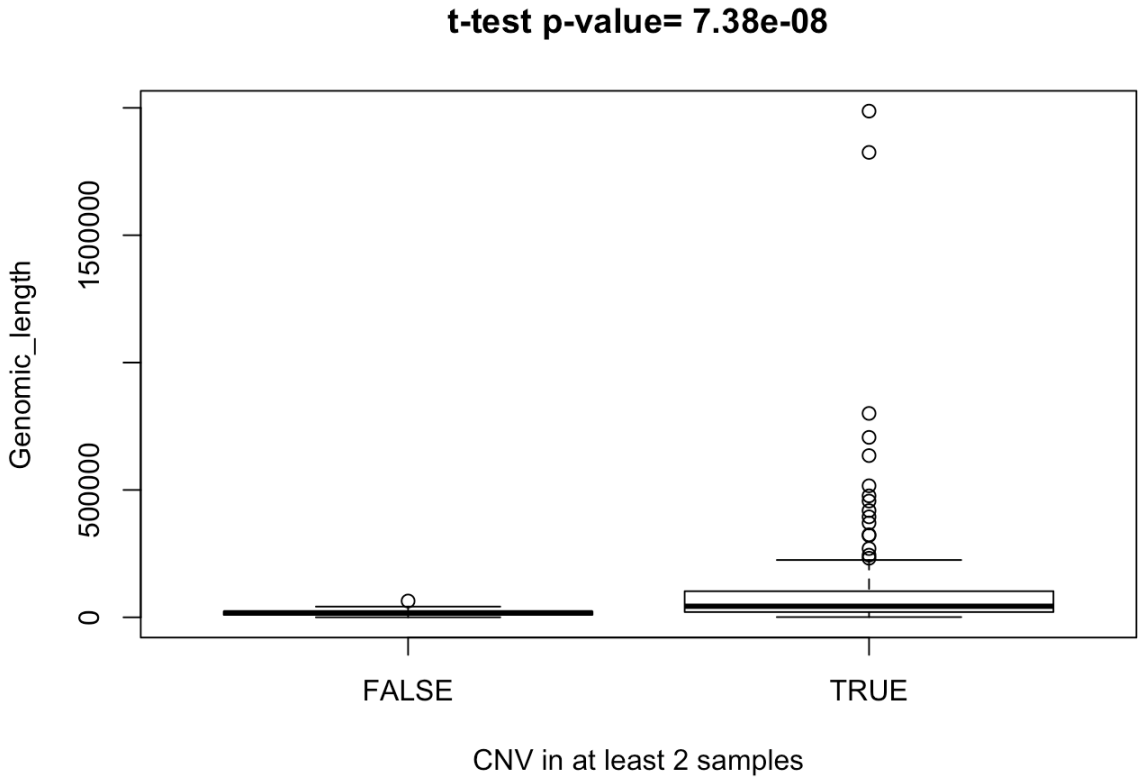
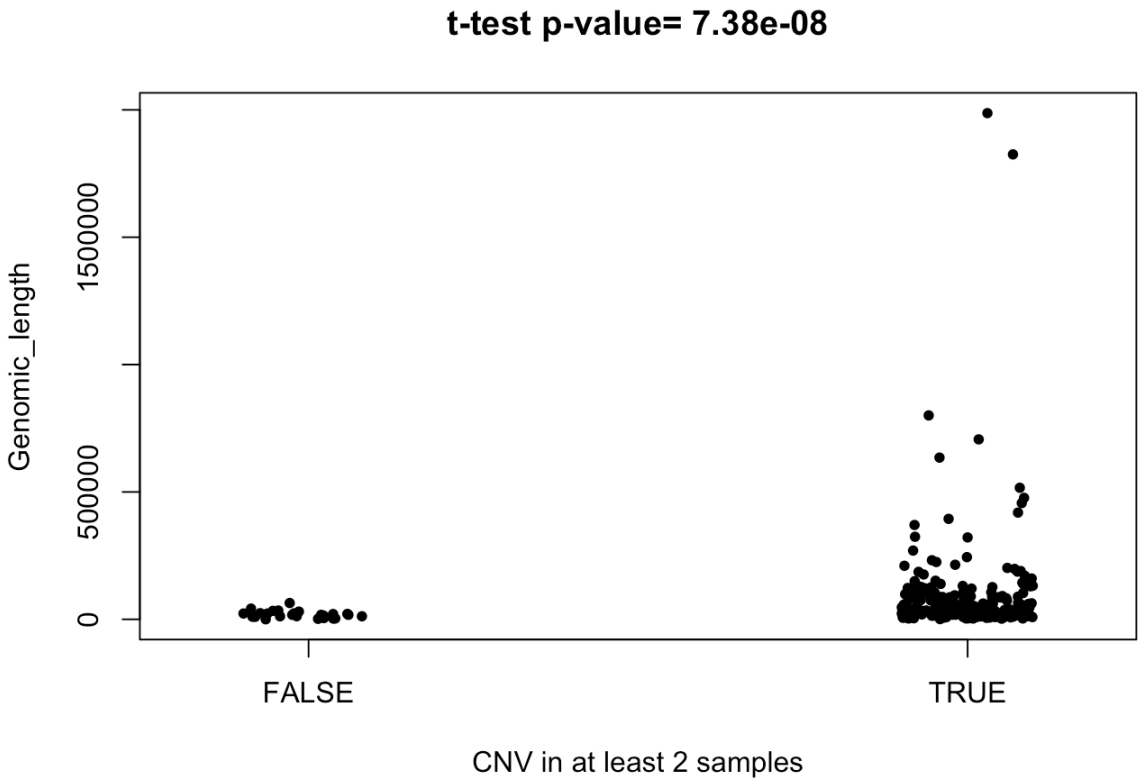
SUPPLEMENTAL FIGURES

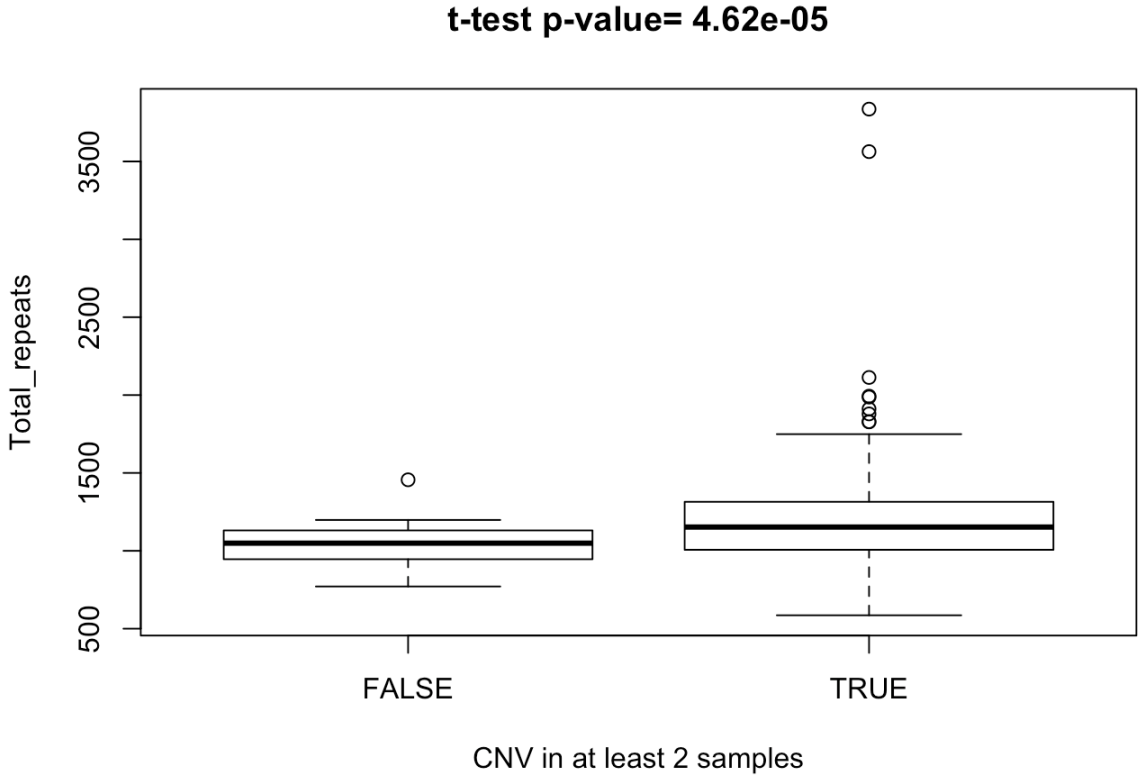
A

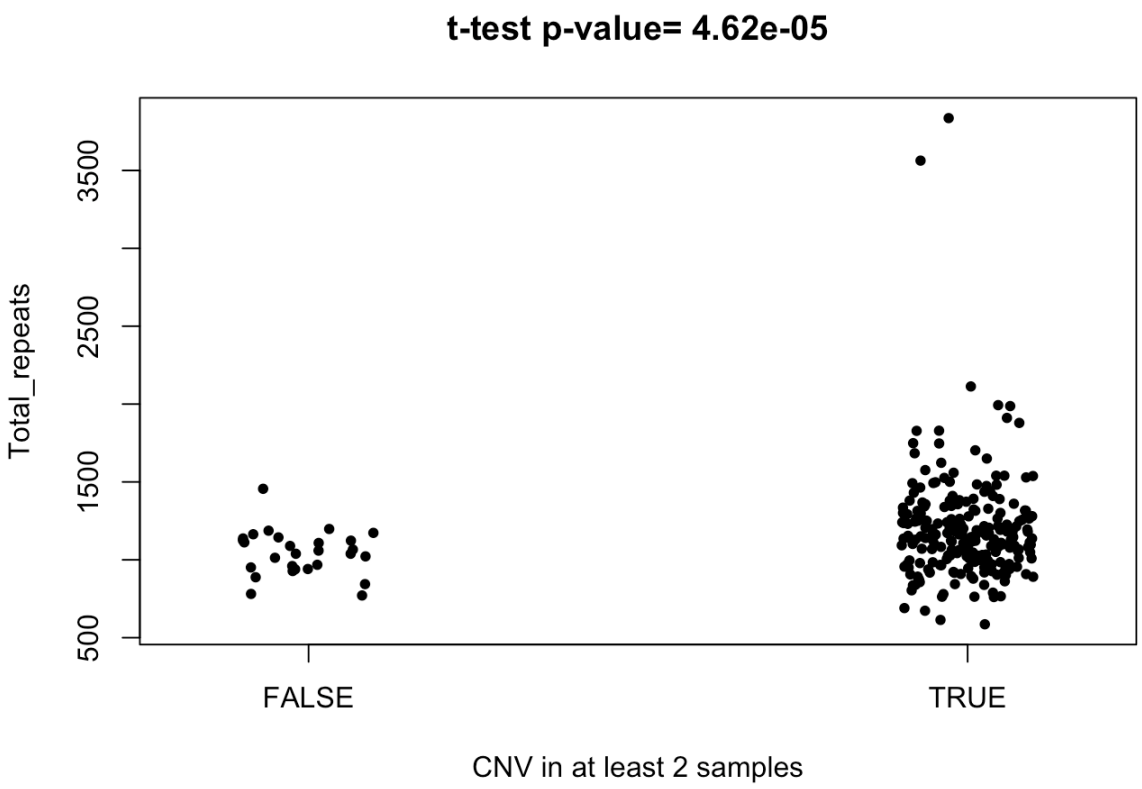
	CNVs.DGV	Total.CNVs.lit	Unique.CNVs.lit
Gene.size	**0.40 ($2.00*10^{-16}$)**	**0.13 (0.0087)**	**0.14 (0.0035)**
Total.repeats	**0.19 ($9.80*10^{-6}$)**	0.016 (0.75)	0.025 (0.62)
Alu.repeats	0.042 (0.34)	-0.051 (0.30)	-0.048 (0.33)
LINE and LTR.repeats	**0.30 ($9.35*10^{-12}$)**	**0.11 (0.029)**	**0.13 (0.011)**
SDs	0.045 (0.33)	0.092 (0.073)	0.080 (0.12)

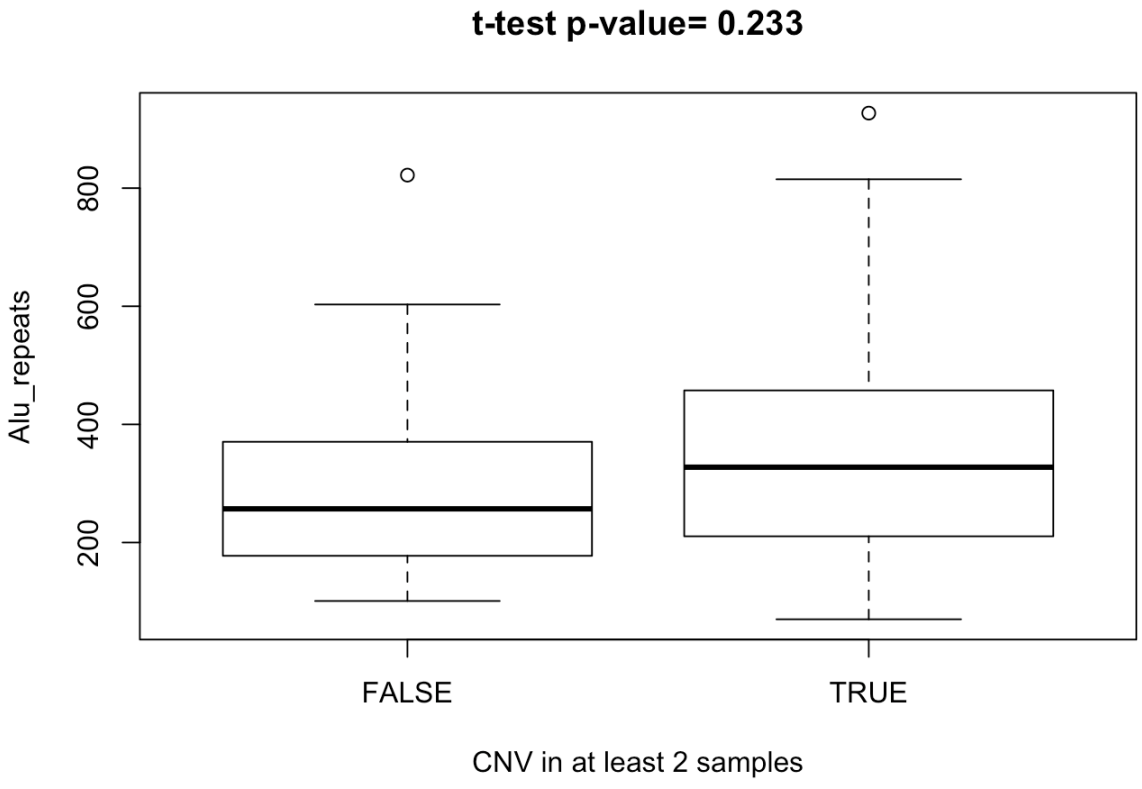
B

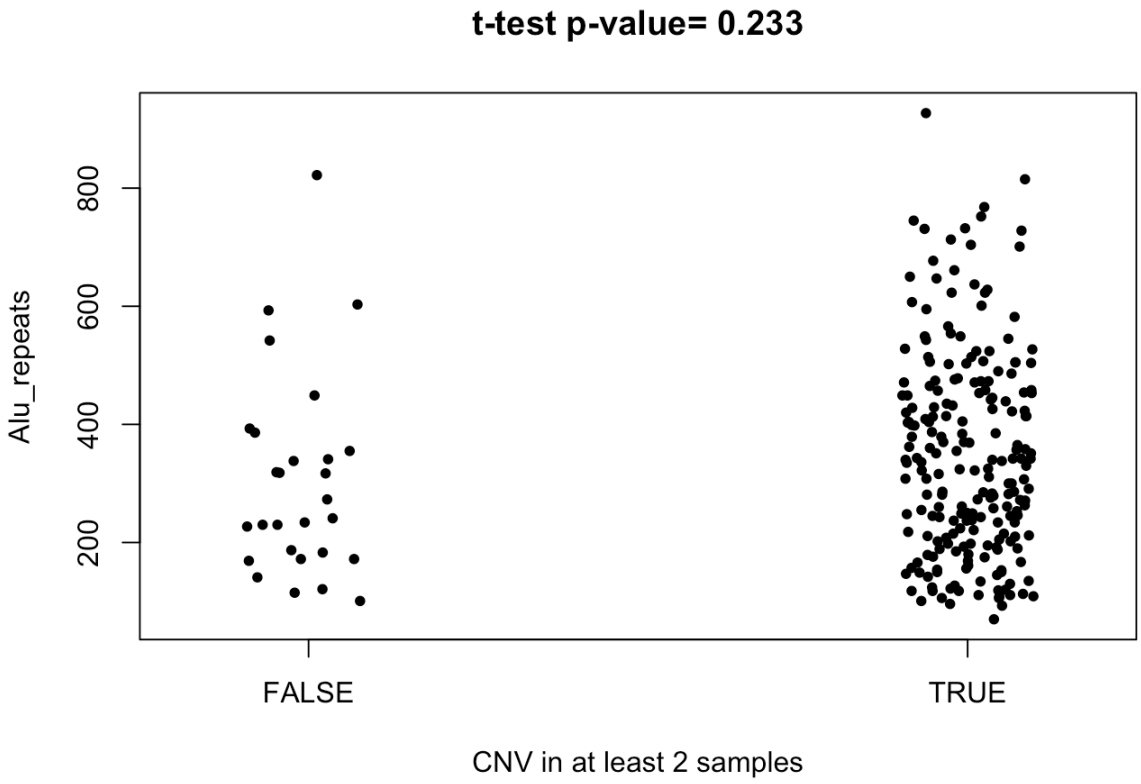


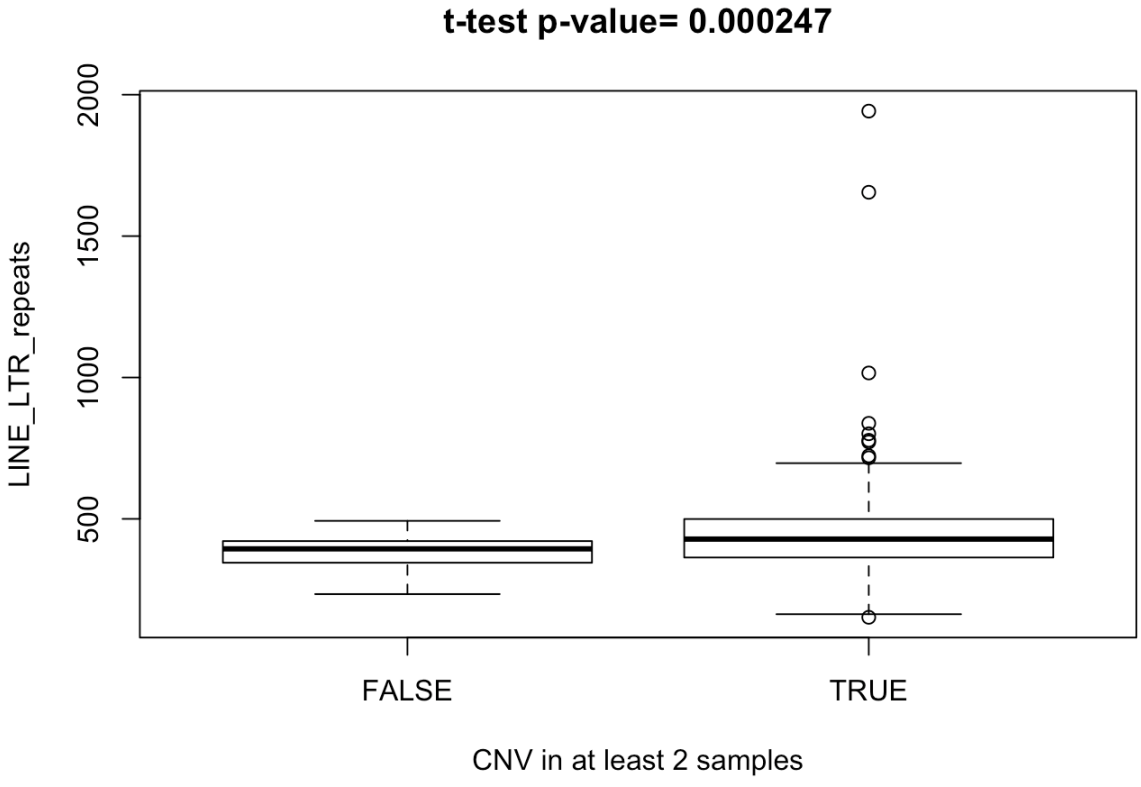

t-test p-value= 7.38e-08
Genomic_length
0
500000
1500000
FALSE
TRUE
CNV in at least 2 samples

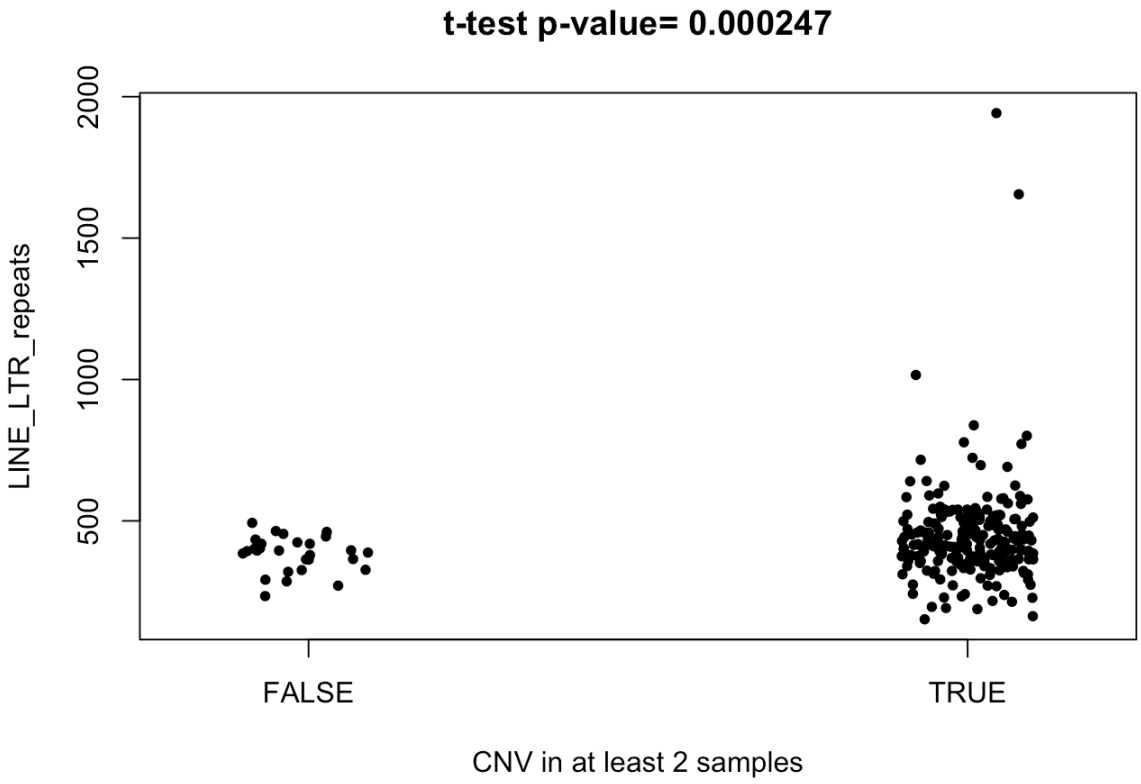

t-test p-value= 4.62e-05
Total_repeats
500
1500
2500
3500
FALSE
TRUE
CNV in at least 2 samples

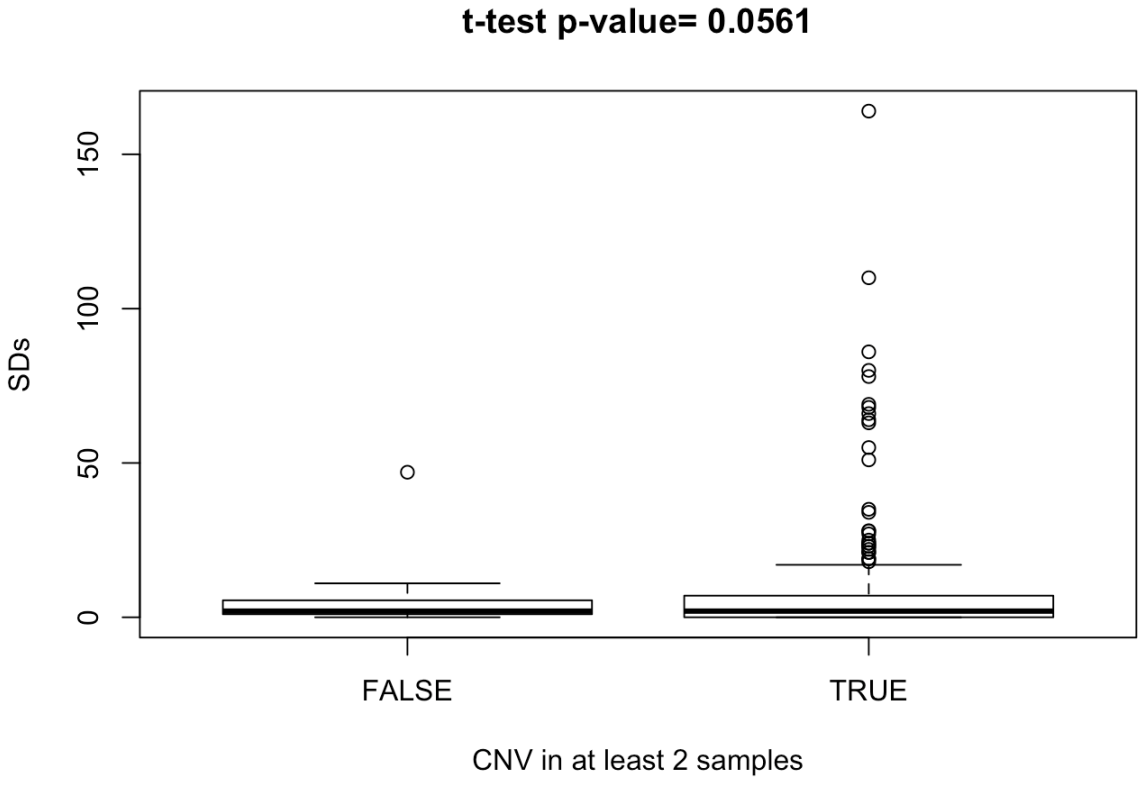

t-test p-value= 4.62e-05
Total_repeats
500
1500
2500
3500
FALSE
TRUE
CNV in at least 2 samples

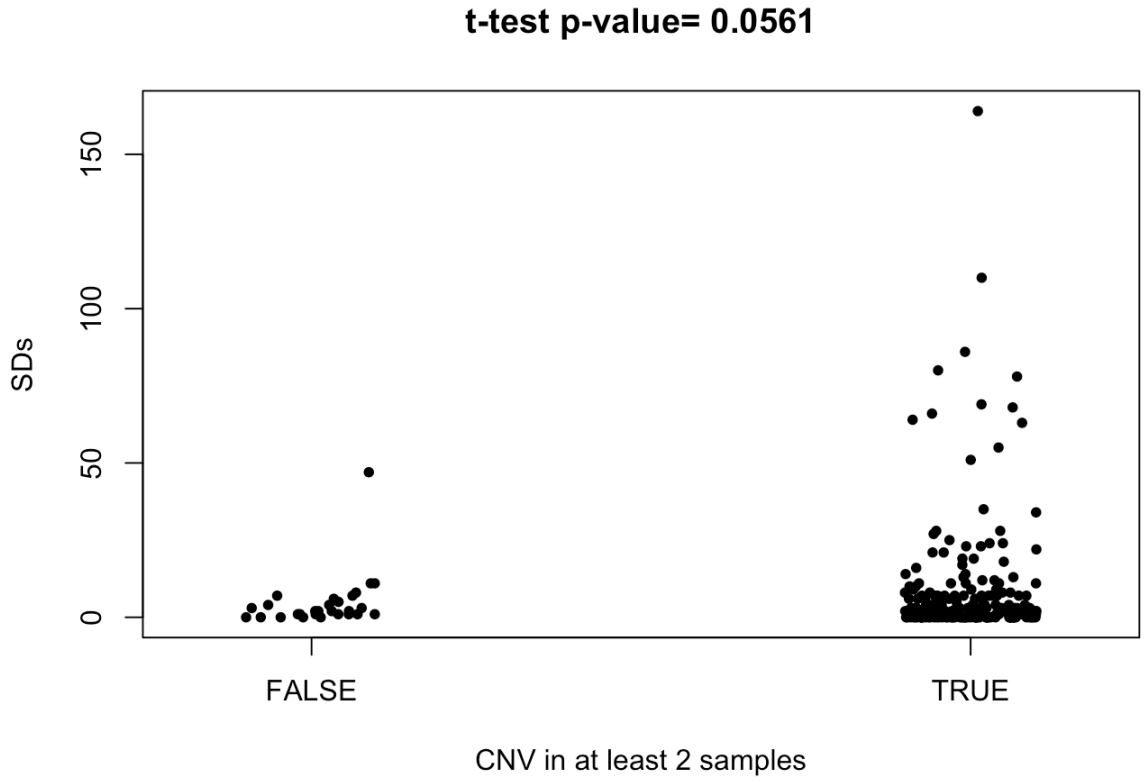

t-test p-value= 0.233
Alu_repeats
200
400
600
800
FALSE
TRUE
CNV in at least 2 samples


t-test p-value= 0.233
Alu_repeats
200
400
600
800
FALSE
TRUE
CNV in at least 2 samples


t-test p-value= 0.000247
LINE_LTR_repeats
500
1000
1500
2000
FALSE
TRUE
CNV in at least 2 samples


t-test p-value= 0.000247
LINE_LTR_repeats
500
1000
1500
2000
FALSE
TRUE
CNV in at least 2 samples


t-test p-value= 0.0561
SDs
0
50
100
150
FALSE
TRUE
CNV in at least 2 samples



C

	Gene.size	Total.repeats	Alu.repeats	LINE+LTR repeats	SDs
Gene.size	**1.00**	**0.81**	**0.038**	**0.89**	**0.034**
Total.repeats	**0.81**	1.00	0.49	0.78	0.060
Alu.repeats	0.038	0.48	1.00	-0.083	0.11
LINE+LTR.repeats	**0.89**	**0.78**	**-0.083**	**1.00**	**0.014**
SDs	0.034	0.060	0.11	0.014	1.00

Figure S1. Overview of the results of the statistical analyses

A. Kendall rank correlation coefficient. Rank correlation analysis was performed using the Kendall rank correlation coefficient or tau. The table lists all tau values with their respective p-values. If tau equals 1, both compared datasets are completely concordant, while tau = -1 is obtained with two completely discordant datasets and datasets that are not correlated lead to tau = 0. All correlations with tau > 0.1 are indicated in bold and are statistically significant (p-value < 0.05).

B. **Genomic feature vs. the presence of at least two CNVs.** Calculation of the statistical difference of the values of the different genomic feautures between the group of genes with one or less CNVs and the group of genes with at least 2 CNVs was performed using a classic t-test. For each comparison a box plot and a scatter plot are shown, together with the respective p-value. ‘Genomic length’ corresponds with gene size.

C. **Correlation of the different genomic features.** Pearson correlation coefficients were calculated to assess which genomic features are correlated with each other. A value of 1 indicates a total positive linear correlation, 0 is no linear correlation, and -1 represents total negative correlation.

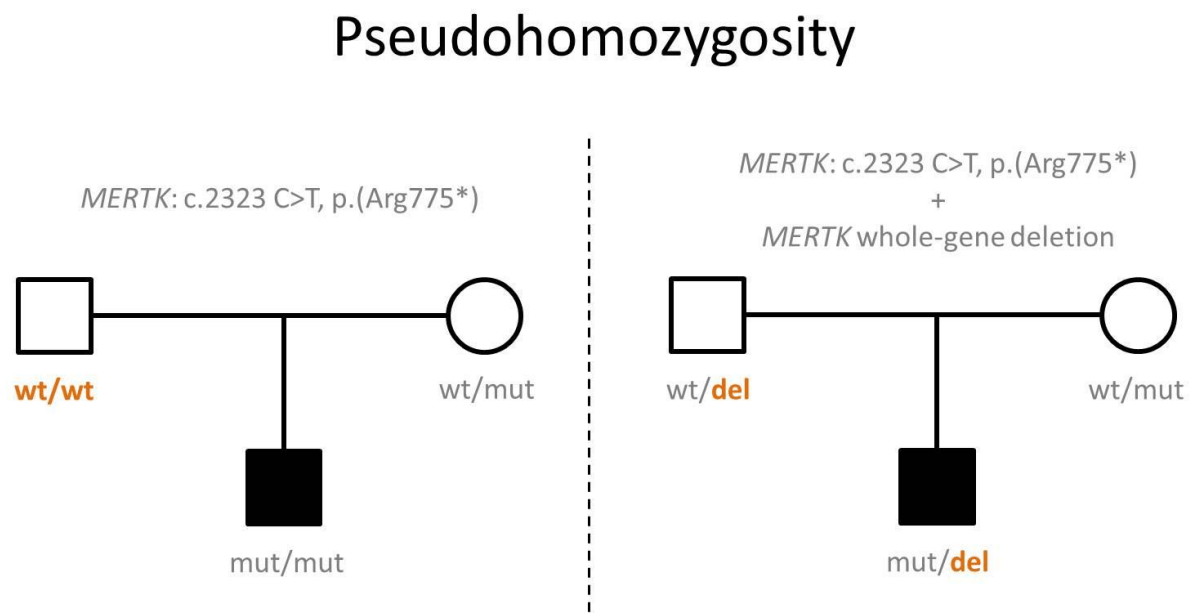


Figure S2. Pseudohomozygosity in patient with whole gene deletion of *MERTK*.

An apparently homozygous nonsense mutation in the *MERTK* gene was initially identified in P6: c.2323C>T, p.(Arg775*) (mut/mut). Segregation analysis demonstrated a recessive carrier state in the mother (wt/mut) but not in the biologically proven father (wt/wt), indicating pseudohomozygosity and illustrated in the left panel. This was confirmed by qPCR analysis, revealing a heterozygous whole gene deletion of *MERTK* in both the index patient (mut/del) and his father (wt/del), represented in the right panel. Wt, wild type; mut, mutation; del, deletion.

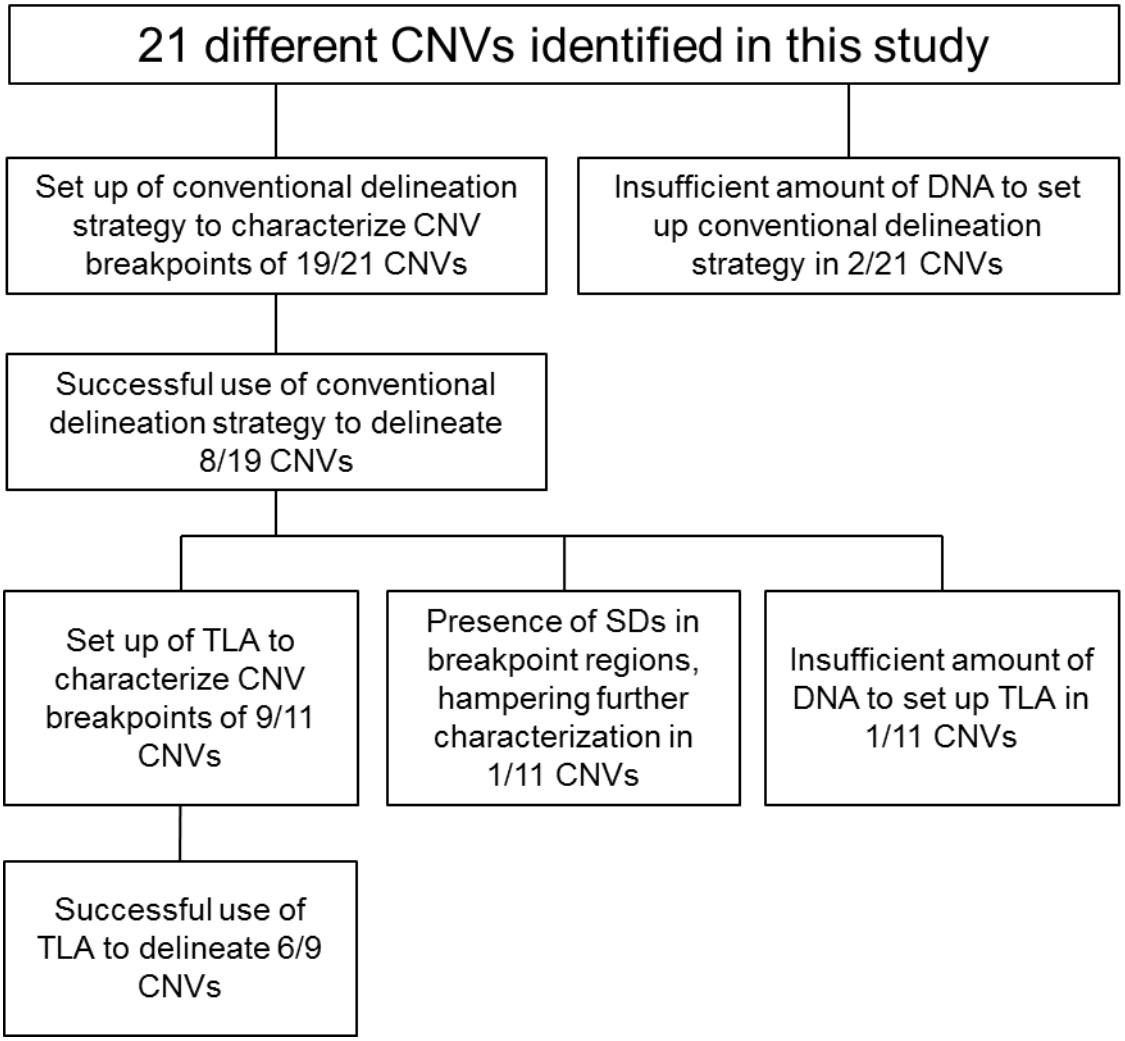


Figure S3. Flowchart of characterization of CNVs.

Flowchart illustrating the number of CNVs delineated at the nucleotide level using either a conventional delineation strategy or TLA.

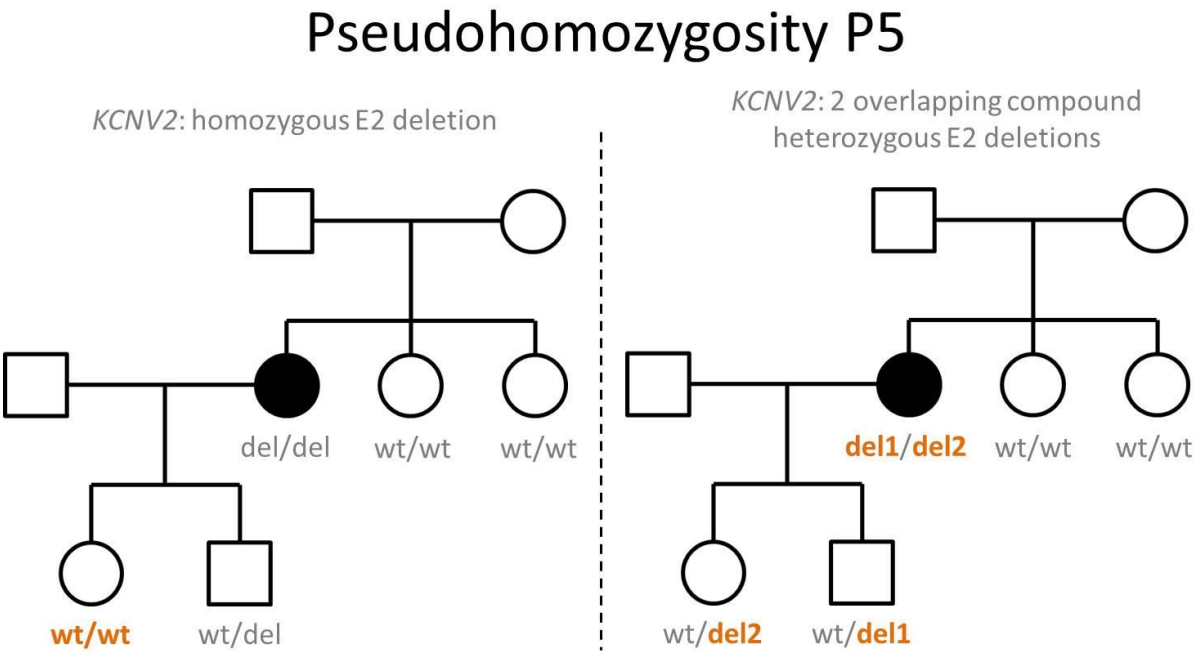


Figure S4. Pseudohomozygosity in patient with overlapping *KCNV2* deletions.

In patient P5 we first performed *KCNV2* mutation screening by PCR and Sanger sequencing. Non-amplification of exon 2 of the gene suggested a homozygous deletion (del/del). After delineation of the deletion, segregation analysis was performed in the family using the junction PCR primers. Although both children of the index patient are obligate carriers of this deletion, segregation of the junction product could only be demonstrated in one of the children (wt/del), illustrated in the left panel. Subsequent qPCR of exon 2 of *KCNV2* in the daughter demonstrated a heterozygous deletion of this exon. The most probable explanation for this finding is the occurrence of two different overlapping heterozygous deletions, only one of which was identified by the initial delineation. This was corroborated using qPCR, revealing a second larger deletion overlapping with the smaller one found in the index case (del 1/del 2) and in the daughter (wt/del2), represented in the right panel. Wt, wild type; del, deletion; E, exon.

Microhomology

BEST1 deletion (E1-2): chr11: g.61711373_61719810del

```
Proximal    cataagtggaaaaggtgaattttacaggacacggattgcccctcaatatt
Deletion    cataagtggaaaaggtgaattttacaggacacggattgcccctcaatatt
Distal      gaatcccacacagactcataggcccacatagtacattaaaaaagagagag

Proximal    aaaattataaaaaaaaagactggaaGGAaatacatcaaaatgataaccttg
Deletion    aaaattataaaaaaaaagactggaaGGAgtctcactgtgttgtccaggctg
Distal      agagagagagagagagagagatGGAgtctcactgtgttgtccaggctg

Proximal    tttgggagtttgttttgttttgttttaaactgctctgtagtttcctaatt
Deletion    gtctcgaactcctaggctcaagcaatccccctgccttagcctcccaaggg
Distal      gtctcgaactcctaggctcaagcaatccccctgccttagcctcccaaggg
```

KCNV2 deletion 1 (E2): chr9: g.2723737_2766822del

```
Proximal    acatggaagcaataacctttgcttctacattcatttgggaagaaaataaa
Deletion    acatggaagcaataacctttgcttctacattcatttgggaagaaaataaa
Distal      ctatacatgctgctatgtctctatacatgctgctagtattagggaagttt

Proximal    acagaaatgattctttttctactgAGGaaaagaaagaacctataaaacat
Deletion    acagaaatgattctttttctactgAGGtaaaacggtcataaatttgatgt
Distal      aacctcaaaaagtttcccagagttAGGtaaaacggtcataaatttgatgt

Proximal    acgagaaggtcagctgttccctcatgggagaacttctgagagtccagata
Deletion    ggaaaaataattgcttcagtatagtcaaggaagacaactaggaaatataa
Distal      ggaaaaataattgcttcagtatagtcaaggaagacaactaggaaatataa
```

KCNV2 deletion 2 (E2): chr9: g.2724293_2730624del

```
Proximal    ttttgtcccagagaccttgctcccatctttctttccctaaggtatgtttt
Deletion    ttttgtcccagagaccttgctcccatctttctttccctaaggtatgtttt
Distal      aaattgtttaagtggatgttatctagtgaaatctctagaccagtggttct

Proximal    cattaccttgccaataggtggcacttacaagatcactgaaattattttat
Deletion    cattaccttgccaataggtggcactccagctaatgcctagagatggtttt
Distal      caaaggcaaattcattcctcagaggccagctaatgcctagagatggtttt

Proximal    ttttgttcattgattgctttgaggaaacccatttgaaaaataaattaggt
Deletion    gattgccaataccgtgaggggcaataagggagacttctggcatctaggag
Distal      gattgccaataccgtgaggggcaataagggagacttctggcatctaggag
```

***PCDH15* deletion (E1): chr10: g.56478660_57777934del**

```
Proximal    agtagctggcactacaggcatgcaccaccatgtccagctaatttttgtat
Deletion    agtagctggcactacaggcatgcaccaccatgtccagctaatttttgtat
Distal      atataaatttttaaaataccatttacagtaacatccaaaaaataaaataa

Proximal    ttttagtagagacggggtttcactATGttggccaggctggtcttgaactc
Deletion    ttttagtagagacggggtttcactATGagttgaaagatctatgtactaaa
Distal      aatatcaagaaataaatttatccaATGagttgaaagatctatgtactaaa

Proximal    ctgacgtcctgatccacccgcctcagtctcccaaagtgctaggattacag
Deletion    aatttcagaaaattgatgcaaaaaattgaagaaaacaaaaataaatggaa
Distal      aatttcagaaaattgatgcaaaaaattgaagaaaacaaaaataaatggaa
```

***PDE6G* deletion (E3-4): chr17: g.79612725_79619171del**

```
Proximal    catccttgccttcctggtgcttgaggccgtggcgggggccctggtggtgg
Deletion    catccttgccttcctggtgcttgaggccgtggcgggggccctggtggtgg
Distal      tttgtgcataagagtgtcatgcccgtgtgtgcgcacacgtgtgagtggac

Proximal    ccctctggggcccgctgcaagacaGCctggagcacaccctgcgtgtggcc
Deletion    ccctctggggcccgctgcaagacaGCggtgcctgtgtgcgcacacgagtg
Distal      actcctcggtgcgttgatgggagcGCggtgcctgtgtgcgcacacgagtg

Proximal    atcgcccactaccaggacgacccagacctgcgcttcctcctcgaccaagt
Deletion    agtggacactcctcggtgcactgatgggagcgagatgcccgtgtgtgcgc
Distal      agtggacactcctcggtgcactgatgggagcgagatgcccgtgtgtgcgc
```

***PRPH2* deletion (E1-3): chr6: g.42631162_42716988delinsTATTTTT**

```
Proximal    ccccttttgtgaatgcttgagtaatactgttattcctctgctgcttcctcc
Deletion    ccccttttgtgaatgcttgagtaatactgttattcctctgctgcttcctcc
Distal      ttccaacatatataacatggagagggtattagaatagtgttccaaggact

Proximal    aagaaatatttttaacaagtaa-------gttttggctcatgacaactat
Deletion    aagaaatatttttaacaagtaaTATTTTTaacaactgccaactgatgtta
Distal      cacatacccatcacccagcttc-------aacaactgccaactgatgtta

Proximal    tacaaagcaatagtttgctttgaagataatgaaataataaatcaaggagg
Deletion    agtaaataagccaggaacagaaagaaaaatatcacatgttctcactcgta
Distal      agtaaataagccaggaacagaaagaaaaatatcacatgttctcactcgta
```

***USH2A* deletion (E3): chr1: g.216589227_216593365del**

```
Proximal    gaaccactgcgcctggcctccattatttcttataacaacttttgagtaat
Deletion    gaaccactgcgcctggcctccattatttcttataacaacttttgagtaat
Distal      tgtatgtatatttggtataaagttatgagtgcaagtatatttttaattta

Proximal    cagtacattccttttttcaaaatctAGGctgggcactctggctcatgcctg
Deletion    cagtacattccttttttcaaaatctAGGttttatataagtagaaataaaag
Distal      ggcaaatagatttagatgacataaAGGttttatataagtagaaataaaag

Proximal    taataccatcactttgggaggccaaggcaggtggatcacctgaggtcagg
Deletion    ttttatataaattctactgcaaattatatttttaacccttcagtagttga
Distal      ttttatataaattctactgcaaattatatttttaacccttcagtagttga
```

***USH2A* deletion (E5-10): chr1: g.216465309_216507632del**

```
Proximal    ttagtcaaatggtgtgaactctttaaaaaacaaaacaaaacaaaacaaca
Deletion    ttagtcaaatggtgtgaactctttaaaaaacaaaacaaaacaaaacaaca
Distal      aacaaaaactatataggtgcaaaattttgcatactcttaaaattaagttg

Proximal    acaacaaaatccttttcctgaaaagatatagtttaagactagagtgctat
Deletion    acaacaaaatccttttcctgaaaaggaattaagatgttaatgataacatc
Distal      gtattaatttgaattagatttttatgaattaagatgttaatgataacatc

Proximal    ttctgaaaactacctagtgatttggaaataggaagatttggtgaactagc
Deletion    ctgggcaacaaacatagtagaataaatgacaagggtattaaattatacac
Distal      ctgggcaacaaacatagtagaataaatgacaagggtattaaattatacac
```

***USH2A* deletion (E12-13): chr1: g.216415487_216435280del**

```
Proximal    tcaacttgcataactacaataataatataattcctcactttgggaggcct
Deletion    tcaacttgcataactacaataataatataattcctcactttgggaggcct
Distal      gataactcactcaataagcctgattccagtaaagtcatgtcactgctgcc

Proximal    aggcaggcagatcacgaagtcaggaGatcgagaccatcctggctaacacg
Deletion    aggcaggcagatcacgaagtcaggaGaagcattcacaaacattactgaca
Distal      acaaactcccacaggttaggtcagtGaagcattcacaaacattactgaca

Proximal    gtgaaaccccatctctgctttaaaaaaaaaaaaaatacaaaaaattagcc
Deletion    tggattacagttgaagaaacttcacagagactacactactgtgcccactc
Distal      tggattacagttgaagaaacttcacagagactacactactgtgcccactc
```

***USH2A* deletion (E22-24): chr1: g.216259404_216323160del**

```
Proximal    tattttaacttcctgggagagacttgggaggcaccctgtgattcttaatg
Deletion    tattttaacttcctgggagagacttgggaggcaccctgtgattcttaatg
Distal      acaacctagctggacgatcagttcttgttaagaatctgacccctcaaact

Proximal    aggacttgggaaacaatcctagaaTAttagcattgcgtttgaagggtgat
Deletion    aggacttgggaaacaatcctagaaTActtagtcatctatagtaccccgac
Distal      ctataacctcaatggaccggacccTActtagtcatctatagtaccccgac

Proximal    ttttttcctatctaaggttaattccattcatgttcattttaattagttt
Deletion    tgccgtccgcctgcaggatcctccccactgggttcaccattccagaataa
Distal      tgccgtccgcctgcaggatcctccccactgggttcaccattccagaataa
```

***USH2A* deletion (E44): chr1: g.216040210_216042178del**

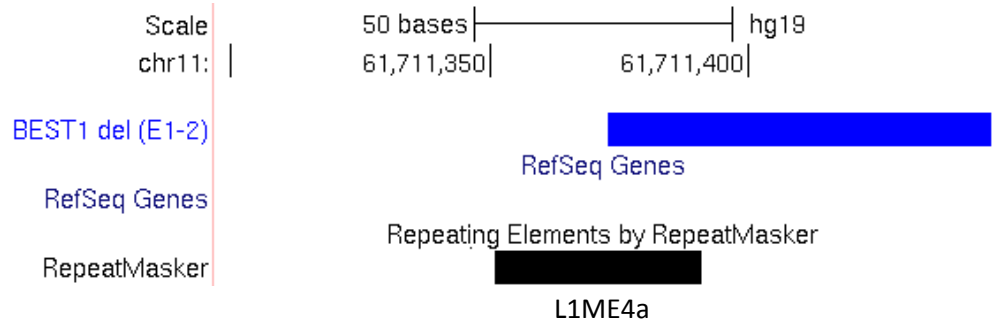
```
Proximal    gttttaatgtgtccatttctaatttatttgggtactaataaaaatggccc
Deletion    gttttaatgtgtccatttctaatttatttgggtactaataaaaatggccc
Distal      ggtgatggccatgtaaaaacagaaacagaaattggagttatgctgtcaca

Proximal    ttctaaagttttcaatgacagccctGtcaatctgcaattggtaaaaattt
Deletion    ttctaaagttttcaatgacagccctGaagttggaagaggcaagaaagggt
Distal      agccaaggatttcctagggctaccaGaagttggaagaggcaagaaagggt

Proximal    accagtaacacttcactatcaaaatgatgtgtacatgggggaggttcata
Deletion    tcttctacagagctttggaagggagtctggtccagaatacaaactggttt
Distal      tcttctacagagctttggaagggagtctggtccagaatacaaactggttt
```

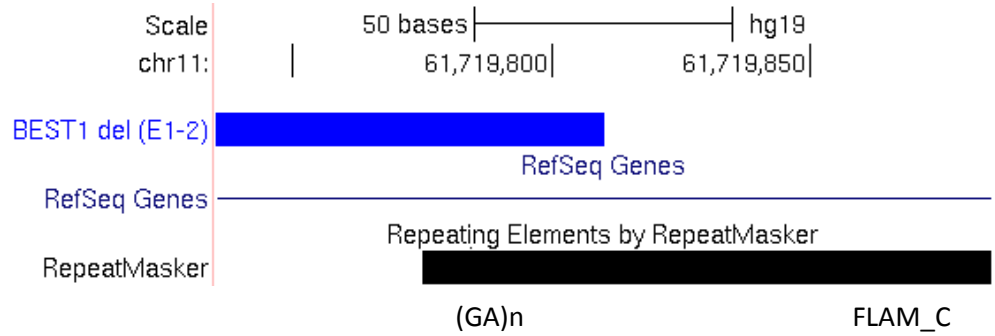
Figure S5. Visualization of microhomology at CNV breakpoints.

Multiple sequence alignments. Sequences of 150 bp surrounding the junctions of each deletion were aligned to the proximal and distal reference sequences using ClustalW. The proximal and distal reference sequences are shown in blue and green respectively. The junction sequences are depicted in the color of the reference sequence they align with. Microhomology between the proximal and distal reference sequence and the junction are shown in red. (Verdin et al., PLoS Genet., 2013)

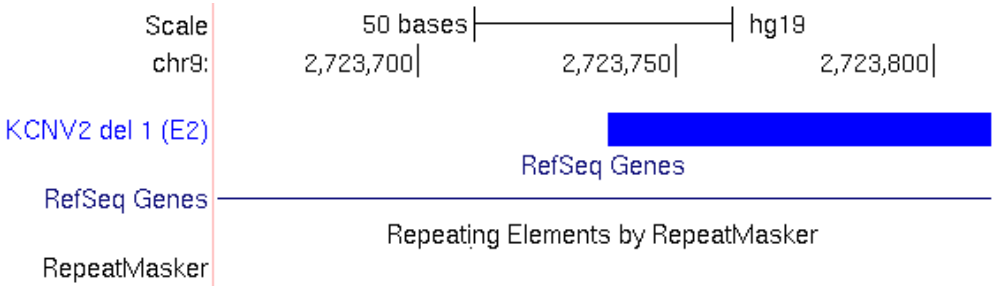
Repetitive elements

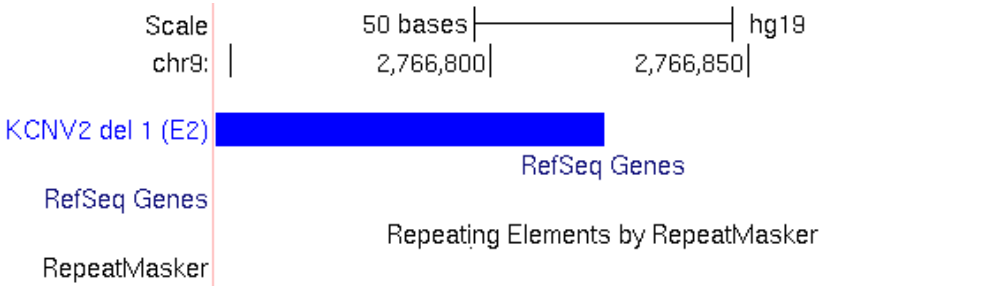
***BEST1* deletion (E1-2): chr11: g.61711373_61719810del**



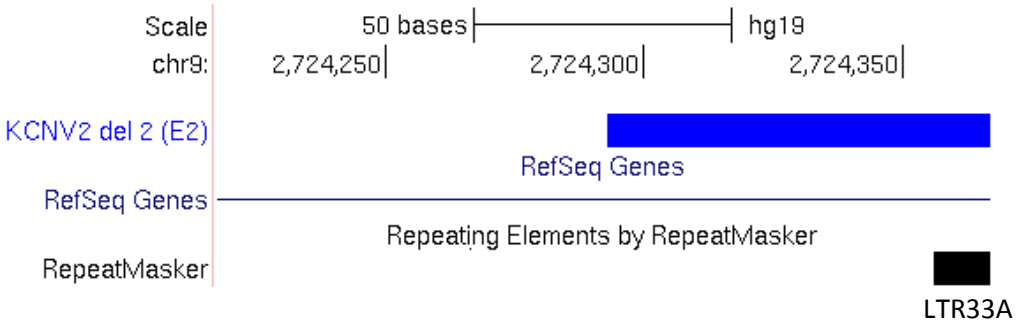


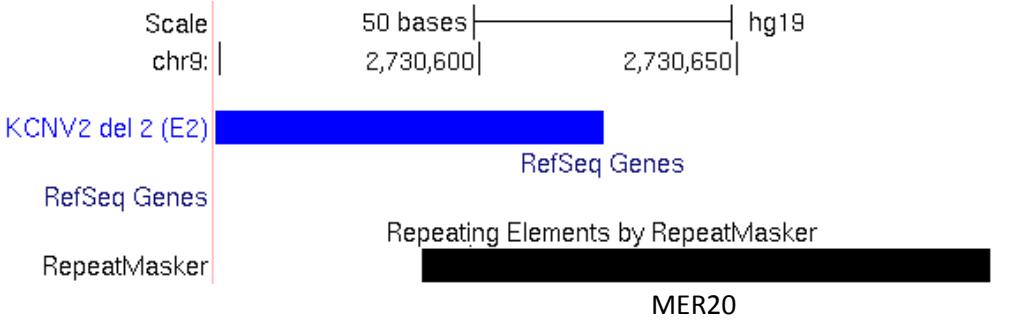
***KCNV2* deletion 1 (E2): chr9: g.2723737_2766822del**



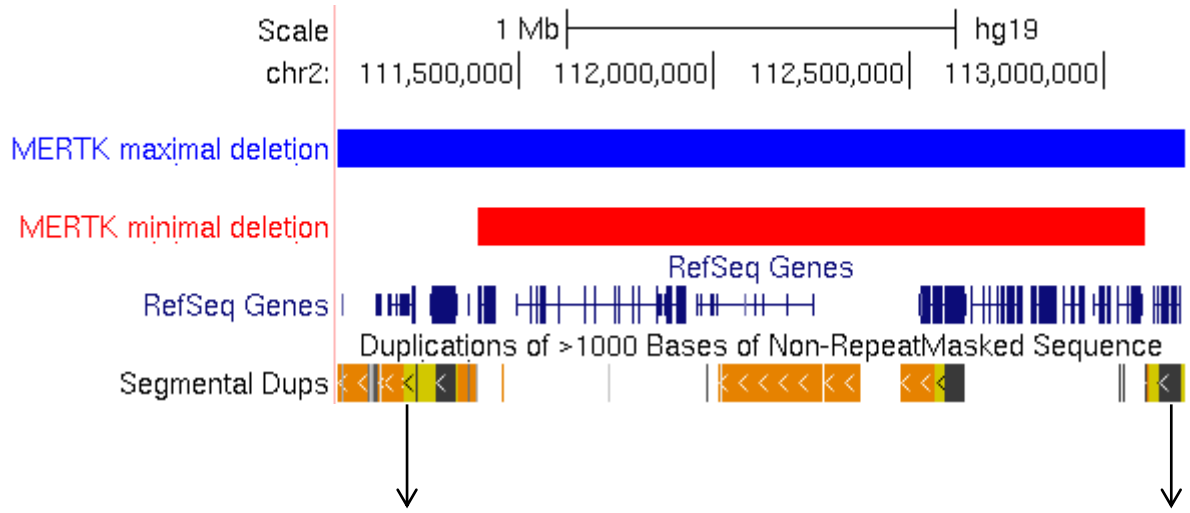


***KCNV2* deletion 2 (E2): chr9: g.2724293_2730624del**



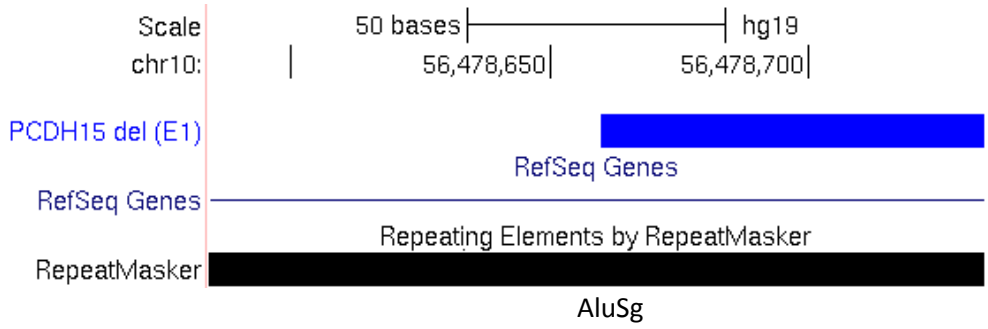


***MERTK* deletion (E1-19): maximal deletion: chr2: g.111033904_113207821del, minimal deletion: chr2: g.111392204_113103622del**

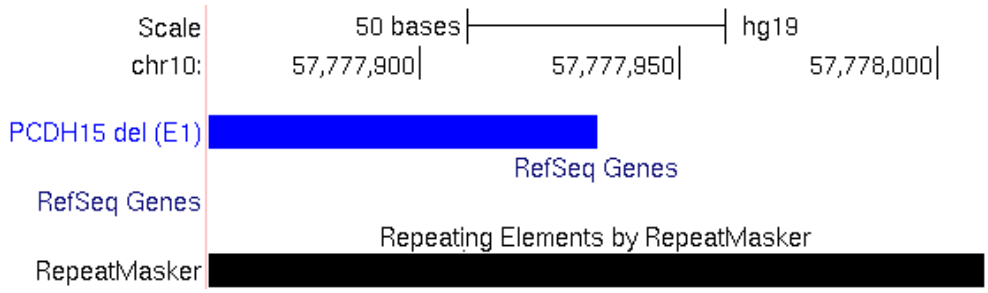


chr2: 111,258,083-111,335,142 ----> 99% identical <---- chr2: 113,112,671-113,191,577

***PCDH15* deletion (E1): chr10: g.56478660_57777934del**

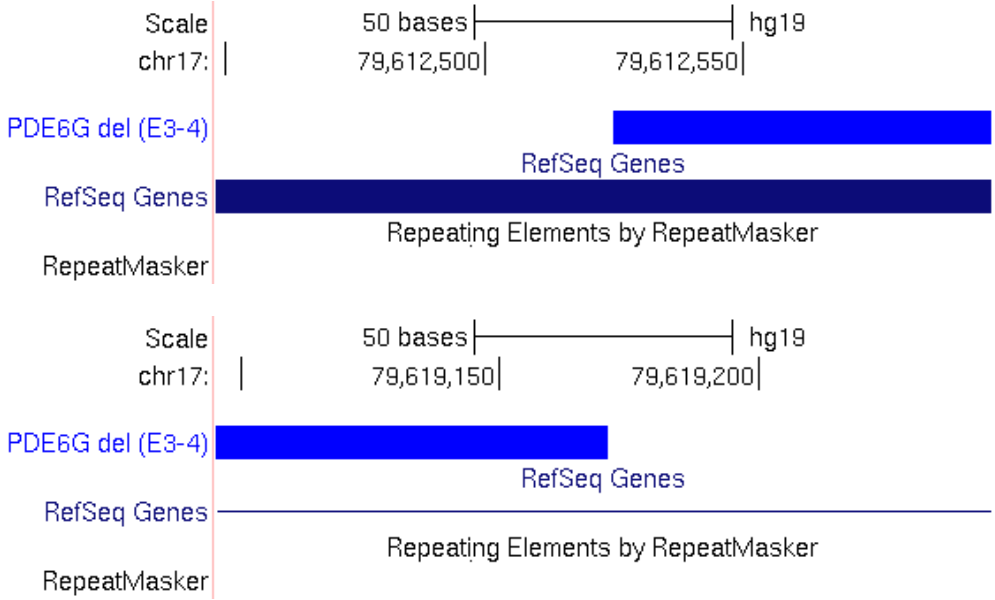


AluSg

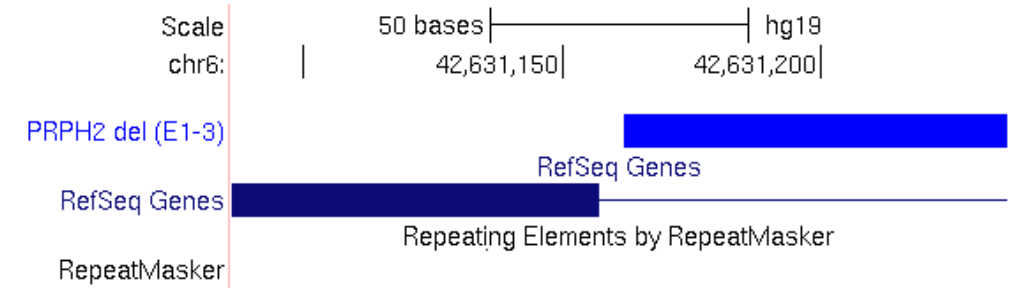


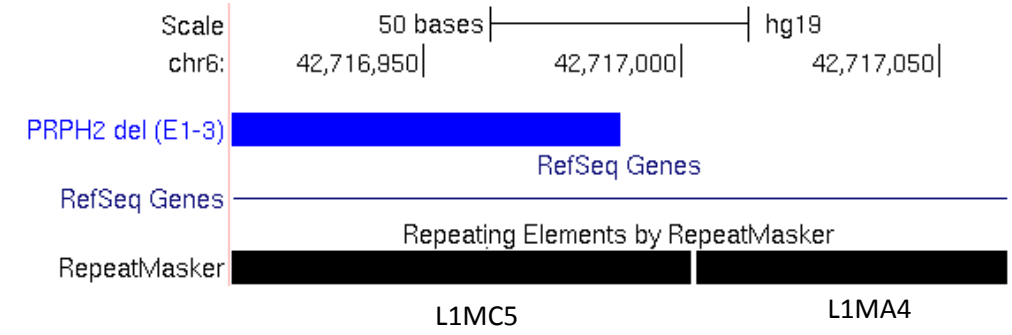
L1MA9

***PDE6G* deletion (E3-4): chr17: g.79612725_79619171del**

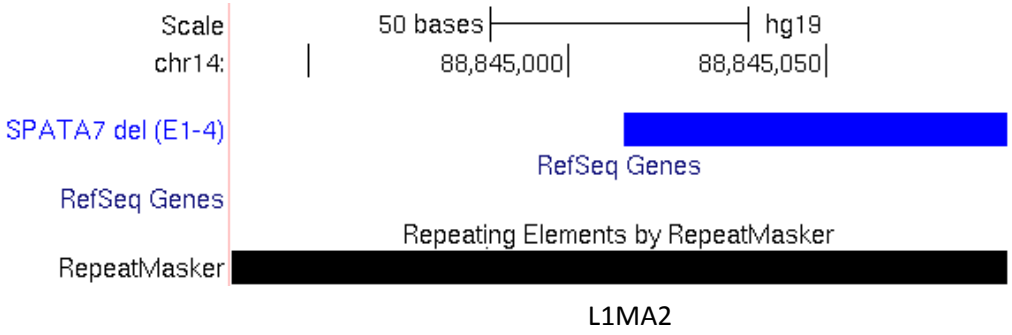


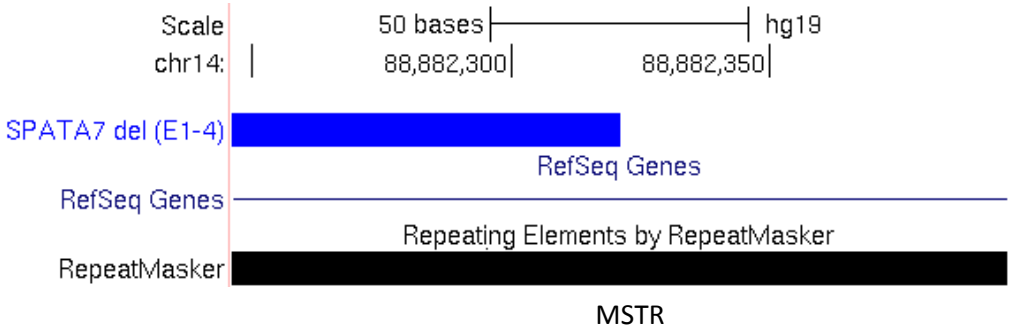
***PRPH2* deletion (E1-3): chr6: g.42631162_42716988delinsTATTTTT**



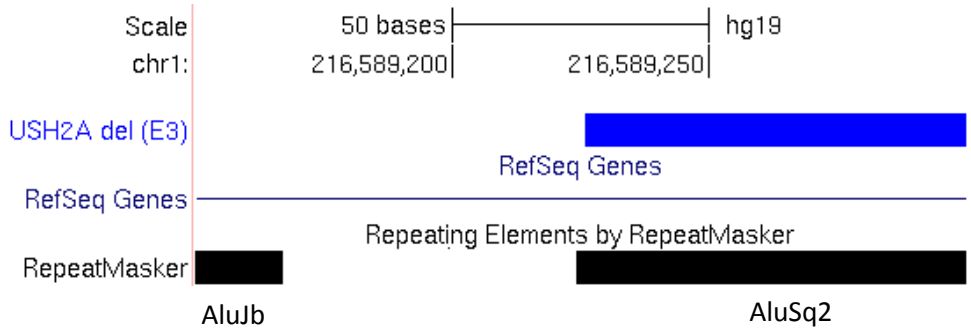


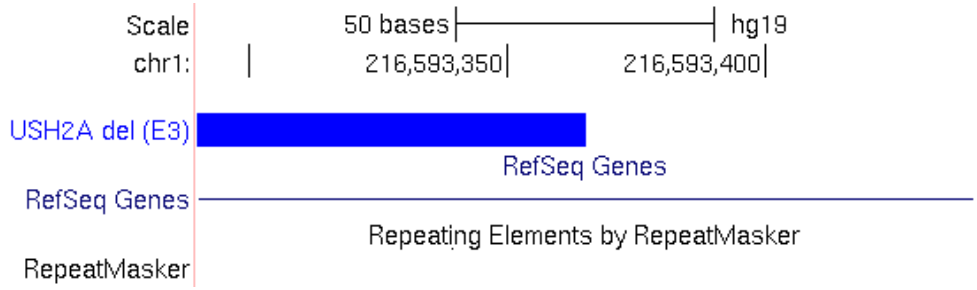
***SPATA*7 deletion (E1-4): chr14: g.88845011_88882321del**



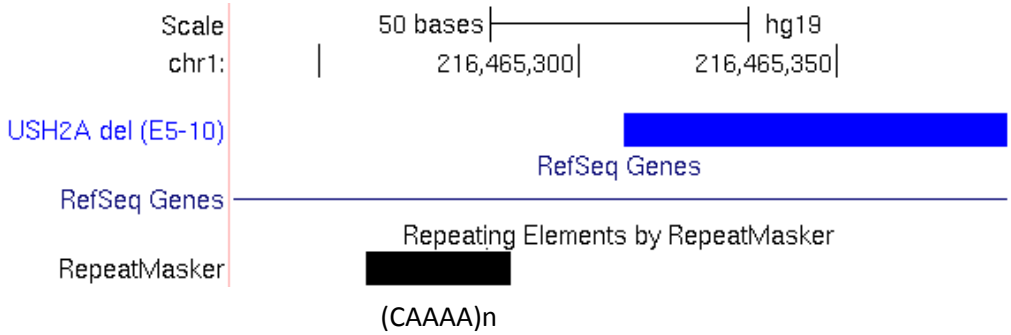


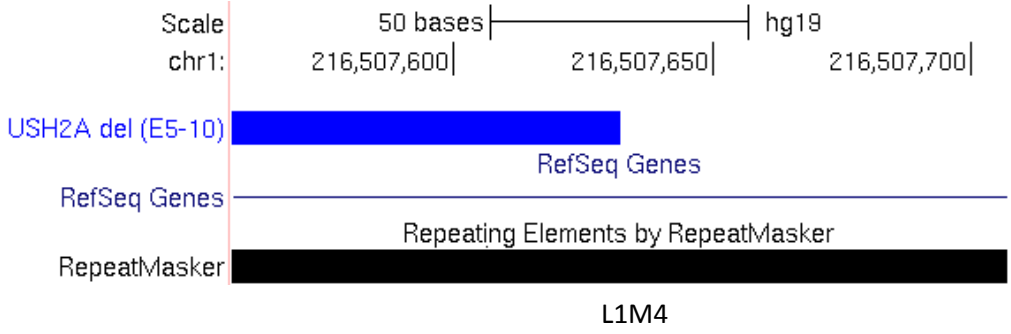
***USH2A* deletion (E3): chr1: g.216589226_216593365del**



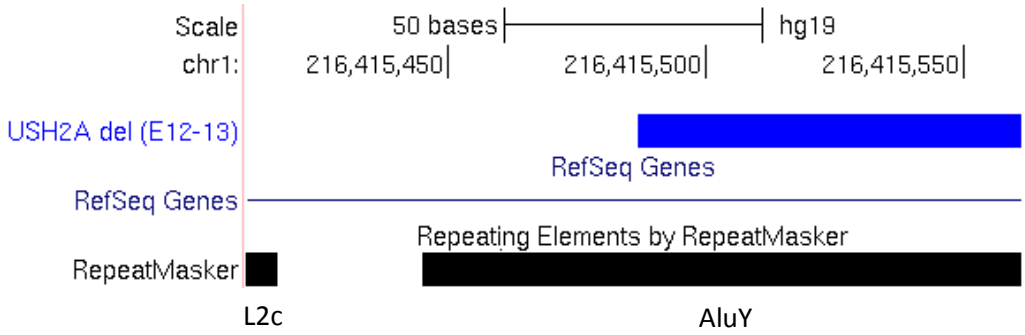


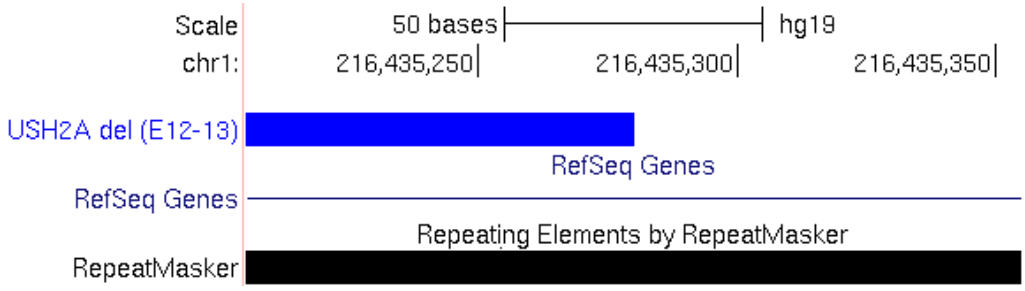
***USH2A* deletion (E5-10): chr1: g.216465309_216507632del**





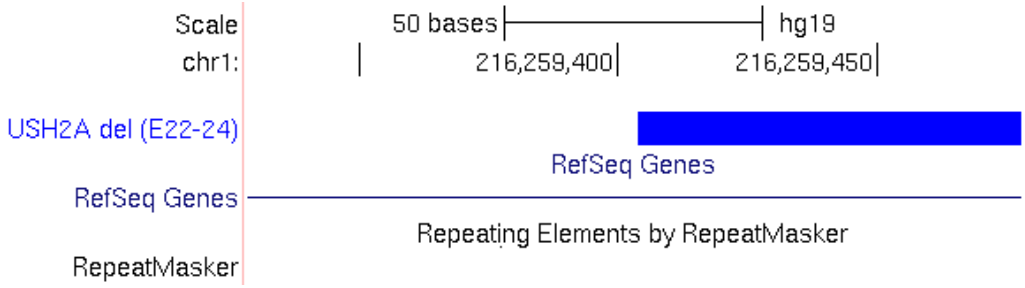
***USH2A* deletion (E12-13): chr1: g.216415487_216435280del**

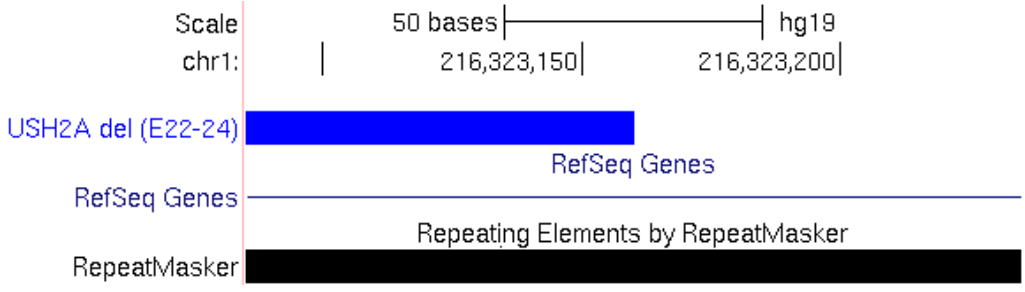




L1MA5

***USH2A* deletion (E22-24): chr1: g.216259404_216323160del**





HERV-int

***USH2A* deletion (E44): chr1: g.216040210_216042178del**

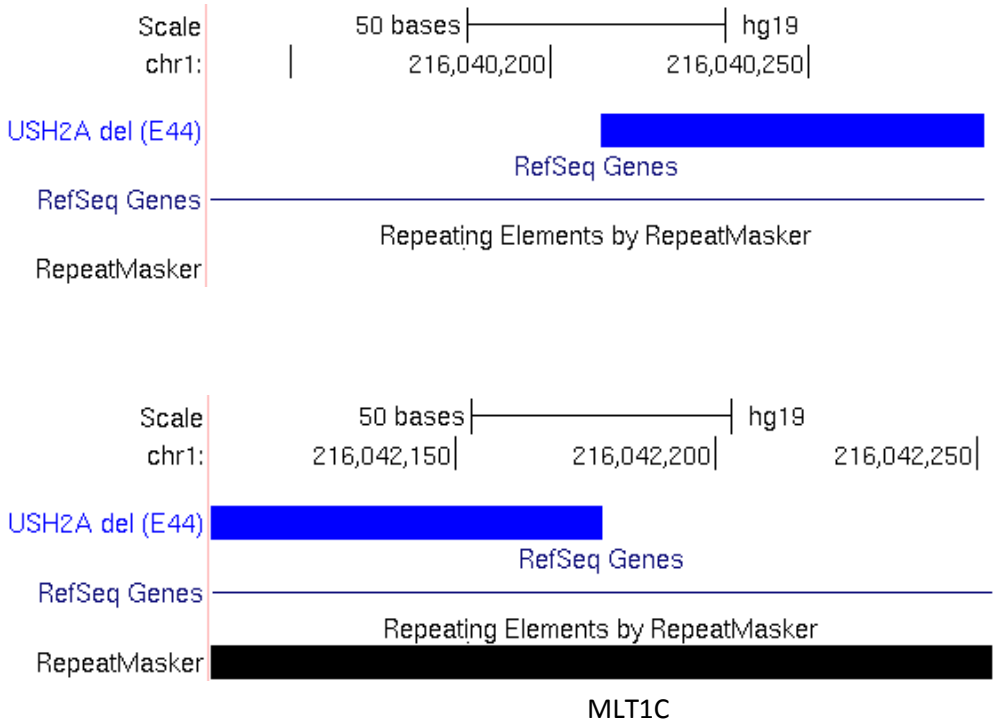


Figure S6. Visualization of repetitive elements at CNV breakpoints.

For every CNV that has been delineated at nucleotide level both breakpoint regions are shown in the UCSC genome browser displaying the RepeatMasker track.

Sequence motifs

***BEST1* deletion (E1-2): chr11: g.61711373_61719810del**

5'BP region

- Consensus SAR 4: 4
- Murine parvovirus recombination hotspot: 2
- Vaccinia topoisomerase I consensus: 1

3'BP region

- Deletion hotspot consensus: 1
- DNA polymerase arrest site: 1
- DNA polymerase a frameshift hotspot 1: 1
- DNA polymerase a/b frameshift hotspot 2: 1
- Heptamer recombination signal: 1
- Ig heavy chain class switch repeat 5: 1
- Murine parvovirus recombination hotspot: 1
- Vaccinia topoisomerase I consensus: 1

***KCNV2* deletion 1 (E2): chr9: g.2723737_2766822del**

5'BP region

- Deletion hotspot consensus: 1
- Murine parvovirus recombination hotspot: 2

3'BP region

- Deletion hotspot consensus: 1
- Murine parvovirus recombination hotspot: 1
- Vaccinia topoisomerase I consensus: 1

***KCNV2* deletion 2 (E2): chr9: g.2724293_2730624del**

5'BP region

- Consensus SAR 2: 2
- Consensus SAR 4: 2
- Deletion hotspot consensus: 1

3’BP region

- Deletion hotspot consensus: 1
- DNA polymerase arrest site: 1
- Vaccinia topoisomerase I consensus: 1

***PCDH15* deletion (E1): chr10: g.56478660_57777934del**

5’BP region

- Consensus SAR 3: 1
- Deletion hotspot consensus: 2
- DNA polymerase arrest site: 1
- Ig heavy chain class switch repeat 3: 1

3’BP region

- Consensus SAR 2: 2
- Consensus SAR 3: 2
- Consensus SAR 4: 3
- Deletion hotspot consensus: 2

***PDE6G* deletion (E3-4): chr17: g.79612725_79619171del**

5’BP region

- Deletion hotspot consensus: 4
- DNA polymerase arrest site: 2
- DNA polymerase b frameshift hotspot 1: 2
- Ig heavy chain class switch repeat 4: 1
- Translin-binding site 2: 1
- Vaccinia topoisomerase I consensus: 1

3’BP region

- DNA polymerase arrest site: 2

***PRPH2* deletion (E1-3): chr6: g.42631162_42716988delinsTATTTTT**

5’BP region

- Consensus SAR 3: 2

- Deletion hotspot consensus: 2
- DNA polymerase arrest site: 2
- Ig heavy chain class switch repeat 5: 1
- Vaccinia topoisomerase I consensus: 2

3'BP region

- Deletion hotspot consensus: 4
- DNA polymerase arrest site: 1
- DNA polymerase b frameshift hotspot 1: 1
- Murine parvovirus recombination hotspot: 1
- Vaccinia topoisomerase I consensus: 1

***SPATA*7 deletion (E1-4): chr14: g.88845011_88882321del**

5'BP region

- DNA polymerase arrest site: 1
- Murine parvovirus recombination hotspot: 2
- Vaccinia topoisomerase I consensus: 4

3'BP region

- Ig heavy chain class switch repeat 3: 1
- Ig heavy chain class switch repeat 4: 1
- Vaccinia topoisomerase I consensus: 2

***USH2A* deletion (E3): chr1: g.216589226_216593365del**

5'BP region

- Consensus SAR 3: 1
- Deletion hotspot consensus: 2
- DNA polymerase arrest site: 1
- Human minisatellites conserved sequence/X-like element: 1
- Ig heavy chain class switch repeat 5: 1
- Murine parvovirus recombination hotspot: 1
- Vaccinia topoisomerase I consensus: 1

3'BP region

- Consensus SAR 2: 1
- Consensus SAR 3: 5

- Consensus SAR 4: 2
- Murine parvovirus recombination hotspot: 1
- Vaccinia topoisomerase I consensus: 1

***USH2A* deletion (E5-10): chr1: g.216465309_216507632del**

5'BP region

- Consensus SAR 3: 1
- Consensus SAR 4: 1
- DNA polymerase a/b frameshift hotspot 2: 1
- Murine parvovirus recombination hotspot: 4
- Vaccinia topoisomerase I consensus: 1

3'BP region

- Translin-binding site 2: 1
- Vaccinia topoisomerase I consensus: 1

***USH2A* deletion (E12-13): chr1: g.216415487_216435280del**

5'BP region

- Consensus SAR 2: 6
- Consensus SAR 3: 1
- Consensus SAR 4: 11
- Deletion hotspot consensus: 1
- DNA polymerase arrest site: 1
- DNA polymerase a/b frameshift hotspot 1: 1
- DNA polymerase a/b frameshift hotspot 2: 1
- Ig heavy chain class switch repeat 3: 1
- Ig heavy chain class switch repeat 4: 1
- Murine MHC recombination hotspot: 1

3'BP region

- Deletion hotspot consensus: 2

***USH2A* deletion (E22-24): chr1: g.216259404_216323160del**

5'BP region

- Deletion hotspot consensus: 1

- DNA polymerase b frameshift hotspot 1: 1
- Vaccinia topoisomerase I consensus: 1

3'BP region

- Deletion hotspot consensus: 1
- DNA polymerase b frameshift hotspot 1: 2
- Ig heavy chain class switch repeat 3: 2
- Ig heavy chain class switch repeat 4: 1

***USH2A* deletion (E44): chr1: g.216040210_216042178del**

5'BP region

- Deletion hotspot consensus: 1
- DNA polymerase a frameshift hotspot 1: 1
- DNA polymerase b frameshift hotspot 1: 1
- Ig heavy chain class switch repeat 2: 1
- Ig heavy chain class switch repeat 4: 1
- Vaccinia topoisomerase I consensus: 1

3'BP region

- DNA polymerase arrest site: 1
- DNA polymerase a/b frameshift hotspot 2: 1
- Ig heavy chain class switch repeat 1: 1
- Ig heavy chain class switch repeat 2: 1
- Vaccinia topoisomerase I consensus: 3

Figure S7. Overview of identified sequence motifs at CNV breakpoints.

For every CNV that has been delineated at nucleotide level the presence of 40 previously described sequence motifs (Abeysinghe et al., Hum. Mutat., 2003) has been assessed in both breakpoint regions using Fuzznuc (Rice et al., Trends Genet., 2000).