | 9 | + | | |
| neADA2-post 243 | | | | | | | 1-17 | 2 | LQHNSYQYT | 9 | + | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA2-post 09# | 1-46 | 5-5 | 3 | 4 | VDTAMRQGDY | 10 | 3-21 | 1 | QVWDSSSDHYV | 11 | + | | |
| neADA2-post 105 | 3-9 | / | / | 4 | DKGMG | 5 | 4-69 | 3 | QTWGTGIHWV | 10 | + | - | - |
| neADA2-post 205# | 3-30 | 3-9 | 2 | 4 | DRGYDILTGYSDY | 13 | 3-21 | 3 | QVWDSSSDHPV | 11 | + | | |
| neADA2-post 209 | 3-74 | 1-26 | 2 | 4 | DTLHSGSYWPDY | 12 | 3-21 | 2 | QVWDSSSDHVV | 11 | + | - | - |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | |
| neADA2-post 03 | 3-23 | 2-2 | 3 | 5 | DLREDVVVVPLGGWFDP | 17 | | | | | + | | |
| neADA2-post 210 | 3-48 | / | / | 3 | DSAIRRYSDAFDI | 13 | | | | | + | | |
| neADA2-post 211 | 3-15 | 4-17 | 3 | 4 | EATVTTFIEIRNFDY | 15 | | | | | + | | |
| neADA2-post 225 | 3-23 | 2-2 | 2 | 4 | AYGYCSSTSCYTEGYYFDY | 19 | | | | | + | | |
| neADA2-post 233 | 4-59 | / | / | 4 | DGDGFDY | 7 | | | | | + | | |
| neADA2-post 236 | 3-66 | / | / | 4 | DLSFSN | 6 | | | | | + | | |
| neADA2-post 239 | 4-34 | 6-13 | 3 | 4 | GEAGIAAAGALYYFDY | 16 | | | | | + | | |
| neADA2-post 246 | 3-13 | 6-19 | 3 | 6 | ALRGAVAGILSGILSGDYYYGMDV | 24 | | | | | + | | |
| neADA2-post 08 | 1-46 | / | / | 4 | ESSYYEY | 7 | | | | | + | | |
| neADA2-post 123 | 4-39 | 5-5 | 2 | 4 | LQGYSYGIDY | 10 | | | | | + | | |
| neADA2-post 204 | 1-2 | 3-10 | 3 | 4 | EVKITMVRGYYFDY | 14 | | | | | + | | |
| neADA2-post 213 | 3-7 | 4-17 | 3 | 6 | DHTVTTYYYGMDV | 13 | | | | | + | | |
| neADA2-post 221 | 4-39 | 2-8 | 2 | 1 | LGYCTNGVCPVFQH | 14 | | | | | + | | |
| neADA2-post 223 | 4-59 | / | / | 4 | AASYFDY | 7 | | | | | + | | |
| neADA2-post 229 | 3-66 | 7-27 | 2 | 4 | DLGDTGYFDY | 10 | | | | | - | | |
| neADA2-post 230 | 4-30-4 | 3-22 | 2 | 4 | APANFNYYDSSGYSY | 15 | | | | | + | | |
| neADA2-post 237 | 3-43 | 4-23 | 3 | 6 | IMSARDTVVTTYRGDLYYYYYGMDV | 25 | | | | | + | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 13:** Repertoire and reactivity of antibodies from new emigrant B cells of ADA-SCID patient 3 after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA3-post 08 | 1-2 | 3-22 | 2 | 4 | DGGTYYYDSSGYRFDY | 16 | 3-20 | 4 | QQYGSSPLT | 9 | + | - | - |
| neADA3-post 10 | 3-23 | 2-15 | 2 | 4 | TSAGYCSGGSCLFILEFWPYFDY | 23 | 3-15 | 1 | QQYNNWPPWT | 10 | + | + | + (C) |
| neADA3-post 12# | 3-23 | 2-21 | 2 | 3 | GFASYCGGDCHVGDAFDI | 18 | 1-8 | 5 | QQYYSYPIT | 9 | + | | |
| neADA3-post 13 | 3-21 | 1-26 | 2 | 3 | ADDSGPYSGSYYPSPGAFDI | 20 | 1-5 | 1 | QQYNSYST | 8 | + | - | - |
| neADA3-post 14 | 3-21 | 1-20 | 2 | 4 | DFGSGGNWNDPLDY | 14 | 2-28 | 4 | MQALQTPQLT | 10 | - | - | - |
| neADA3-post 15 | 4-39 | 2-15 | 2 | 4 | TKDLGYCSGGSCYSYFYY | 18 | 3-15 | 2 | QQYNNWPPYS | 10 | + | - | - |
| neADA3-post 16 | 3-9 | 1-26 | 2 | 1 | DSGSYFEAEYFQH | 13 | 3-20 | 5 | QQYGSSPT | 8 | + | - | - |
| neADA3-post 17 | 3-7 | 1-7 | 2 | 4 | ETNWIGY | 7 | 1-27 | 3 | QKYNSAPPNT | 10 | + | - | - |
| neADA3-post 18 | 1-69 | 3-22 | 2 | 4 | HSSGYFFDY | 9 | 1-39 | 1 | QQSYSTPRT | 9 | + | + | + (C) |
| neADA3-post 20 | 1-69 | 3-22 | 2 | 1 | TYYYDSSGYLQDPVGYFQH | 19 | 4-1 | 4 | QQYYSTPLT | 9 | + | - | - |
| neADA3-post 22 | 3-43 | 6-19 | 3 | 3 | GNQARIGPISIAVAVPKDAFDI | 22 | 2-30 | 2 | MQGTHWPPYS | 10 | + | - | - |
| neADA3-post 23 | 4-31 | 3-10 | 3 | 5 | DPGVRGVSYWFDP | 13 | 4-1 | 3 | QQYYSTPPT | 9 | + | - | - |
| neADA3-post 25 | 3-15 | 3-22 | 2 | 4 | DLPLDYYDSSGYYDVY | 16 | 3-11 | 1 | QQRSNWPWT | 9 | + | - | + (C) |
| neADA3-post 27 | 3-30 | 6-13 | 2 | 4 | EKYSSSWYVYYFDY | 14 | 3-11 | 4 | QQRSNWPLT | 9 | + | + | - |
| neADA3-post 28 | 3-30 | 6-19 | 2 | 6 | DQQGYSSGWSTTDLYYYYYYGMDV | 24 | 1-8 | 2 | QQYYSYPYS | 9 | + | + | + (C) |
| neADA3-post 36 | 4-31 | 2-2 | 3 | 6 | DWVVVVPAAKTFGTNNYYYGMDV | 23 | 1-39 | 2 | QQSYSTPYS | 9 | - | - | - |
| neADA3-post 37 | 1-8 | 3-3 | 1 | 4 | APPRFLEFN | 9 | 1-39 | 4 | QQSYSTPLT | 9 | - | - | - |
| neADA3-post 39 | 1-2 | 1-26 | 2 | 4 | DTTRRGSYDRGGYDY | 15 | 4-1 | 3 | QQYYSTFPT | 9 | + | - | + (C) |
| neADA3-post 40 | 1-2 | 3-10 | 3 | 6 | ERMVPVKEYYYYYGMDV | 17 | 1-5 | 4 | QQYNSYSLT | 9 | + | - | + (C) |
| neADA3-post 41# | 1-69 | 2-2 | 2 | 3 | RYCSSTSCYSGAFDI | 15 | 3-15 | 1 | QQYNNWPPWT | 10 | - | | |
| neADA3-post 42 | 4-34 | 7-27 | 1 | 2 | GKEEKLGTPPFYWYFDL | 17 | 1-5 | 4 | QQYNSYPLT | 9 | + | - | - |
| neADA3-post 43 | 3-23 | 6-6 | 2 | 4 | DRGWYSSSSNYFDY | 14 | 3-11 | 5 | QQRSNWPIT | 9 | + | + | - |
| neADA3-post 44 | 3-53 | / | / | 4 | VGRLDYFDY | 9 | 3-20 | 2 | QQYGSSPMYS | 10 | + | - | - |
| neADA3-post 46# | 5-51 | 2-2 | 3 | 3 | QDIVVVPAAMDAFDI | 15 | 3-20 | 1 | QQYGSSPRT | 9 | - | | |
| neADA3-post 07 | | | | | | | 1-8 | 1 | QQYYSYPRT | 9 | + | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Staining |
| neADA3-post 02 | 3-33 | / | / | 6 | ESSGMDV | 7 | 1-40 | 1 | QSYDSSLSAPYV | 12 | + | - | - |
| neADA3-post 04 | 5-51 | 5-5 | 3 | 4 | QVGDTAMVSFNYFDY | 15 | 2-11 | 2 | CSYAGSYTVV | 10 | + | - | - |
| neADA3-post 05 | 3-11 | 2-2 | 2 | 5 | DNCMYGSSTSCYDP | 14 | 1-44 | 1 | AAWDDSLNGYV | 11 | - | - | - |
| neADA3-post 06# | 3-15 | 3-3 | 3 | 4 | DPESITIFGVVIY | 13 | 2-14 | 1 | SSYTSSSTYV | 10 | - | | |
| neADA3-post 07 | 4-34 | 3-22 | 2 | 4 | LSYYDSSGEGDY | 12 | 1-47 | 3 | AAWDDSLSGSWV | 12 | + | - | - |
| neADA3-post 08 | 1-2 | 3-22 | 2 | 4 | DGGTYYYDSSGYRFDY | 16 | 3-1 | 2 | QAWDSSTVV | 9 | + | - | + (C) |
| neADA3-post 11 | 3-48 | 6-19 | 2 | 4 | ALSSSGVYFDY | 11 | 3-1 | 3 | QAWDSSTAWV | 10 | - | - | - |
| neADA3-post 19 | 1-8 | 3-10 | 1 | 4 | GMIVGEGVFFGDY | 13 | 3-25 | 3 | QSADSSGTWV | 10 | + | - | - |
| neADA3-post 21 | 4-31 | 3-22 | 2 | 2 | SYDSSGYWYFDL | 12 | 2-14 | 3 | SSYTSSSTLGV | 11 | - | - | - |
| neADA3-post 24 | 4-4 | 2-15 | 2 | 6 | DPGYCSGGSCYLDYYYDMDV | 20 | 2-23 | 3 | CSYAGSSTFEV | 11 | + | - | - |
| neADA3-post 29 | 4-31 | / | / | 5 | RGSANNWFDP | 10 | 3-21 | 2 | QVWDSSSDHVV | 11 | + | - | - |
| neADA3-post 37# | 1-8 | 3-3 | 1 | 4 | APPRFLEFN | 9 | 3-25 | 2 | QSADSSGTYQV | 11 | + | | |
| neADA3-post 38 | 3-9 | 3-22 | 2 | 4 | MKGSSGYYPFDY | 12 | 3-21 | 3 | QVWDSSSDRNWV | 12 | + | - | - |
| neADA3-post 45 | 1-2 | 3-3 | 2 | 5 | DRSGASIRFNWFDP | 14 | 1-44 | 2 | AAWDDSHVV | 9 | + | - | + (C) |
| neADA3-post 47 | 4-39 | 4-17 | 2 | 5 | RDGDYPFNWFDP | 12 | 1-40 | 2 | QSYDSSLSVV | 10 | - | - | - |
| neADA3-post 30 | | | | | | | 3-1 | 2 | QAWDSSTVV | 9 | + | | |
| neADA3-post 31 | | | | | | | 3-1 | 3 | QAWDSSTAWV | 10 | + | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | |
| neADA3-post 09 | 3-11 | 2-15 | 2 | 6 | DGYRGCSGGSCYSSYYGMDV | 20 | | | | | + | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 14:** Repertoire and reactivity of antibodies from mature naive B cells of ADA-SCID patient 1 after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA1-post 51 | 7-4-1 | 6-19 | 2 | 5 | GGLGYSSVEWWFDP | 14 | 3-15 | 1 | QQYNNWPRT | 9 | - | - | - | - |
| mADA1-post 52 | 4-b | 6-13 | 2 | 4 | EEISSSWYSGGVDY | 14 | 1-27 | 3 | QKYNSAPFT | 9 | + | + | + | - |
| mADA1-post 58# | 4-59 | 4-17 | 2 | 4 | HDDYGDYYFDY | 11 | 1-39 | 2 | QQSYSTPYT | 9 | + | | | |
| mADA1-post 62# | 4-34 | 4-17 | 3 | 3 | GLLLRTVTTRVAFDI | 15 | 1-16 | 3 | QQYNSYPFT | 9 | + | | | |
| mADA1-post 63# | 1-18 | 2-15 | 3 | 3 | GRSDIVVVVAAELDAFDI | 18 | 1-27 | 3 | QKYNSAPFT | 9 | + | | | |
| mADA1-post 67 | 4-34 | 6-13 | 3 | 5 | GIAAAGTFRDPNWFDP | 16 | 4-1 | 1 | QQYYSTPWT | 9 | + | - | - | - |
| mADA1-post 70# | 1-2 | 6-6 | 2 | 4 | LLYSSSSWDFDY | 12 | 1-16 | 5 | QQYNSYPIT | 9 | + | | | |
| mADA1-post 72 | 3-15 | 3-10 | 2 | 6 | DYGSGSYYYYYYGMDV | 16 | 1-33 | 3 | QQHDNLPLT | 9 | - | - | - | - |
| mADA1-post 75 | 3-9 | 2-15 | 2 | 4 | DITSSGGSGGSCYSS | 15 | 3-15 | 2 | QQYNNWPCS | 9 | + | + | + | + (C) |
| mADA1-post 81# | 4-59 | 6-13 | 3 | 4 | ETHIAAAGRAYYFDY | 15 | 3-20 | 5 | QQYGSSIT | 8 | + | | | |
| mADA1-post 84# | 3-15 | 2-15 | 2 | 4 | RPYCSGGSCYSFFHY | 15 | 3-15 | 1 | QQYNNWPPT | 9 | - | | | |
| mADA1-post 86 | 3-21 | 3-3 | 3 | 4 | RVFGVANLYYFDY | 13 | 3-11 | 4 | QQRSNWPLT | 9 | + | + | - | - |
| mADA1-post 87# | 3-23 | 5-12 | 3 | 4 | DRVNDDY | 7 | 3-11 | 1 | QQRSNWPPWT | 10 | - | | | |
| mADA1-post 88# | 4-b | 3-22 | 2 | 5 | EGYFLPLDYDSSP | 13 | 3-11 | 1 | QQRSNWPPWT | 10 | + | | | |
| mADA1-post 90# | 1-3 | 1-26 | 2 | 3 | YSGSYFLDAFDI | 12 | 3-20 | 4 | QQYGSSLT | 8 | + | | | |
| mADA1-post 92# | 1-2 | 2-15 | 3 | 3 | ASIVVVVAATRIYDAFDI | 18 | 2-28 | 2 | MQALQTPRT | 9 | + | | | |
| mADA1-post 54 | | | | | | | 3-20 | 4 | QQYGSSPLT | 9 | + | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA1-post 55 | 1-8 | / | / | 2 | GGVNIFDL | 8 | 1-36 | 1 | AAWDDSLNGYV | 11 | + | - | - | - |
| mADA1-post 59 | 4-4 | / | / | 4 | VNIWIDY | 7 | 2-11 | 1 | CSYAGSYTFPYV | 12 | + | + | + | + (C) |
| mADA1-post 65 | 3-48 | 2-15 | 3 | 3 | DLPRLPLRVVAATFAFDI | 18 | 2-14 | 2 | SSYTSSSTLVV | 11 | + | - | - | - |
| mADA1-post 66# | 1-18 | 4-17 | 2 | 4 | RAYGDYLPGFDY | 12 | 7-43 | 2 | LLYYGGAQLV | 10 | + | | | |
| mADA1-post 68 | 4-59 | 4-17 | 2 | 1 | SYGDYVVEYFQH | 12 | 2-11 | 2 | CSYAGSYTLV | 10 | + | - | - | - |
| mADA1-post 71 | 4-39 | 6-19 | 3 | 4 | LVLYSMTGIDY | 11 | 2-23 | 2 | CSYAGSSTLV | 10 | - | - | - | - |
| mADA1-post 74 | 1-18 | 3-10 | 1 | 4 | NGLLWFRELLSLYYFDY | 17 | 1-44 | 3 | AAWDDSLNGWV | 11 | + | + | + | + (C) |
| mADA1-post 77# | 3-33 | 2-21 | 2 | 4 | EGNAYCGGDCPFDY | 14 | 3-21 | 2 | QVWDSSSDHHVV | 12 | + | | | |
| mADA1-post 80 | 1-69 | 4-23 | 2 | 4 | SQGRTYDYGGNSPFDY | 16 | 2-11 | 2 | CSYAGSYTSVV | 11 | - | + | - | - |
| mADA1-post 83 | 1-8 | 4-17 | 2 | 6 | ASYGDYYYYYYMDV | 14 | 1-44 | 2 | AAWDDSLNGVV | 11 | + | - | - | - |
| mADA1-post 91 | 3-11 | 7-27 | 3 | 2 | ELTEPYWYFDL | 11 | 2-8 | 1 | SSYAGSNKLGV | 11 | + | - | - | - |
| mADA1-post 96# | 1-2 | 3-10 | 2 | 5 | VAYYGSGSLNWFDP | 14 | 3-21 | 3 | QVWDSSSDHWV | 11 | - | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | | |
| mADA1-post 57 | 3-23 | 6-6 | 2 | 4 | DRWEGRSSSFSPDY | 14 | | | | | - | | | |
| mADA1-post 69 | 3-9 | 6-13 | 3 | 4 | ASSLIAAAGFDY | 12 | | | | | + | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 15:** Repertoire and reactivity of antibodies from mature naive B cells of ADA-SCID patient 2 after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA2-post 85# | 4-39 | 6-19 | 3 | 4 | HRIAVAGDFDY | 11 | 1-33 | 4 | QQSGT | 5 | + | | | |
| mADA2-post 186 | 3-30 | 3-10 | 1 | 4 | DPRRFGELFPYYFDY | 15 | 1-5 | 2 | QQYNSYSMYT | 10 | + | - | - | - |
| mADA2-post 255 | 3-30 | 3-22 | 2 | 3 | NYYDSSGWPFDAFDI | 15 | 4-1 | 2 | QQYYSTPHT | 9 | - | - | - | - |
| mADA2-post 258 | 3-64 | 2-15 | 2 | 2 | ANCSGGSCYYNWYFDL | 16 | 1-8 | 2 | QQYYSYPYT | 9 | - | - | - | - |
| mADA2-post 265 | 3-21 | 6-13 | 1 | 4 | ENEQQLVRELDY | 12 | 1-5 | 1 | QQYNSYSRT | 9 | + | - | - | - |
| mADA2-post 267 | 3-43 | 1-26 | 2 | 4 | DSPSSGSYRYFDY | 13 | 1-33 | 2 | QQYDNLPYT | 9 | - | - | - | - |
| mADA2-post 275 | 3-53 | 4-4 | 3 | 6 | DVFRTTTVTTYYGMDV | 16 | 1-9 | 4 | QQLNSYPPLT | 10 | - | - | - | - |
| mADA2-post 280 | 3-11 | 3-10 | 3 | 6 | GLDTMVRGVIIDNHNYYYGMDV | 22 | 2-28 | 3 | MQALQTPIFT | 10 | - | - | - | - |
| mADA2-post 283 | 3-21 | 6-13 | 2 | 4 | DQVPRYSSSRFFYYFDY | 17 | 3-15 | 1 | QQYNNWPRAT | 10 | + | + | + | + (C) |
| mADA2-post 284# | 3-23 | 2-2 | 3 | 4 | DLILDIVVVPAAILLDY | 17 | 3D-15 | 5 | QQYNNWPPVT | 10 | + | | | |
| mADA2-post 285# | 1-58 | / | / | 6 | EPKTVFNGAEHGMDV | 15 | 1-39 | 4 | QQSYSTP | 7 | + | | | |
| mADA2-post 287 | 3-33 | 2-2 | 2 | 6 | DMGYCSSTSCLGVLKLTYYYGMDV | 24 | 1-39 | 3 | QQSYSTPFT | 9 | + | + | + | + (C+N) |
| mADA2-post 290 | 3-48 | 4-23 | 2 | 3 | PGGDYGNDAFDI | 12 | 3-11 | 3 | QQRSNWPPGGL | 11 | + | - | - | - |
| mADA2-post 291 | 3-30-3 | 3-22 | 2 | 3 | AYYYDSSGYYVRAFDI | 16 | 1-5 | 3 | QQYNSYSGFT | 10 | + | - | - | - |
| mADA2-post 293# | 1-69 | 1-7 | 1 | 4 | VRLEGMGKRYYFDY | 14 | 3-15 | 1 | QQYNNWPPWT | 10 | + | | | |
| mADA2-post 295 | 1-69 | 3-3 | 2 | 4 | ETLQEERSGYLDY | 13 | 3-11 | 4 | QQRSNWPLT | 9 | + | - | - | - |
| mADA2-post 64 | | | | | | | 3-20 | 3 | QQYGSSAFT | 9 | - | | | |
| mADA2-post 73 | | | | | | | 1-5 | 2 | QQYNSYSRA | 9 | + | | | |
| mADA2-post 96 | | | | | | | 1-5 | 2 | QQYNSYLYT | 9 | + | | | |
| mADA2-post 52 | | | | | | | 1-33 | 4 | QQYDNLPLT | 9 | + | | | |
| mADA2-post 66 | | | | | | | 3-15 | 1 | QQYNNWPPTWT | 11 | + | | | |
| mADA2-post 273 | | | | | | | 1-33 | 2 | QQYDNLYT | 8 | - | | | |
| mADA2-post 279 | | | | | | | 1-39 | 1 | QQSYSTPRT | 9 | + | | | |
| mADA2-post 281 | | | | | | | 1-33 | 4 | QQYDNLLT | 8 | - | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA2-post 53# | 4-4 | 3-10 | 2 | 4 | SRTYYYGSGSYYNGPYYFDY | 20 | 2-14 | 2 | SSYTSSSTLDVV | 12 | - | | | |
| mADA2-post 56 | 3-48 | 6-13 | 3 | 4 | IPLAADFDY | 9 | 2-23 | 1 | CSYAGSSTHYV | 11 | - | - | - | - |
| mADA2-post 68 | 1-69 | 5-24 | 2 | 5 | ESEDGYNYGNWFDP | 14 | 1-40 | 3 | QSYDSSLSGWV | 11 | + | - | - | - |
| mADA2-post 153# | 3-7 | / | / | 4 | EGGPPSVDLDY | 11 | 1-47 | 7 | AAWDDSLSGAV | 11 | + | | | |
| mADA2-post 156# | 4-59 | 6-19 | 2 | 2 | LGVSGWYESWYFDL | 14 | 1-40 | 3 | QSYDSSLSGWV | 11 | - | | | |
| mADA2-post 157 | 4-30-2 | / | / | 2 | GINWYFDL | 8 | 3-21 | 3 | QVWDSSSDHPV | 11 | - | + | - | - |
| mADA2-post 253 | 3-33 | 3-16 | 1 | 6 | DVAQGGELSPYYYYYGMDV | 19 | 2-18 | 3 | SSYTSSSTLV | 10 | + | - | - | - |
| mADA2-post 260 | 3-23 | 5-5 | 2 | 4 | DLGYSYGYQYYFDY | 14 | 2-14 | 3 | SSYTSSSTRV | 10 | + | - | - | - |
| mADA2-post 263 | 3-23 | 6-19 | 2 | 4 | ALPMYSSGGIYYFDY | 15 | 2-8 | 2 | SSYAGSNNLV | 10 | + | - | - | - |
| mADA2-post 274 | 3-30-3 | 3-10 | 1 | 4 | GRSVKFGELLFVDY | 14 | 2-18 | 3 | SSYTSSSTWV | 10 | + | - | - | - |
| mADA2-post 286 | 3-21 | 3-10 | 2 | 4 | SGNRGSGLSYLDY | 13 | 3-21 | 3 | QVWDSSSDHWV | 11 | - | - | - | - |
| mADA2-post 294# | 1-69 | 1-1 | 1 | 4 | VRLEGMGKRYYFDY | 14 | 7-43 | 3 | LLYYGGPWV | 9 | + | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | | |
| mADA2-post 87 | 3-21 | 6-19 | 2 | 5 | DGSYSSGWYGGNWFDP | 16 | | | | | + | | | |
| mADA2-post 169 | 1-46 | 3-22 | 2 | 4 | VQSYDSSGYPNGYFDY | 16 | | | | | + | | | |
| mADA2-post 180 | 3-11 | 5-24 | 3 | 4 | DPVLGMATIGDYFDY | 15 | | | | | + | | | |
| mADA2-post 190 | 3-7 | 3-10 | 1 | 4 | ADESPPPWFGELFRVRLYYFDY | 22 | | | | | - | | | |
| mADA2-post 256 | 3-21 | 2-15 | 2 | 6 | FQYCSGGSCLGMDV | 14 | | | | | + | | | |
| mADA2-post 277 | 1-69 | 3-22 | 2 | 4 | VGVGYYDSSGYYSPFDY | 17 | | | | | + | | | |
| mADA2-post 292 | 3-53 | 2-2 | 2 | 4 | GSLPFSAFDY | 10 | | | | | + | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 16:** Repertoire and reactivity of antibodies from mature naive B cells of ADA-SCID patient 3 after gene therapy

| Ig | HEAVY | | | | | | LIGHT | | | | GT | REACTIVITY | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | VH | D | RF | JH | CDR3 (aa) | Length | Vκ | Jκ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA3-post 50 | 4-34 | 3-22 | 2 | 2 | DYYDSSGLLRAYSWYFDL | 18 | 1-33 | 4 | QQYDNLPLT | 9 | - | - | - | - |
| mADA3-post 53 | 3-30 | 3-22 | 2 | 4 | DELRYYDSSGLNYFDY | 16 | 1-39 | 3 | QQSYSTLFT | 9 | + | - | - | - |
| mADA3-post 61 | 6-1 | 6-19 | 1 | 4 | EPKPKSQWLVHGAIDY | 16 | 1-39 | 1 | QQSYSTPWT | 9 | - | + | + | + (N) |
| mADA3-post 63 | 3-30 | 1-7 | 2 | 3 | EFRNYEGAFDI | 11 | 4-1 | 4 | QQYYSSSSST | 10 | - | - | - | - |
| mADA3-post 64 | 6-1 | 6-6 | 2 | 5 | SSHSPNWFDP | 10 | 1-39 | 2 | QQSYSTPPT | 9 | - | - | - | - |
| mADA3-post 65 | 3-11 | 1-26 | 2 | 4 | DRGGSYLRGGFDY | 13 | 1-33 | 3 | QQYDNLLFT | 9 | - | - | - | - |
| mADA3-post 71 | 4-39 | 3-22 | 2 | 4 | PPGDYDSSGAHY | 12 | 3-11 | 4 | QQRSNWPPLT | 10 | - | - | - | - |
| mADA3-post 75 | 1-2 | 7-27 | 3 | 4 | LITGAQLDY | 9 | 1-6 | 2 | LQDYNYPPT | 9 | + | - | - | - |
| mADA3-post 84 | 3-33 | 3-22 | 2 | 4 | GPYYYDSSGYYYGYYFDY | 18 | 3-11 | 2 | QQRSNWPPYT | 10 | + | - | - | - |
| mADA3-post 85 | 4-61 | 3-10 | 2 | 4 | GPAPYYYGSGSYYERFDY | 18 | 4-1 | 4 | QQYYSTPLT | 9 | + | + | + | + (C) |
| mADA3-post 86 | 4-b | 3-22 | 2 | 3 | EGVAHYDSSGYYYDAFDI | 18 | 3-20 | 1 | QQYGSSRT | 8 | + | - | - | - |
| mADA3-post 87 | 4-34 | 3-3 | 2 | 3 | GIYDFWSGYRGAFDI | 15 | 1-5 | 1 | QQYNSYSRT | 9 | + | + | + | + (C) |
| mADA3-post 88 | 3-48 | 6-19 | 2 | 4 | ALSSSGVYFDY | 11 | 1D-8 | 1 | QQYYSFPWT | 9 | + | - | - | - |
| mADA3-post 89 | 3-21 | 6-19 | 3 | 4 | DRPSLAVAGRGCDY | 14 | 3-11 | 4 | QQRSNWPLT | 9 | + | - | - | - |
| mADA3-post 90 | 4-34 | 2-21 | 3 | 6 | RMVVTATPYYYYGMDV | 16 | 3-20 | 1 | QLGSGT | 6 | + | - | - | - |
| mADA3-post 91 | 5-51 | 3-3 | 1 | 2 | LSFLEWLDQRYFDL | 14 | 3-11 | 2 | QQRSNWLMYT | 10 | + | - | - | - |
| mADA3-post 96 | 3-23 | 3-3 | 2 | 4 | DKDFWGYLRPLFDY | 14 | 4-1 | 4 | QQYYSTPPLT | 10 | + | + | + | + (C) |
| mADA3-post 52 | | | | | | | 3-11 | 1 | QQRSNWPWT | 9 | + | | | |
| mADA3-post 62 | | | | | | | 1-5 | 4 | QQYNSYSLT | 9 | + | | | |
| mADA3-post 69 | | | | | | | 1-9 | 4 | QQLNSYPLT | 9 | + | | | |
| mADA3-post 77 | | | | | | | 3-20 | 4 | QQYGSSLT | 8 | + | | | |
| mADA3-post 80 | | | | | | | 3-20 | 2 | QQYGSSPMYT | 10 | + | | | |
| mADA3-post 82 | | | | | | | 2-28 | 1 | MQALQTPWT | 9 | + | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | Vλ | Jλ | CDR3 (aa) | Length | NeoR | Poly | Hep2 | Staining |
| mADA3-post 49# | 4-34 | 5-5 | 1 | 5 | TEIQLWYNWLDP | 12 | 4-60 | 2 | ETWDSNTRV | 9 | + | | | |
| mADA3-post 51 | 1-2 | 6-13 | 3 | 4 | IHLKISAAGSKSFYFDY | 17 | 3-21 | 2 | QVWDSSSDHHVV | 12 | - | - | + | + (C+N) |
| mADA3-post 70 | 1-69 | 3-22 | 2 | 4 | DGYYDSSGYYFDY | 13 | 2-23 | 2 | CSYAGSVV | 8 | + | + | + | + (C) |
| mADA3-post 73 | 1-8 | 2-2 | 2 | 6 | TFPAFLGYCSSTSCYGHGYYYYGMDV | 26 | 1-44 | 2 | AAWDDSLNGVV | 11 | + | - | - | - |
| mADA3-post 74 | 4-34 | 7-27 | 1 | 3 | PGLGRRSAFDI | 11 | 2-14 | 1 | SSYTSSSTRYV | 11 | + | - | - | - |
| mADA3-post 76 | 3-11 | 3-10 | 1 | 4 | YPTLLWFGEDKPSHFDY | 17 | 2-11 | 2 | CSYAGSYTVV | 10 | + | - | - | - |
| mADA3-post 83 | 1-69 | 3-3 | 3 | 6 | APVPITIFGVVTSTGYYYGMDV | 22 | 2-14 | 2 | SSYTSSSPVV | 10 | + | + | + | + (C) |
| mADA3-post 94 | 5-51 | 2-2 | 2 | 4 | IRTRDCSSTSCYGGYFDY | 18 | 2-14 | 1 | SSYTSSSTLYV | 11 | + | - | - | - |
| mADA3-post 77 | | | | | | | 3-1 | 3 | QAWDSSTAWV | 10 | + | | | |
| mADA3-post 93 | | | | | | | 2-14 | 2 | SSYTSSSTLV | 10 | + | | | |
| | VH | D | RF | JH | CDR3 (aa) | Length | | | | | NeoR | | | |
| mADA3-post 55 | 1-8 | 2-15 | 3 | 4 | GPSHPNVVVVAATEISDY | 18 | | | | | + | | | |
| mADA3-post 67 | 4-34 | 3-22 | 3 | 4 | GREIVVDMTLKPTDY | 15 | | | | | + | | | |
| mADA3-post 68 | 3-21 | 1-7 | 2 | 4 | EENNWNYGDY | 10 | | | | | - | | | |
| mADA3-post 72 | 4-34 | 1-26 | 3 | 4 | AVGATHD | 7 | | | | | + | | | |
| mADA3-post 79 | 3-23 | / | / | 4 | ANRRYFDY | 8 | | | | | + | | | |
| mADA3-post 95 | 1-46 | 4-4 | 3 | 6 | VVETTFSYYYYGMDV | 15 | | | | | + | | | |

RF, reading frame; #, antibody failed to be expressed;-, non-reactive; +, reactive;

C, diffuse cytoplasmic staining; S, mitotic spindle staining; N, nuclear staining; F, cytoplasmic fibers

**Supplemental Table 17:** Serum autoantibodies in ADA-SCID patients 6yrs postGT

| | ADA1 | | ADA2 | | ADA3 | |
|---|---|---|---|---|---|---|
| GT | preGT | postGT | preGT | postGT | preGT | postGT |
| Direct or indirect Coombs tests | - | - | N.D. | - | - | - |
| anti-neutrophil cytoplasmic antibodies (ANCA) | N.D. | - | N.D. | - | - | - |
| anti-liver kidney microsomal antibodies (LKM) | N.D. | - | N.D. | - | - | - |
| anti-smooth muscle antibodies | N.D. | - | N.D. | - | - | - |

N.D., not done; -, non-reactive;