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Supplemental Data

A Common Molecular Mechanism Underlies Two Phenotypically Distinct 17p13.1 Microdeletion Syndromes

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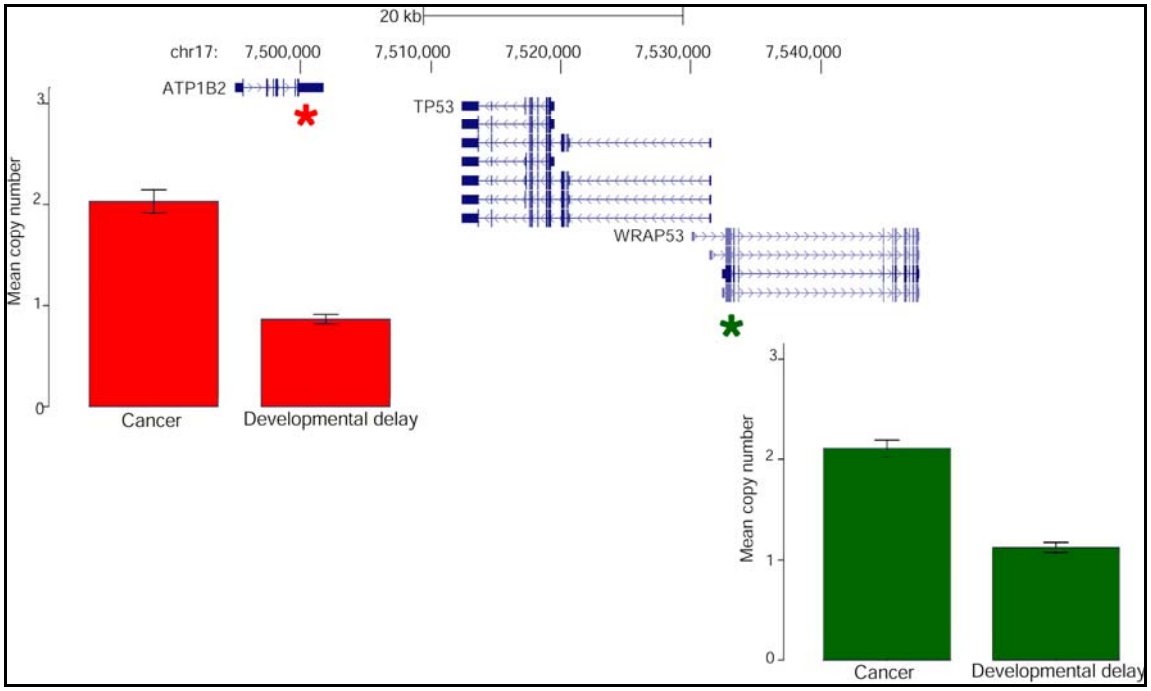
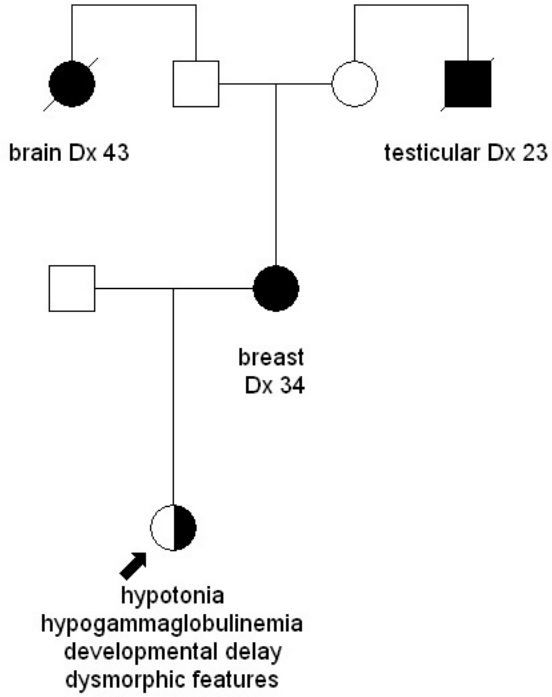
Figure S1. Copy Number of *ATP1B2* and *WRAP53, TP53*'s Neighboring Genes



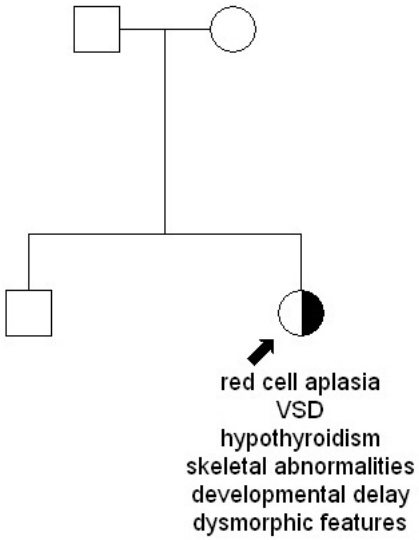
To determine the extent of 17p13.1 CNVs, probes were designed in *TP53*'s neighboring genes, *ATP1B2* and *WRAP53* and assessed by qPCR. Probes were located in the closest exon to *TP53* (red and green asterisks). While all patients' harbored deletions at *TP53*, only the developmental delay patients' deletions included *ATP1B2* (telomeric) and *WRAP53* (centromeric). Both *ATP1B2* and *WRAP53* were diploid in all cancer patients (mean copy number = 2.03 and 2.11, respectively). However, all patients with DD were hemizygously deleted for both flanking genes (mean copy number = 0.87 [*ATP1B2*] and 1.13 [*WRAP53*]), a significant reduction as compared to the cancer patients ($p=2.90 \times 10^{-4}$ [*ATP1B2*] and 2.42×10^{-8} [*WRAP53*]).

Figure S2. Pedigrees

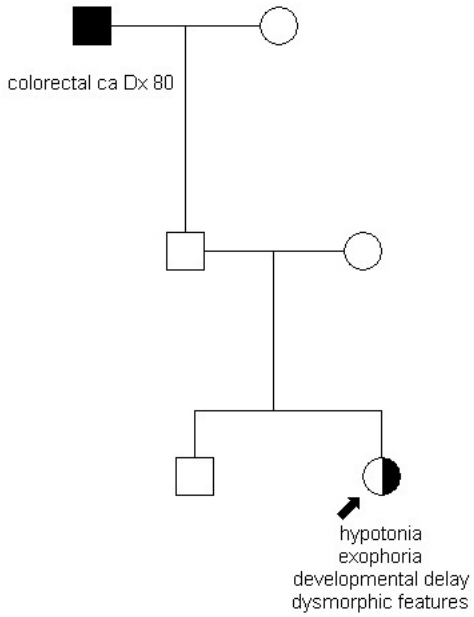
Patient 2723 - DD



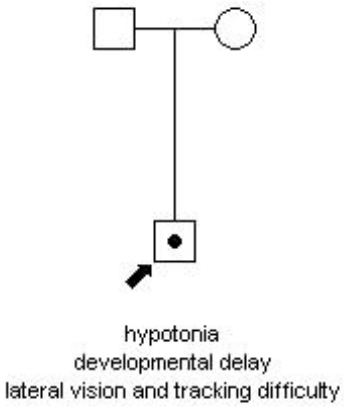
Patient 3026 - DD



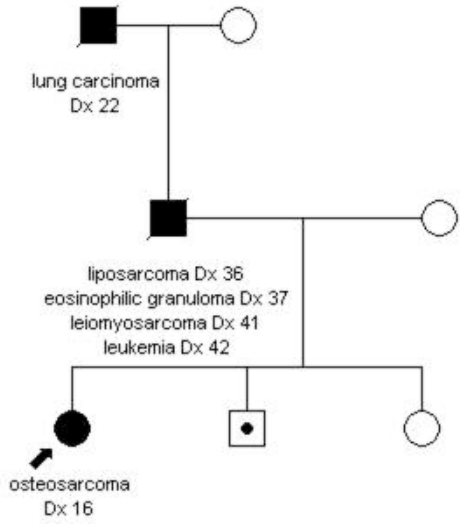
Patient 3148 - DD



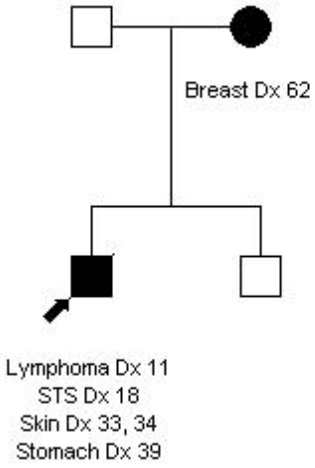
Patient 3354 – DD



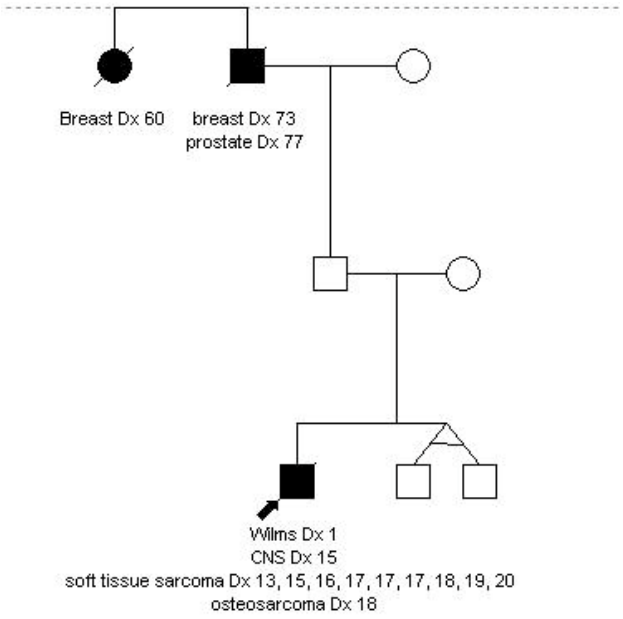
Patient 3332 - Cancer



Patient Y47- Cancer



Patient Y20 - Cancer



Patient 2760 - Cancer

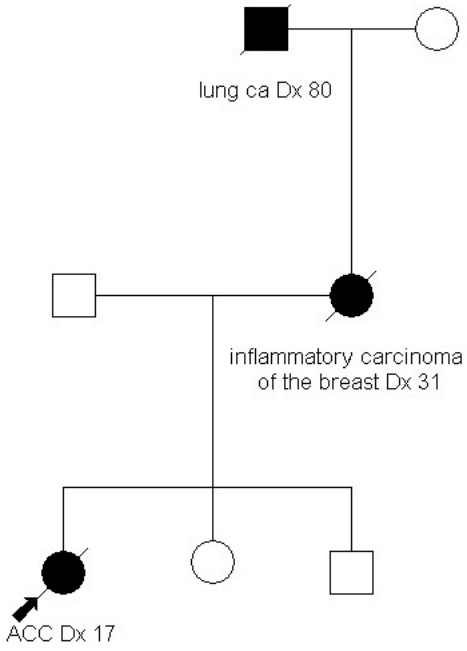
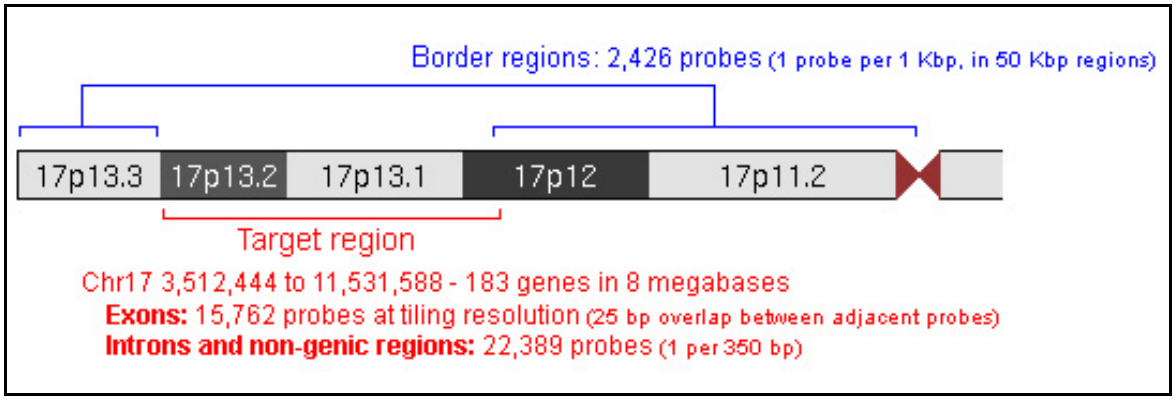
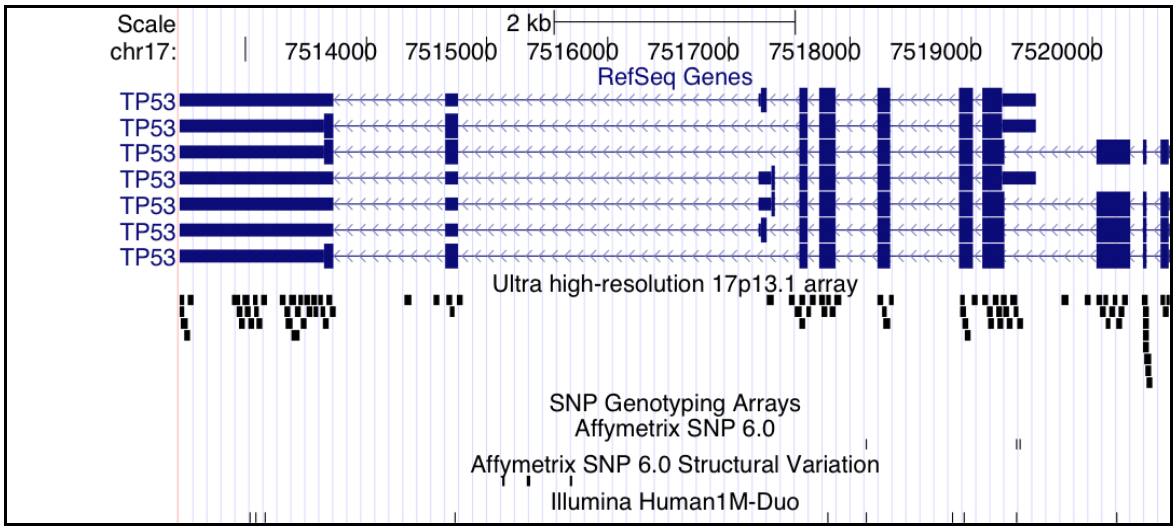


Figure S3. Design of Ultra High-Resolution Array

A



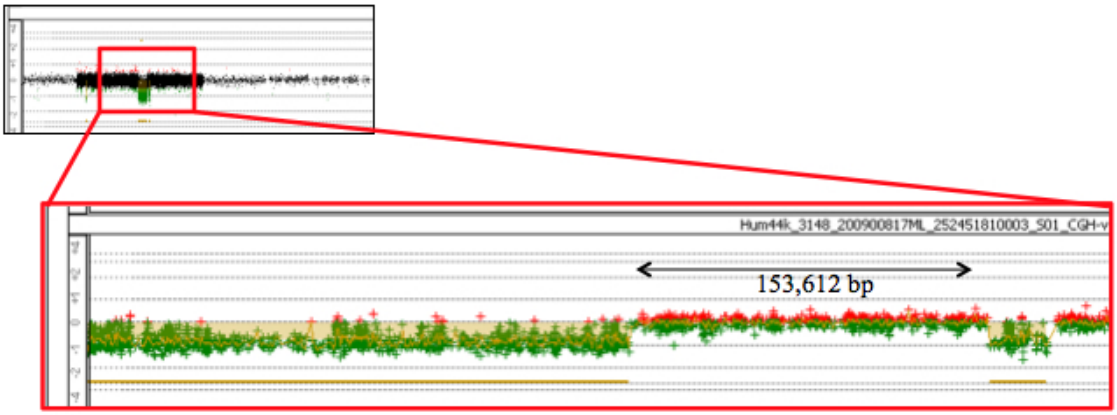
B



(a) 40,577 oligonucleotide probes were placed on the short arm of chromosome 17, in which an 8 Mb target region around *TP53* (red) was covered in ultra high-resolution. Of the 38,061 in the target region, 15,762 (41%) were designed on exons. Exonic probes were overlapping and tiled across all exons, of all alternative splice variants, for every gene (183) in the target region. The remaining probes in the target region (22,389) were placed in intronic or non-genic regions at a resolution of one probe per 350 nucleotides. An additional 2,426 probes were placed in the border regions (blue) from the target region to the telomere and centromere. The borders regions were covered at low resolution with 1 probe per 1 Kbp, in 50 Kbp regions. The following 183 genes are covered at tiling resolution by this novel platform: **Telomere**-…-TAX1BP3-TMEM93-P2RX5-ITGAE-GSG2-C17orf85-CAMKK1-P2RX1-ATP2A3-ZZEF1-CYB5D2-ANKFY1-UBE2G1-SPNS3-SPNS2-MYBBP1A-GGT6-SMTNL2-ALOX15-PELP1-ARRB2-MED11-CXCL16-ZMYND15-TM4SF5-VMO1-GLTPD2-PSMB6-PLD2-MINK1-CHRNE-LOC100130311-GP1BA-SLC25A11-RNF167-PFN1-ENO3-SPAG7-CAMTA2-INCA1-KIF1C-GPR172B-ZFP3-ZNF232-USP6-ZNF594-C17orf87-RABEP1-NUP88-RPAIN-C1QBP-DHX33-DERL2-MIS12-NLRP1-WSCD1-AIPL1-FAM64A-PITPNM3-KIAA0753-TXNDC17-MED31-C17orf100-SLC13A5-XAF1-FBXO39-TEKT1-ALOX12P2-ALOX12-RNASEK-C17orf49-BCL6B-SLC16A13-SLC16A11-CLEC10A-ASGR2-ASGR1-DLG4-ACADVL-DVL2-PHF23-

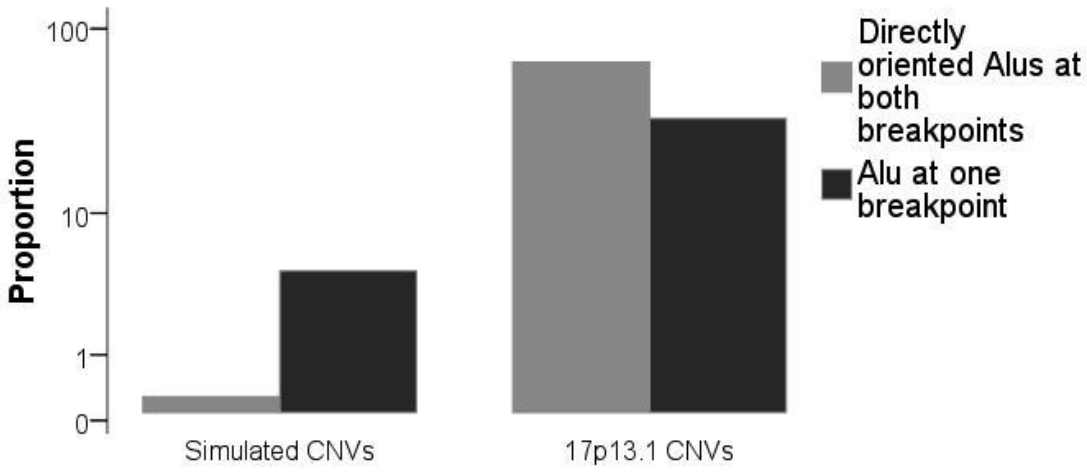
GABARAP-DULLARD-C17orf81-CLDN7-SLC2A4-YBX2-EIF5A-GPS2-NEURL4-ACAP1-KCTD11-TMEM95-TNK1-PLSCR3-C17orf61-NLGN2-SPEM1-C17orf74-TMEM102-FGF11-CHRNB1-ZBTB4-AMAC1L3-POLR2A-TNFSF12-TNFSF12/TNFSF13-TNFSF13-SENP3-EIF4A1-SNORA48-SNORD10-SNORA67-CD68-MPDU1-SOX15-FXR2-SHBG-SAT2-SHBG-ATP1B2-**TP53**-WRAP53-EFNB3-DNAH2-RPL29P2-KDM6B-TMEM88-LSMD1-CYB5D1-CHD3-SCARNA21-LOC284023-KCNAB3-TRAPPC1-CNTROB-GUCY2D-ALOX15B-ALOX12B-ALOXE3-HES7-PER1-VAMP2-TMEM107-C17orf59-AURKB-C17orf44-C17orf68-PFAS-SLC25A35-RANGRF-ARHGEF15-ODF4-LOC100128288-KRBA2-RPL26-RNF222-NDEL1-MYH10-CCDC42-SPDYE4-MFSD6L-PIK3R6-PIK3R5-NTN1-STX8-WDR16-USP43-DHRS7C-GLP2R-RCVRN-GAS7-MYH13-MYH8-MYH4-MYH1-MYH2-MYH3-SCO1-C17orf48-TMEM220-PIRT-FLJ45455-DNAH9-…-**Centromere. (b)** Shown are the positions of probes (black squares and rectangles) across the region of 17p13.1 containing *TP53*. All exons (solid blue boxes), introns (dashed) and alternative transcripts of *TP53* are covered. Our arrays' coverage is contrasted to that of the Affymetrix 6.0 and Illumina 1M Duo microarrays. All genes within the target region have identical coverage as *TP53*, which is here shown to demonstrate the resolution of the platform in genic regions.

Figure S4. A Complex Event Near 17p13.1 Deletion Breakpoint



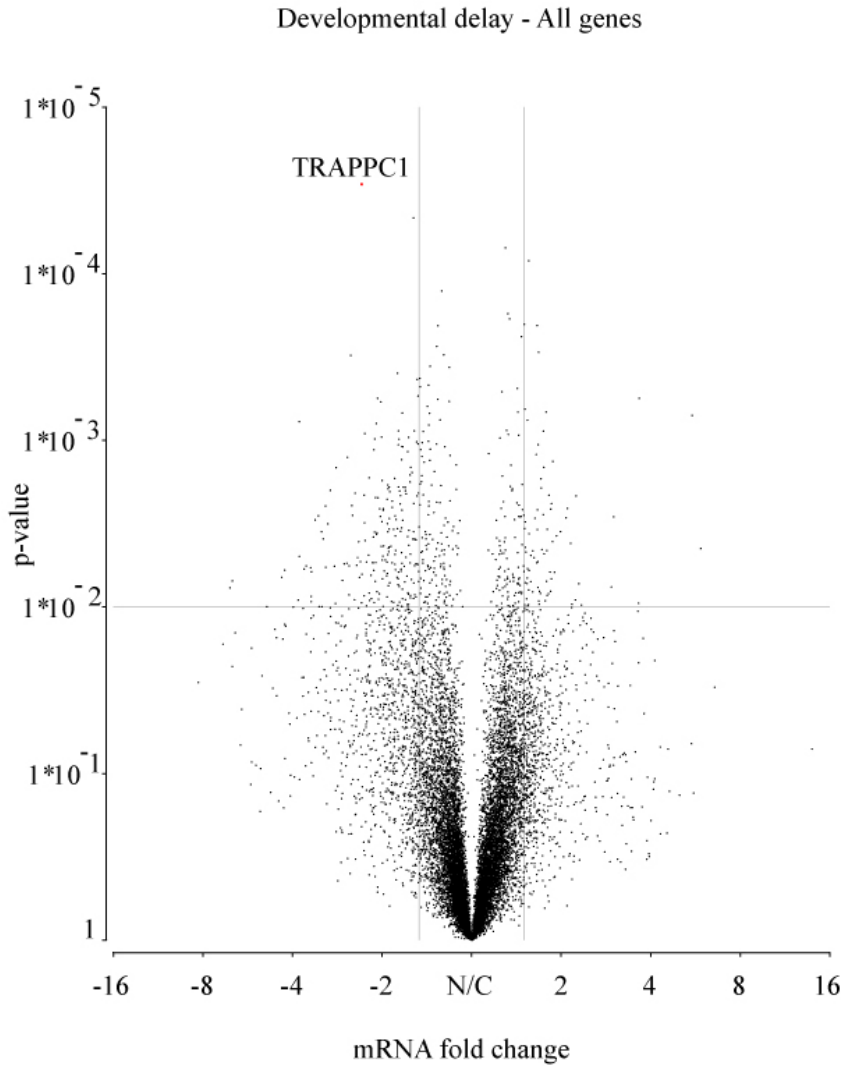
By ultra high-resolution array, an additional deletion was observed in one patient. This secondary deletion, which is distal to the primary deletion and is also hemizygous, is not a polymorphic CNV as it was not seen in other hybridizations using this platform, and is absent from the Database of Genomic Variants[36] and from the ultra high-resolution data released from the Genome Structural Variation Consortium[37]. The secondary deletions contains two genes: *EFNB3* and *DNAH2*.

Figure S5. Breakpoint Simulation



Shown are the proportion of CNVs whose breakpoints overlap with an Alu retrotransposon. We performed 10,000 simulation experiments in which randomly distributed CNVs were assessed for Alu overlap. Experiments were done on simulated CNVs sized 10 Kb, 100 Kb, 1 Mb and 2 Mb. Across all size ranges, few simulated CNVs were found to have directly oriented Alus at both breakpoints (~1%), or an Alu at only one breakpoint (<10%). In contrast, all 17p13.1 CNV breakpoints intersected with at least one Alu element: Most 17p13.1 CNV breakpoints were found to have directly oriented Alus at both breakpoints (67%), and the remainder had an intersecting Alu at only one breakpoint (33%).

Figure S6. Genome-wide Analysis of Gene Expression Differences in a Developmental Delay Patient



We used Affymetrix Exon arrays to look for gene expression differences in available blood-derived RNA. Gene expression values (x axis), expressed as a fold change relative to controls, are shown for all genes (circles). The p value of each genes' expression change is indicated (y axis, in reverse order). Grey gridlines delineate regions of the plot containing significantly under expressed genes (top left; $p<0.01$ and <-1.5 fold change) or significantly over expressed genes (top right; $p<0.01$ and >1.5 fold change). For a patient with a large 17p13.1 CNV and DD, *TRAPPC1* (red) was found to be the most significantly under expressed gene in the transcriptome.

Table S1. Initial Sample Ascertainments

	Hospital	**Patients screened**	**Patients with 17p13.1 microdeletion**	**Reason for screen**	**Method**
1	The Hospital for Sick Children	230	2	Suspected LFS, *TP53* sequencing wild type	MLPA
2	Stanford University School of Medicine	400	1	Many	Array CGH (44K array)
3	Emory University School of Medicine	3,374	1	Many	Array CGH (EmArray Cyto6000)
4	Children's Hospital of Philadelphia	487	1	Neurological or developmental issues	Illumina Hap550 BeadChip
5	Hospital A.C. Camargo	13	2	Suspected LFS, *TP53* sequencing wild type	MLPA
6	University Health Network, Toronto	20	1	Presence of multiple issues, including dysmorphologies, congenital delay or learning disabilities	Array CGH (Gene DX)
	Total:	**4,524**	**8**		

Table S2. Primer Probes Used for qPCR Assay

Primer name	Orientation	Sequence
chr17:7,067,522	TAAGAAAACAAATACCTGGAAGCAC	Forward
chr17:7,067,522	GATACTTGTTCTGGGTCAGTTCATC	Reverse
Primer 5'200k	AGGTACCCGCCCACCTCT	Forward
Primer 5'200k	GGGGTGGCTGCCTTTAAGT	Reverse
chr17:7,085,488	AAGTAGAACTGACCAACTGCAAAAG	Forward
chr17:7,085,488	CTCTGTGACTTTCTTGCATCTTGTA	Reverse
chr17:7,089,927	AATAACTTGCATTTCGACTTACAGC	Forward
chr17:7,089,927	GGGATTCCCTAGATTATCCCTAGTT	Reverse
5'200K#1	TGAGAAAACAGCCATAAAGATCA	Forward
5'200K#1	CCCGAGAGGATCACAAAGTT	Reverse
chr17:7,103,403	CGGTTTTATTAGGAGAGCAGTACAA	Forward
chr17:7,103,403	CACCATTTGGTACTGGGATAGATAC	Reverse
chr17:7,108,123	ATTCTCACATACCCTGCTTATTCTG	Forward
chr17:7,108,123	AGAGGAAAATGAAATGAGAGAGAGC	Reverse
5'200K#2	CAAGGCAAATGATATTCCAGGT	Forward
5'200K#2	GCCCAAGAATGAAGTCCTGA	Reverse
chr17:7,126,819	AATGCACGTGTTAATTTATGAAACC	Forward
chr17:7,126,819	TGTTAACAATATCCTTGACAGTGCT	Reverse
chr17:7,128,845	CTGCCATCACTTCTTCTTCTCC	Forward
chr17:7,128,845	GCTGGTCGAATAATAGAAAACCTAGAG	Reverse
chr17:7,129,661	GGTTAGGTTTCACTTCTCTGAGTTGAG	Forward
chr17:7,129,661	GAGAATCCAGTTGAGGCCTTTC	Reverse
chr17:7,130,108	AGGGAGCTGACCTAGATTGGATAG	Forward
chr17:7,130,108	CTTGGAGTAAGGTAAGTTATGCCACTG	Reverse
chr17:7,130,443	AACACCTCTTTCTCCACCTGTC	Forward
chr17:7,130,443	GCAAATAGAAGGAAGACGTAGGG	Reverse
chr17:7,130,605	CTCTTTTAGAGCAGGAGGTGAAAC	Forward
chr17:7,130,605	CCCTAAATACTCAAGTTCTGTGCTG	Reverse
chr17:7,131,853	CTATAAAGTCACTGCTGAAGACAAGC	Forward
chr17:7,131,853	ATCCTCAAGTACCTCCACAATAGG	Reverse
chr17:7,134,553	CCAGTTACATTAGTGGCTTCTGC	Forward
chr17:7,134,553	TCACTATTCCCTTACCACCTTCC	Reverse
chr17:7,134,917	CTAGAGAAGAAACCCAGACACTGC	Forward
chr17:7,134,917	GTCCTAAGTATGTCCCTGTCATCG	Reverse
chr17:7,137,705	CTTCAAGCCAACCAGGTTTC	Forward
chr17:7,137,705	GACTAGGGTAGGAATACCTTGAAGC	Reverse
chr17:7,137,709	AAGCCAACCAGGTTTCACTAGTAGC	Forward
chr17:7,137,709	GGTGGACTAGGGTAGGAATACCTTG	Reverse
chr17:7,137,790	CTTTCAAAAAGCCTTCCAACTCTAAGTC	Forward
chr17:7,137,790	AGGTGAGTACCCAGTTCTAGGTGAAG	Reverse
chr17:7,137,847	CTTCACCTAGAACTGGGTACTCACC	Forward
chr17:7,137,847	GCTAGAAAAACCCTCAGTTATGACC	Reverse
chr17:7,139,824	GCTGAGTTTTGATCAATGTGTC	Forward
chr17:7,139,824	AGTCTAGGGCGGTAGACATCAG	Reverse
chr17:7,140,384	GAGGCTCCAGCAGTATCTCCTC	Forward
chr17:7,140,384	CTGCCCTGGCTAAAAATACAAC	Reverse

Primer name	Orientation	Sequence
chr17:7,140,656	ACCCTCCACTTGCTATCTATCG	Forward
chr17:7,140,656	TCCGAATCTTCAACTCTGAAAG	Reverse
chr17:7,140,822	TAGACTGTTAAGGACCGACTTACCC	Forward
chr17:7,140,822	ATTTCTTGAGCTCAGGGCTAGG	Reverse
chr17:7,143,180	CCTTCTTTTTCTGTCGCTTCC	Forward
chr17:7,143,180	AACACTTCAGGTGAGCAGGAAC	Reverse
chr17:7,144,737	GAGTTTACGTCATCACCACTTATCC	Forward
chr17:7,144,737	AGACTGTTCTACGCAAAATGACTGT	Reverse
chr17:7,154,486	GAATTTCTTGTGGTGTTTTTCCTTA	Forward
chr17:7,154,486	CAAGTCAAAAGATGGAAAAACTGAT	Reverse
chr17:7,155,157	GACTTCCAGGTATGTAGATGGTCTG	Forward
chr17:7,155,157	CATAAACCGCTAACCCATCCTC	Reverse
chr17:7,157,782	GAACTCGCCTACAAACTCCTTG	Forward
chr17:7,157,782	GATACACACAAGTCACTGGGTTTC	Reverse
chr17:7,159,682	ACGGTGTTTATTGGCTCCTTAG	Forward
chr17:7,159,682	CCTCTGTACAGTTGTTACTGACTCTG	Reverse
chr17:7,161,133	CTAGCAAGACGAGATGAGAAAAGG	Forward
chr17:7,161,133	ACTGGGGTCTTCATGATTCG	Reverse
chr17:7,163,222	CCACTGTCGGTTTAGGAAATCTATC	Forward
chr17:7,163,222	GAGTAGTTCTTTCTTCCCCAGGTG	Reverse
chr17:7,165,352	AGCAGAAGACCTCAACCATCC	Forward
chr17:7,165,352	GTAGACCTGAACGCTTCCTTG	Reverse
chr17:7,165,804	CTTCTCATCTAGCTCTTCCACTTTC	Forward
chr17:7,165,804	GCTTATGTTTTCTCTGTCTCGTAGG	Reverse
5'200k#8	ACCTGGAGCCCCCAGAAG	Forward
5'200k#8	CTGTGAGCTGCCGAGTGCTA	Reverse
5'200K#9	AGGGTTCCTCCACATCCAG	Forward
5'200K#9	TTCTGGGCAGCTCAGTCC	Reverse
Primer 5'50k	TGAGGTGGGCATCACTAACA	Forward
Primer 5'50k	CCCAGAAATGGACGTTTGAC	Reverse
ATP1B2ex7b	TGTGAAGTTCCTGAATGTGACC	Forward
ATP1B2ex7b	CGACATTCTACATTCACCTCCA	Reverse
ATP1B2exon7	CAGACGATGAGCGAGACAAG	Forward
ATP1B2exon7	TGTTGATGCGGAGTTTGAAG	Reverse
ATP1B2-p53	TCCTTGGTTTGTCTACCACTCC	Forward
ATP1B2-p53	ACTGCTGGAATGAGGTGGAT	Reverse
ATP1B2-p53#1	ACTAAGTTCTCTAAAATCTGGCAAGG	Forward
ATP1B2-p53#1	AGAATGGAGAGGGGGTCCTA	Reverse
ATP1B2-p53#2	TTCCAAACATCCACTCTAACCA	Forward
ATP1B2-p53#2	TGCATTCAAGAGGAACAAAAA	Reverse
ATP1B2-p53#3	GCTCTTGACCTTAATTGAAATTATCT	Forward
ATP1B2-p53#3	TGATAATGGAATACCAATTTTCCTAA	Reverse
ATP1B2-p53#4	AGGTTGTGAATTACAAGTTTAGACTGA	Forward
ATP1B2-p53#4	AATCTCCCAATTTCCTCATGG	Reverse
ATP1B2-p53#5	CAGAGGCAAGGACAGGGACT	Forward
ATP1B2-p53#5	ACCCAGCAGGCAAACTAGAG	Reverse
ATP1B2-p53#6	GGCTCAGCCTTGCTAAATCA	Forward
ATP1B2-p53#6	GCCTCTCACCAAGGATTACG	Reverse

Primer name	Orientation	Sequence
ATP1B2-p53#7	ATCTCCCAAACATCCCTCAC	Forward
ATP1B2-p53#7	CATACCAGCTTAGATTTTAAGGTTTTT	Reverse
p53exon11	TTCTGACGCACACCTATTGC	Forward
p53exon11	TTTGGGTTTTGGGTCTTTGA	Reverse
p53exon10	TCTGAGTCAGGCCCTTCTGT	Forward
p53exon10	CAGTCTACCTCCCGCCATAA	Reverse
p53exon10-9	ATTAGGCCCCTCCTTGAGAC	Forward
p53exon10-9	CCAGGGACGAGTGTGGATAC	Reverse
p53exon9	CTGGGCATCCTTGAGTTCC	Forward
p53exon9	CTTCGAGATGTTCCGAGAGC	Reverse
p53exon9-8	TGTCTTTGAGGCATCACTGC	Forward
p53exon9-8	TACCTGCAATTGGGGCATT	Reverse
p53exon8	TCCATCCAGTGGTTTCTTCTTT	Forward
p53exon8	CCAACAACACCAGCTCCTCT	Reverse
p53exon8-7	GAGGCATAACTGCACCCTTG	Forward
p53exon8-7	AAGCAAGCAGGACAAGAAGC	Reverse
chr17:7,519,754	CAAAAGCCAAGGAATACACG	Forward
chr17:7,519,754	CTGGGGATGCAGAACTTTTC	Reverse
chr17:7,519,934	CCAAAGGGTGAAGAGGAATC	Forward
chr17:7,519,934	CCCCCTGCATTTCTTTTG	Reverse
chr17:7,520.031	ACTGACCGTGCAAGTCACAG	Forward
chr17:7,520.031	TCTGGGCTTCTTGCATTCTG	Reverse
chr17:7,520,227	TCATCTGGACCTGGGTCTTC	Forward
chr17:7,520,227	TCCCCGGACGATATTGAAC	Reverse
chr17:7,520,453	GACAAGAGCAGAAAGTCAGTCC	Forward
chr17:7,520,453	TAGCAGAGACCTGTGGGAAG	Reverse
chr17:7,523,833	CACTCCATTAATGCAAGTACACCTC	Forward
chr17:7,523,833	CTGTTGACTGAATAGATCCCTGAAG	Reverse
chr17:7,523,886	AGGAGGTGCTGTCCTGAAATG	Forward
chr17:7,523,886	TCCAAGTGGGATTGAGACTTTG	Reverse
chr17:7,524,009	TTTTCAGAGCAGGACCTTCC	Forward
chr17:7,524,009	TGTATCCTGGCCCACTGATG	Reverse
chr17:7,530,048	CAGGTGCATGGGAAGAAACT	Forward
chr17:7,530,048	ATCTCTTAGCTCGCGGTTGTT	Reverse
chr17:7,533,018	AAACGGGAGCCTTTCTGAAG	Forward
chr17:7,533,018	TTTTCCAGACCCCAACTCTG	Reverse
chr17:7,533,886	CCCCGAATTCTGAAGTTATTTAGAG	Forward
chr17:7,533,886	TCCTAATTAAAGAAAGGGGAAACCT	Reverse
chr17:7,539,366	GTGTGTACTGAGTCTTCTTCCCAAT	Forward
chr17:7,539,366	GAAAGGAGAAAACCCGAGAAACTAT	Reverse
Primer 3'50k	AAGCGATCCTCCTGGCTAAC	Forward
Primer 3'50k	ATTCAAGCATGGTGGCATGT	Reverse
Primer 3'200k	CACATTTTCCCTGAGGCAGT	Forward
Primer 3'200k	CCCTTCCTCATTTACCTTTCC	Reverse
3'200K#1	CACTGGGTCCACAGAATCAT	Forward
3'200K#1	CAATGTTGCATGGCTTGAA	Reverse
3'200K#2	TCCCAGCAAAGGCTGTTAAA	Forward
3'200K#2	TGAAGGAGAAGCTCCTGGAA	Reverse

Primer name	Orientation	Sequence
3'200K#9	TCAGCGTTCAGGTCATTGAA	Forward
3'200K#9	TTCCAGAAACCCCTCTCCTT	Reverse
3'200K#10	CTCTGTTAGCTGCCGTGTTG	Forward
3'200K#10	GCAGGAAGGAGGAAAAATGA	Reverse
chr17:8,056,064	TATTCCCACTTCTGAAAGCACTTAG	Forward
chr17:8,056,064	GTTTCCTTCTCAAATCCTCAAAACT	Reverse
chr17:8,056,181	TTATGGTTATTGGATCAGGCATAAG	Forward
chr17:8,056,181	CTAGCTGGCCCACTTAAATCAC	Reverse
chr17:8,059,157	CATTGTCTCTTCTGTGGAGAGG	Forward
chr17:8,059,157	TGATGACCAACCATCTCTGC	Reverse
chr17:8,059,212	GGACTTAAGTTTTGCCTTTAATGTTC	Forward
chr17:8,059,212	CAAACAGTTCCATGACTCTATACACC	Reverse
chr17:8,061,386	AGTCAAATCTTAACACATTTTGC	Forward
chr17:8,061,386	CCACCTGTATGCCTTTTAGTTC	Reverse
chr17:8,063,581	TCGCCAGAGTTTCTATGCTGTTAC	Forward
chr17:8,063,581	GAAGAACTGCTGATGACCTGAG	Reverse
chr17:8,065,022	ATAACGGGCGGACTTTTGTC	Forward
chr17:8,065,022	GCGCTGACAAATCTTAAAGGTG	Reverse
chr17:8,066,390	AGACTTTACACTCTGCGTGCTG	Forward
chr17:8,066,390	AAGAACAGAACCCCTAGGAACC	Reverse
chr17:8,067,624	GCTTCCGGACTGTAAGATCAG	Forward
chr17:8,067,624	AGCAGAGGTTAGACTGTAGGGTAGG	Reverse
chr17:8,069,257	AGGCCAGATTAGATTCCACTTCTC	Forward
chr17:8,069,257	AGCACTAACGGCACTGTAAGAAC	Reverse
chr17:8,070,736	AGGCTTAGGGCTTCGTTTTC	Forward
chr17:8,070,736	GTCAGTCAAGGGAAGTAAGAAAGG	Reverse
chr17:8,071,660	CAACTCCCCAATTTACTTCCTTTAC	Forward
chr17:8,071,660	CAGAAGAAATGGTACCAAAATCAGT	Reverse
chr17:8,081,793	TCCCTTCCACAGTAAAGAATAACCT	Forward
chr17:8,081,793	GGGCTATGGACATTTTGTATCATTA	Reverse
chr17:8,131,927	ATACCCAGGTTGGCTCTCACTTTC	Forward
chr17:8,131,927	CCAGGTTAGGATTCAGGTCTCAC	Reverse
chr17:8,331,999	TGCTAAGAGGTACAGAGGACTAAGC	Forward
chr17:8,331,999	TCCTCACAGCTCTAGGAAACAG	Reverse

QPCR cycling conditions (repeated for 40 cycles): 95°C for 10 seconds; 60°C for 15 seconds; and 72°C for 10 seconds. Preceded by 95°C for 5 minutes. Abbreviations: Tm, melting temperature.