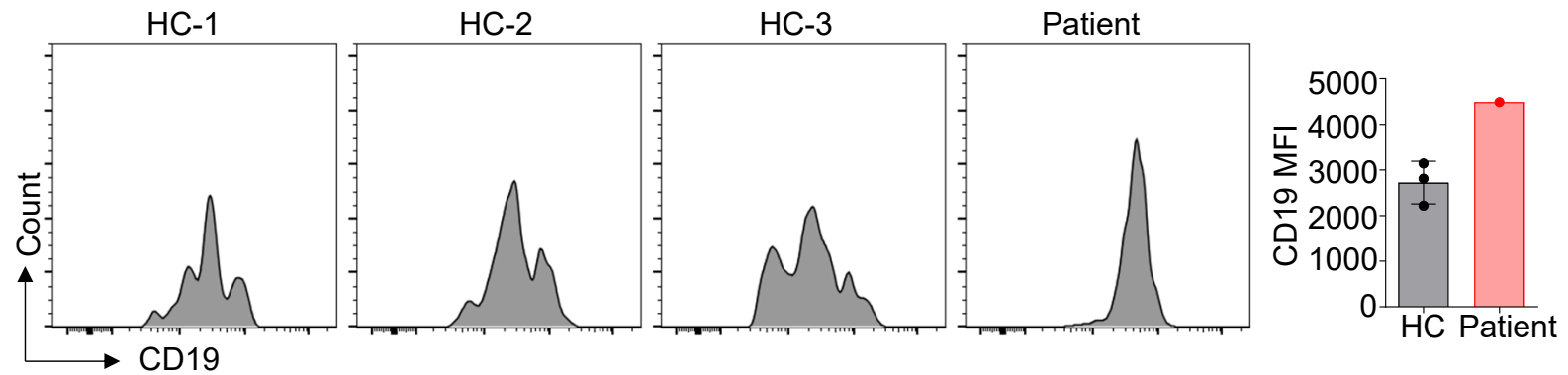


Exon 1

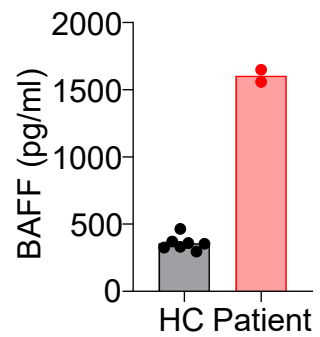
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Exon 2

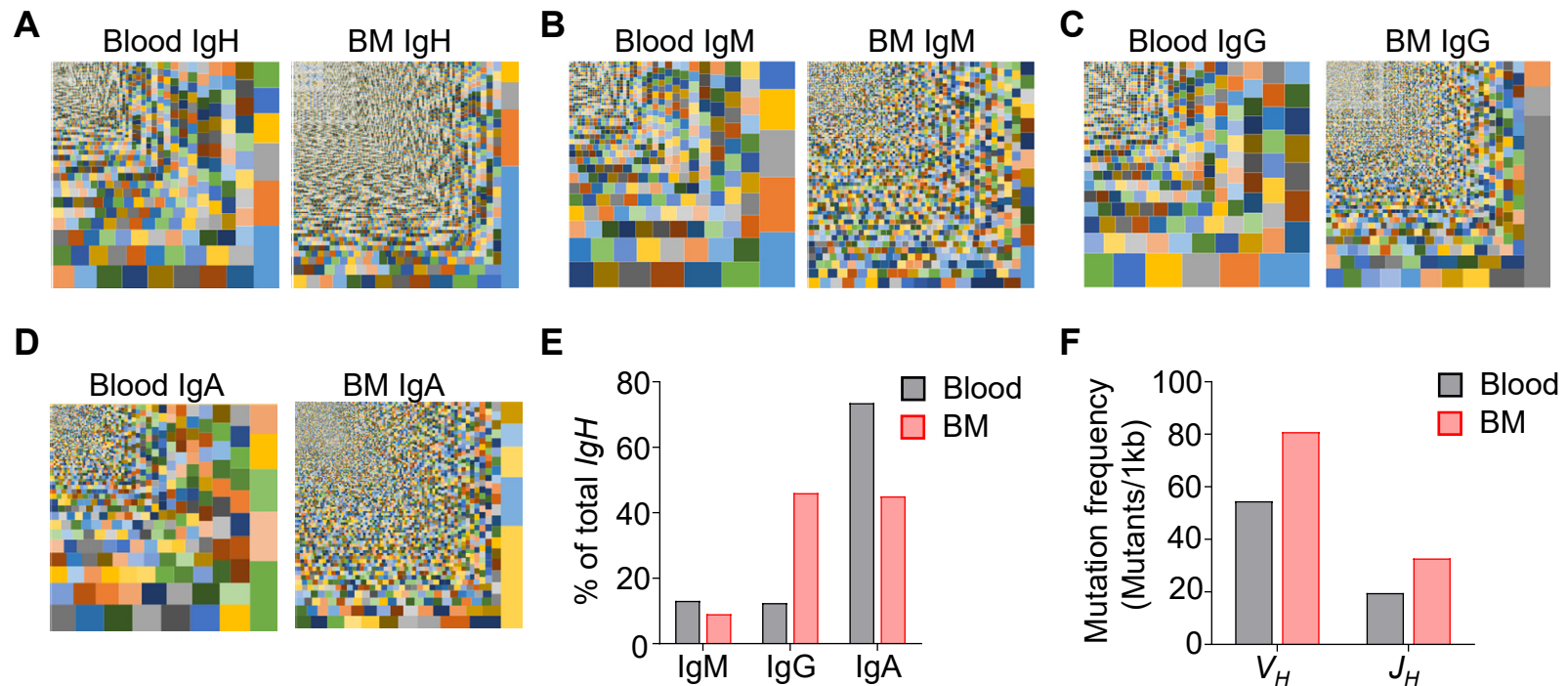
Supplemental Figure 1 Product of the aberrantly spliced *RAG1* mRNA in the patient. The sequence includes the entire exon 1 (highlighted in grey), retained intron 1 (black) and part of exon 2 (highlighted in grey). The mutated nucleotide is shown in red. Eleven out-of-frame ATGs in intron 1 are underlined in red and an ATG in intron 1 that is in-frame with the authentic ATG (highlighted in blue) is underlined in black. Eleven premature stop codons are squared.



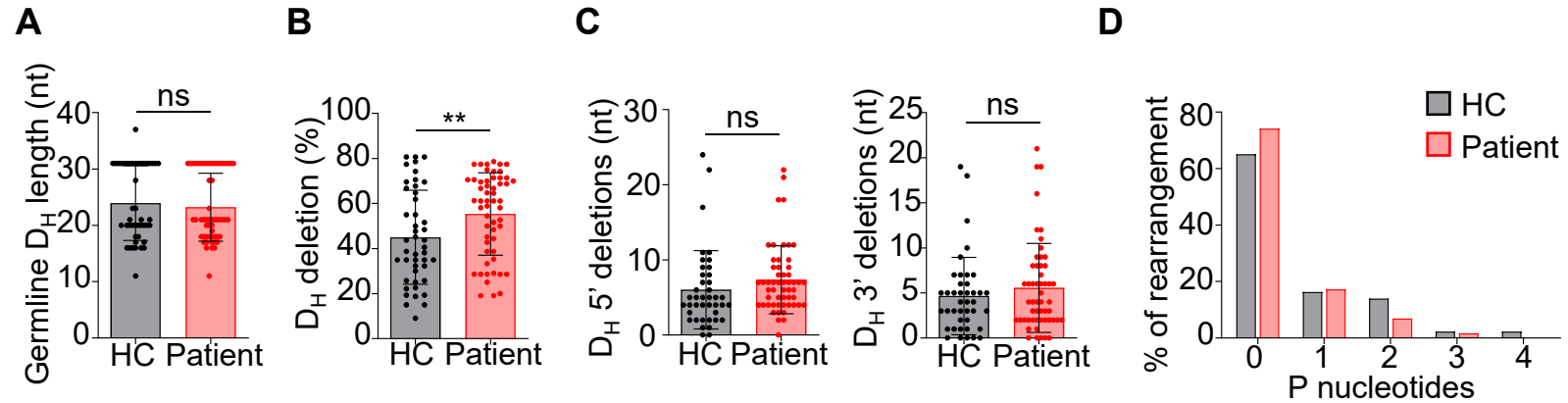
Supplemental Figure 2 The patient IgD⁻CD27⁻ DN B cells expressed higher levels of CD19 than did those from HC. The DN B cells from HC and the patient (Fig. 3B, lower panels) were analyzed for their CD19 expression.



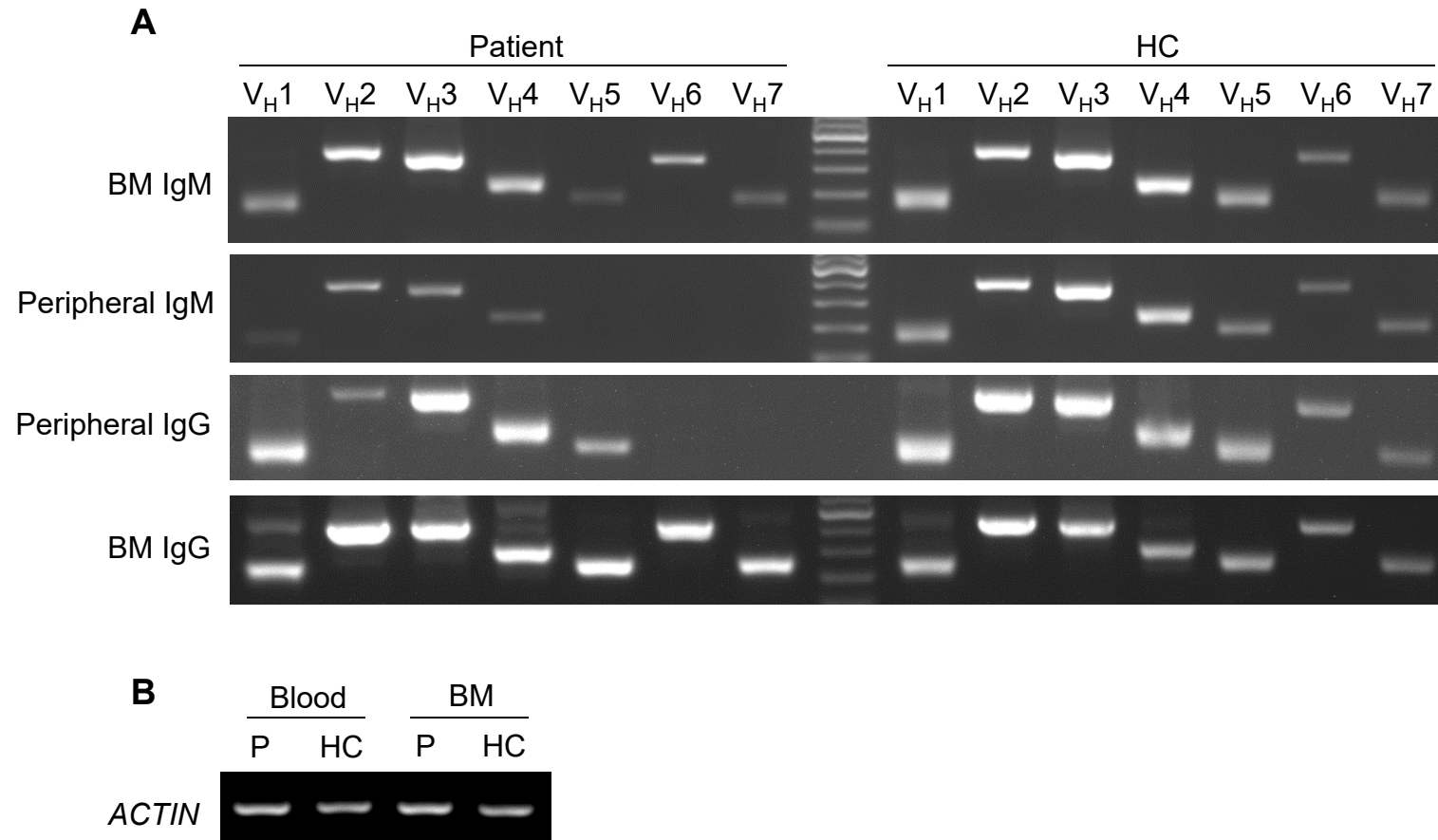
Supplemental Figure 3 The patient serum contained an elevated level of BAFB than the sera from 7 HC. The concentration of BAFB was analyzed using ELISA. The patient sample was independently analyzed twice.



Supplemental Figure 4 (A-D) Tree maps showing CDR3 amino acid sequences from the patient peripheral blood and BM. (A) Total IgH, (B) IgM, (C) IgG and (D) IgA. Each rectangle represents a unique CDR3 amino acid sequence and the size of the rectangle represents the relative frequency of an individual CDR3. (E) Frequency of IgM, IgG and IgA sequences among all CDR3s in the patient peripheral blood and BM. (F) Mutation frequency in the V_H and J_H genes of the patient peripheral blood and BM.



Supplemental Figure 5 *IgH* CDR3 profiles in the BM. **(A)** Average length of the germline D_H segments used by the patient and HC. **(B)** Proportion of the deleted nucleotides (nt) from germline D_H segment. **(C)** Number of nt deleted from the 5' (left) and 3' (right) ends of the D_H segments in the patient and HC. **(D)** The distribution of P-nucleotide addition. Mean \pm SD is shown. ** $p < 0.01$; ns, not significant.



Supplemental Figure 6 Amplification of V_H genes using cDNA derived from the BM and peripheral blood of the patient and HC. (A) V_H -specific forward primers and a C μ (for IgM) or a C γ (for IgG) reverse primer were used for amplification. (B) As a control, the *ACTIN* gene was amplified from the same cDNA.

Supplemental Table 1. Serum Ig levels in the patient at age 7

IgG subclass	Value (g/L)	Reference (g/L)
IgG ₁	20.5	3.77-11.31
IgG ₂	1.02	0.68-3.88
IgG ₃	0.945	0.158-0.890
IgG ₄	0.0687	0.012-1.699

Supplemental Table 2. Clinical and immunological features of the PID cohort

	Sex	Age	Genetic defects	Clinical features	B cell (cells/ul)	Reference values	T cell (cells/ul)	Reference values	Naive B (% of CD19 ⁺)	Naive B (cells/ul)	DN B (% of CD19 ⁺)	DN B (cells/ul)	Memory B (% of CD19 ⁺)	Memory B (cells/ul)	IgG (g/L)	Reference values	IgM (g/L)	Reference values	IgA (g/L)	Reference values	Auto-antibody
PID001*	M	8Y	RAG1	Fever; infections; granulomas	4	300-600	14	1100-2200	17.3	0.60896	59.5	2.38	22.1	0.884	32.1	6.09-12.85	2.73	0.67-2.01	2.06	0.52-2.16	ANA, pANCA, Coombs
PID002	M	9M	RAG1	Recurrent infections	801	300-800	1958	1800-3300	70.9	567.909	7.1	56.871	11.8	94.518	14.66	3.50-8.90	3.38	0.36-1.20	1.12	0.06-0.54	Coombs
PID003	M	11Y	PIK3R1	CVID	32	300-600	796	1100-2200	42.3	13.71789	54.8	17.536	2.54	0.8128	0.54	6.09-12.85	0.1	0.67-2.01	0.14	0.52-2.16	SSA
PID004*	F	1Y5M	Unknown	Recurrent infections	397	300-600	1984	1900-2900	12.1	48.037	13.9	55.183	71	281.87	38.5	5.52-11.46	2.98	0.60-2.12	1.65	0.06-0.74	ANA
PID005*	F	4Y4M	Unknown	Eosinophilia	739	300-600	1812	1900-2900	7.97	58.8983	77.4	571.986	14.3	105.677	24.35	4.95-12.74	1.08	0.65-2.01	1.65	0.33-1.89	SSB
PID006	M	4Y9M	PIK3CD	Recurrent fever and diarrhea	211	300-600	1303	1500-2900	76.8	162.048	8.01	16.9011	13.6	28.696	19 [#]	4.95-12.74	3.85	0.65-2.01	1.54	0.33-1.89	PR3, GBM
PID007	M	13Y	Unknown	Hypogammaglobulinemia	303	300-600	1282	1100-2200	84.7	256.641	6.12	18.5436	5.32	16.1196	5 [#]	6.98-14.26	1.54	0.56-2.16	2.09	0.92-2.50	(-)
PID008	F	13Y	TNFRF13B	Thrombocytopenic	301	300-500	680	1300-2200	74.6	224.546	6.86	20.6486	11.4	34.314	10.79 [#]	6.98-14.26	0.54	0.56-2.16	1.32	0.92-2.50	(-)
PID009	M	16Y	Unknown	CVID	203	300-600	759	1100-2200	93.9	190.617	1.78	3.6134	1.06	2.1518	ND	7.54-16.62	0.02	0.72-2.28	0.05	0.89-3.24	(-)
PID010	M	5Y6M	Unknown	Recurrent fever; encephalopathy	494	300-600	2112	1500-2900	90.6	447.564	3.95	19.513	3.21	15.8574	9.68 [#]	4.95-12.74	0.82	0.65-2.01	1.65	0.33-1.89	(-)
PID011	M	15Y	PIK3CD	Hyper IgM syndrome	169	300-600	909	1100-2200	66.5	112.385	17.5	29.575	5.69	9.6161	5.26 [#]	7.54-16.62	18.3	0.72-2.28	0.99	0.89-3.24	(-)
PID012	F	9M	Unknown	Thrombocytopenic; agranulocytosis	1307	300-800	3371	2800-3500	95.1	1242.957	0.92	12.0244	1.44	18.8208	19.37	3.50-8.90	0.46	0.36-1.20	0.11	0.06-0.54	(-)
PID013	F	4Y8M	PIK3CD	Hypogammaglobulinemia; lymphadenopathy	36	300-600	746	1900-2900	42	15.12	48.3	17.388	7.91	2.8476	3.04 [#]	4.95-12.74	11.06	0.65-2.01	0.24	0.33-1.89	(-)
PID014	M	4Y4M	PIK3CD	APDS	278	300-600	1138	1500-2900	80.5	223.79	10.6	29.468	6.66	18.5148	9.15 [#]	4.95-12.74	3.82	0.65-2.01	0.55	0.33-1.89	(-)
PID015	M	2Y	Unknown	Recurrent fever and diarrhea	310	300-600	2696	1500-2900	80.2	248.62	7.23	22.413	11.6	35.96	19.9 [#]	5.52-11.46	1.25	0.60-2.12	2.97	0.06-0.74	(-)
PID016	M	5Y4M	Unknown	Recurrent fever	285	300-600	901	1500-2900	77.7	221.445	8.46	24.111	10	28.5	9.68	4.95-12.74	0.96	0.65-2.01	2.31	0.33-1.89	(-)
PID017	M	3M	WAS	WAS	301	300-800	2143	1800-3300	91.3	274.813	2.95	8.8795	2.43	7.3143	21.58	3.70-8.30	0.31	0.33-1.25	0.39	0.14-0.50	(-)
PID018	M	11Y	Unknown	Recurrent fever; Chronic EBV infection	168	300-600	845	1100-2200	37.1	62.328	59.6	100.128	2.77	4.6536	13.28 [#]	6.09-12.85	0.36	0.67-2.01	2.2	0.52-2.16	(-)
PID019	M	2Y	Unknown	agammaglobulinemia	12	300-600	880	1500-2900	19.3	2.35074	80	9.6	0.57	0.0684	8.05	5.52-11.46	0.12	0.60-2.12	0.29	0.06-0.74	(-)
PID020	F	7Y	PIK3CD	Recurrent pulmonary infections; EBV infection	152	300-500	1052	1300-2200	63.5	96.52	30.5	46.36	5.01	7.6152	11.62 [#]	6.09-12.85	3.81	0.67-2.01	1.32	0.52-2.16	(-)
PID021	F	3Y3M	Unknown	Recurrent pulmonary infections	935	300-600	2781	1900-2900	91.4	854.59	3.01	28.1435	2.84	26.554	6.92	4.95-12.74	11.53	0.65-2.01	0.44	0.33-1.89	(-)
PID022	M	10Y	Unknown	CVID	170	300-600	618	1100-2200	94.9	161.33	0.85	1.445	0.51	0.867	0.09	6.09-12.85	0.2	0.67-2.01	0.51	0.52-2.16	(-)
PID023	F	17Y	PIK3CD	Hyper IgM syndrome	52	300-500	820	1300-2200	17.1	8.892	17.1	8.892	14.5	7.54	5.81 [#]	7.54-16.62	36.86	0.72-2.28	6.64	0.89-3.24	(-)
PID024	M	5Y7M	Unknown	agammaglobulinemia	176	300-600	1758	1500-2900	90	158.4	1.37	2.4112	2.62	4.6112	ND	4.95-12.74	0.04	0.65-2.01	0.08	0.33-1.89	(-)
PID025	M	1Y11M	Unknown	Recurrent fever	750	300-600	2572	1500-2900	94.3	707.25	1.56	11.7	1.42	10.65	8.15 [#]	5.52-11.46	0.33	0.60-2.12	0.3	0.06-0.74	(-)
PID026	F	1Y5M	ADA2	Autoinflammatory; fever; skin rash	1192	300-600	1482	1500-2900	88.9	1059.688	2.63	31.3496	4.69	55.9048	9.96 [#]	5.52-11.46	1.99	0.60-2.12	1.21	0.06-0.74	(-)
PID027	F	5M	Unknown	Agranulocytosis; inflammatory	725	300-800	2485	2800-3500	8.15	59.0875	0.47	3.4075	1.39	10.0775	7.47	3.70-8.30	0.9	0.33-1.25	0.36	0.14-0.50	(-)
PID028	M	5Y	Unknown	Eosinophilia	422	300-600	2478	1500-2900	84.1	354.902	4.91	20.7202	6.23	26.2906	14.39	4.95-12.74	1.14	0.65-2.01	2.09	0.33-1.89	(-)
PID029*	M	2M	Unknown	SCID	2.17	300-800	6.7	1800-3300	1.6	0.03472	44.8	0.97216	19.2	0.41664	4.15	3.70-8.30	0.05	0.33-1.25	0.03	0.14-0.50	(-)
PID030	M	5M	Unknown	Recurrent fever	1105	300-800	1346	1800-3300	97	1071.85	0.83	9.1715	0.69	7.6245	15.77	3.70-8.30	1.26	0.33-1.25	0.08	0.14-0.50	(-)
PID031	M	9M	Unknown	Recurrent severe infections	452	300-800	1956	1800-3300	93.7	423.524	2.45	11.074	1.68	7.5936	8.58	4.95-12.74	1.51	0.65-2.01	0.46	0.33-1.89	UD
PID032	M	1Y	BTK	XLA	1.9	300-600	4257	1500-2900	62.5	1.1875	28.1	0.5339	6.25	0.11875	ND	5.52-11.46	0.08	0.60-2.12	0.02	0.06-0.74	(-)
PID033	M	1Y	BTK	XLA	7	300-600	4965	1500-2900	93.6	6.552	5.32	0.3724	1.06	0.0742	9.41 [#]	5.52-11.46	0.08	0.60-2.12	0.25	0.06-0.74	UD
PID034	M	2Y10M	Unknown	Recurrent infections	837	300-600	2268	1500-2900	90.4	756.648	6.25	52.3125	2.25	18.8325	27.1 [#]	5.52-11.46	0.67	0.60-2.12	2.31	0.06-0.74	(-)
PID035*	F	5Y10M	Unknown	Recurrent infections	258	300-600	2688	1900-2900	59.5	153.51	12.3	31.734	18.3	47.214	14.39	4.95-12.74	1.34	0.65-2.01	2.9	0.33-1.89	UD
PID036	F	3Y7M	Unknown	Recurrent infections	379	300-600	1889	1900-2900	81.6	309.264	6.66	25.2414	6.79	25.7341	9.4	4.95-12.74	0.98	0.65-2.01	0.77	0.33-1.89	UD
PID037*	F	3M	RAG1	SCID	0.79	300-800	0.4	2800-3500	36.4	0.28756	36.4	0.28756	27.3	0.21567	7.75	3.70-8.30	0.06	0.33-1.25	0.04	0.14-0.50	UD
PID038	F	1M	IKBKG	Skin rash; Recurrent infections	1507	300-800	3603	2800-3500	94.2	1419.594	1.78	26.8246	1.32	19.8924	8.2	3.70-8.30	1.25	0.33-1.25	0.33	0.14-0.50	UD
PID039	M	6Y	SH2D1A	XLP	920	300-600	3984	1500-2900	96.1	884.12	0.59	5.428	0.3	2.76	5.4 [#]	4.95-12.74	0.12	0.65-2.01	0.18	0.33-1.89	UD
PID040	M	1Y3M	BTK	XLA	8.32	300-800	1836	1500-2900	89.6	7.45472	6.25	0.52	0	0	5.4 [#]	5.52-11.46	0.06	0.60-2.12	0.12	0.06-0.74	UD
PID041	M	2M	Unknown	SCID	1082	300-600	11.3	1800-3300	97.7	1057.114	0.32	3.4624	0.034	0.36788	11.7 [#]	3.70-8.30	0.18	0.33-1.25	0.04	0.14-0.50	UD
PID042	M	2Y11M	Unknown	Recurrent infections	756	300-600	2782	1500-2900	80	604.8	4.45	33.642	8.47	64.0332	7.47	5.52-11.46	1.15	0.60-2.12	0.47	0.06-0.74	UD
PID043	F	5M	RAG1	SCID	42	300-800	727	2800-3500	12.6	5.292	30.5	12.81	54.6	22.932	14.39 [#]	3.70-8.30	1.42	0.33-1.25	0.27	0.14-0.50	UD
PID044	M	8Y	Unknown	Agammaglobulinemia; agranulocytosis	9.63	300-600	475.8	1100-2200	90.6	8.72478	3.12	0.300456	3.12	0.300456	4.9 [#]	6.09-12.85	0.19	0.67-2.01	0.33	0.52-2.16	UD
PID045	M	5Y2M	CD40LG	Hyper IgM syndrome	473	300-600	2918	1500-2900	89.4	422.862	1.03	4.8719	1.56	7.3788	2.9	4.95-12.74	2.62	0.65-2.01	1.98	0.33-1.89	UD
PID046	F	5M	Unknown	Recurrent infections	43	300-800	407	2800-3500	90.9	39.087	7.27	3.1261	0	0	4.7	3.70-8.30	2.6	0.33-1.25	0.8	0.14-0.50	UD
PID047	M	5Y	Unknown	Fever; encephalopathy	435	300-600	426	1500-2900	86.4	375.84	2.94	12.789	5.67	24.6645	ND	4.95-12.74	0.04	0.65-2.01	0.18	0.33-1.89	UD
PID048	M	14Y	BTK	Hypogammaglobulinemia	1.38	300-500	1676.7	1100-2200	10	0.138	50	0.69	35	0.483	7.5 [#]	6.98-14.26	0.03	0.56-2.16	0.55	0.92-2.50	UD
PID049	M	1Y11M	MHC II	Recurrent fever	572	300-600	4119	1500-2900	90.2	515.944	2.88	16.4736	3.88								