Whole genome sequencing reveals complex chromosome rearrangement disrupting *NIPBL* in infant with Cornelia de Lange syndrome

Morasha Plesser Duvdevani,[1,*] Maria Pettersson,[2,*] Jesper Eisfeldt,[2,3] Ortal Avraham,[1] Judith Dagan,[1] Ayala Frumkin,[1] James R. Lupski,[4,5,6,7] Anna Lindstrand,[2,8] Tamar Harel[1]

1. Department of Genetic and Metabolic Diseases, Hadassah-Hebrew University Medical Center, Jerusalem, Israel 9112001
2. Department of Molecular Medicine and Surgery, Center for Molecular Medicine, Karolinska Institute, Stockholm, Sweden
3. Science for Life Laboratory, Karolinska Institutet Science Park, Solna, Sweden
4. Department of Molecular and Human Genetics, Baylor College of Medicine, Houston, TX, USA
5. Human Genome Sequencing Center, Baylor College of Medicine, Houston, TX, USA
6. Department of Pediatrics, Baylor College of Medicine, Houston, TX, USA
7. Texas Children's Hospital, Baylor College of Medicine, Houston, TX, USA
8. Department of Clinical Genetics, Karolinska University Hospital, Stockholm, Sweden
* These authors contributed equally to this work

Supplemental Files:

Figures:

Figure S1. Breakpoint junction analysis confirms interruption of *NIPBL*

Figure S2. Breakpoint junctions and molecular signatures suggest chromothripsis event as underlying mechanism

Figure S3. Fluorescent in situ hybridization (FISH) of proband and parents.

Figure S4. Paternally inherited chromosomal rearrangement.

Tables:

Table S1. Primers designed to amplify specific breakpoint junctions

Table S2. Single nucleotide polymorphisms (SNPs) and primers tested to determine parental origin of rearrangement allele

Table S3. Chromosomal segments as delineated by breakpoint junctions *(provided separately in xls format)*

Table S4. Paternally inherited duplication: breakpoint junctions

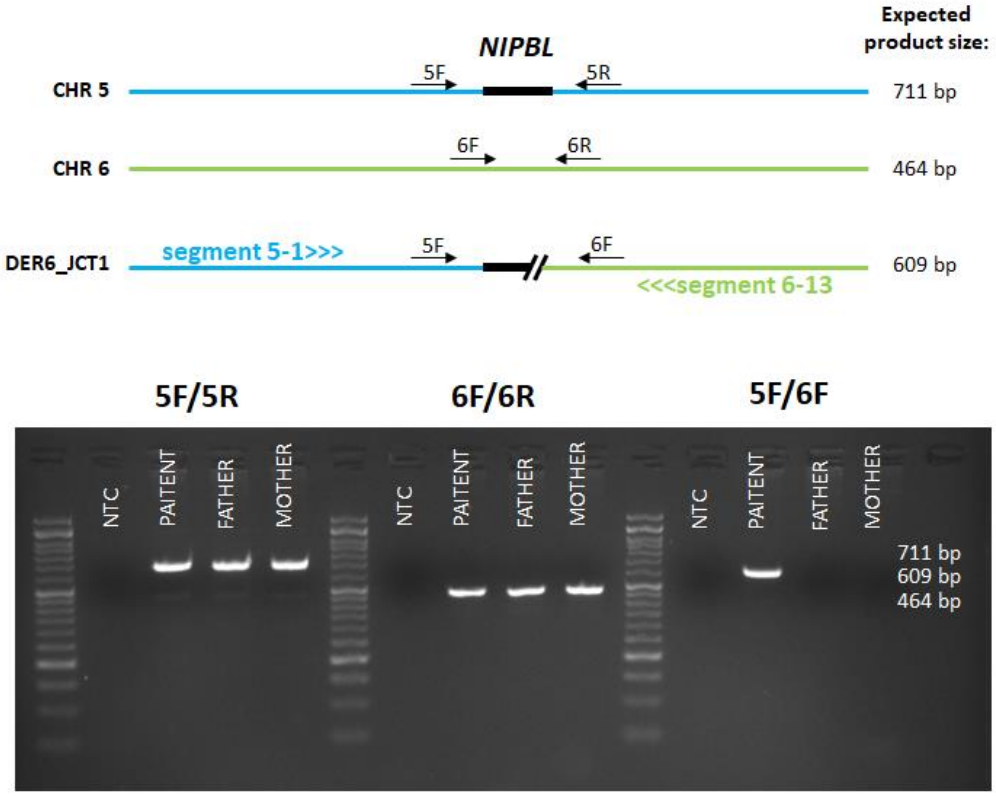
NIPBL
Expected product size:
CHR 5
5F
5R
711 bp
CHR 6
6F
6R
464 bp
DER6_JCT1
segment 5-1>>>
5F
6F
609 bp
<<<segment 6-13
5F/5R
6F/6R
5F/6F
NTC
PAITENT
FATHER
MOTHER
711 bp
609 bp
464 bp

Figure S1. Breakpoint junction analysis confirms interruption of *NIPBL*. Primers were designed to amplify the wild-type alleles on chromosome 5 (primers 5F/5R) and chromosome 6 (primers 6F/6R). The breakpoint junction can be amplified by primers 5F and 6F, since the segment of chromosome 6 is in reverse orientation. The breakpoint junction is only amplified in the proband, confirming that the complex rearrangement occurred *de novo*.

Figure S2. Breakpoint junctions and molecular signatures suggest chromothripsis as underlying mechanism

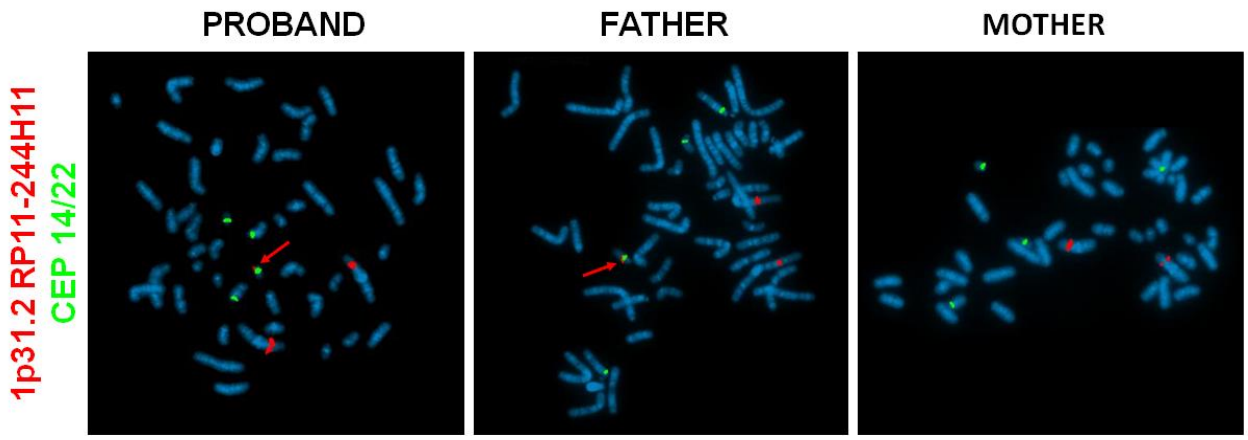

Figure S3. Fluorescent in situ hybridization (FISH) of proband and parents. Hybridization with probes targeting centromeres 14 and 22 (CEP 14/22, green) and 1p31.2 (red) confirmed localization of the duplication to chromosome 22 (red arrows) in the proband and father. The mother's sample was used as a control.

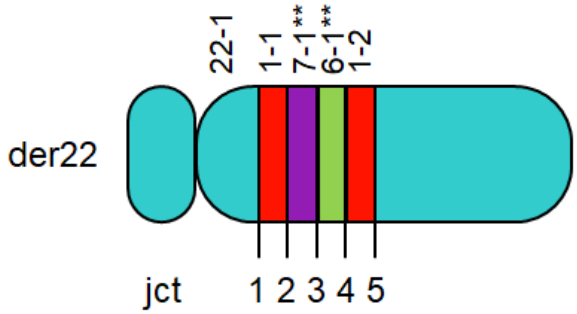

Figure S4. Paternally inherited chromosomal rearrangement. The rearrangement involves a ~56kb region originating from chromosome 1 (segments 1-1, 1-2), with interspersed segments from chromosomes 6 (segment 6-1**) and 7 (segment 7-1**). Double asterisks (**) are used to differentiate these segments from those of the main chromothriptic event (Figure S2), when the same chromosomes are involved.

Table S1. Primers designed to amplify specific breakpoint junctions

Primer name	Forward primer	Primer Name	Reverse primer	Size (bp)
CHR5_control_F (5F)	TTCTGCCTGTAACCATGGAA	CHR5_control_R (5R)	CCCCAACCTATCCATCCTCT	711
CHR6_control_F (6F)	GGGAATTTTCTGCAAAACCA	CHR6_control_R (6R)	GGCTGTCAACACAGCAGAGA	464
DER6_JCT1_NIPBL_F	CHR5_control_F (5F)	DER6_JCT1_NIPBL_R	CHR6_control_F (6F)	609
DER5_JCT7-8_F	CCAGCACAAGATAACCAGCC	DER5_JCT7-8_R	AGCAAGAGTAGCATGGAGGT	685
PAT_DUP_A_F	CAGATTCAGACACCACATTTTGA	PAT_DUP_A_R	AAAAATGAAAAGGACAGATCAAGT	563
PAT_DUP_B_F	CCCAGGAAAAGCAGATGAGA	PAT_DUP_B _R	TCAGGGATAATAAAAGTTCAAACAGA	564

Table S2. Single nucleotide polymorphisms (SNPs) and primers tested to determine parental origin of rearrangement allele

Primer name	Forward primer	Reverse primer	Size (bp)
SNP_RS1408258	GCCCAGCTCATAGCAATAAAA	ACAGCAAGTACTGGAGCACA	397
SNP_RS73718756	GCACAAGGCTGTGAACAGAA	AGCAAGAGTAGCATGGAGGTG	825
SNP_RS1398412976	TGCACCACACCGAGTTTTTA	TGCAGTTCCAAAGTCGCTTA	348

Table S3. Chromosomal segments as delineated by breakpoint junctions *(provided separately in xls format)*

Table S4. Paternally inherited duplication: breakpoint junctions

Breakpoint junction	Genomic coordinate (prox end)	Genomic coordinate (distal end)	BL/INS/ MH	Size (bp)	Source
Der22_jct1	22:NA	1:69119683	NA	-	NR
Der22_jct2	1:69119765	7:38494198	INS	57	Chr6: 166661181-166661157
Der22_jct3	7:38494277	6:166661187	INS	82	NR
Der22_jct4	6:166668890	1:69063596	BL	-	NR
Der22_jct5	1:69119677	22:NA	INS	-	NR