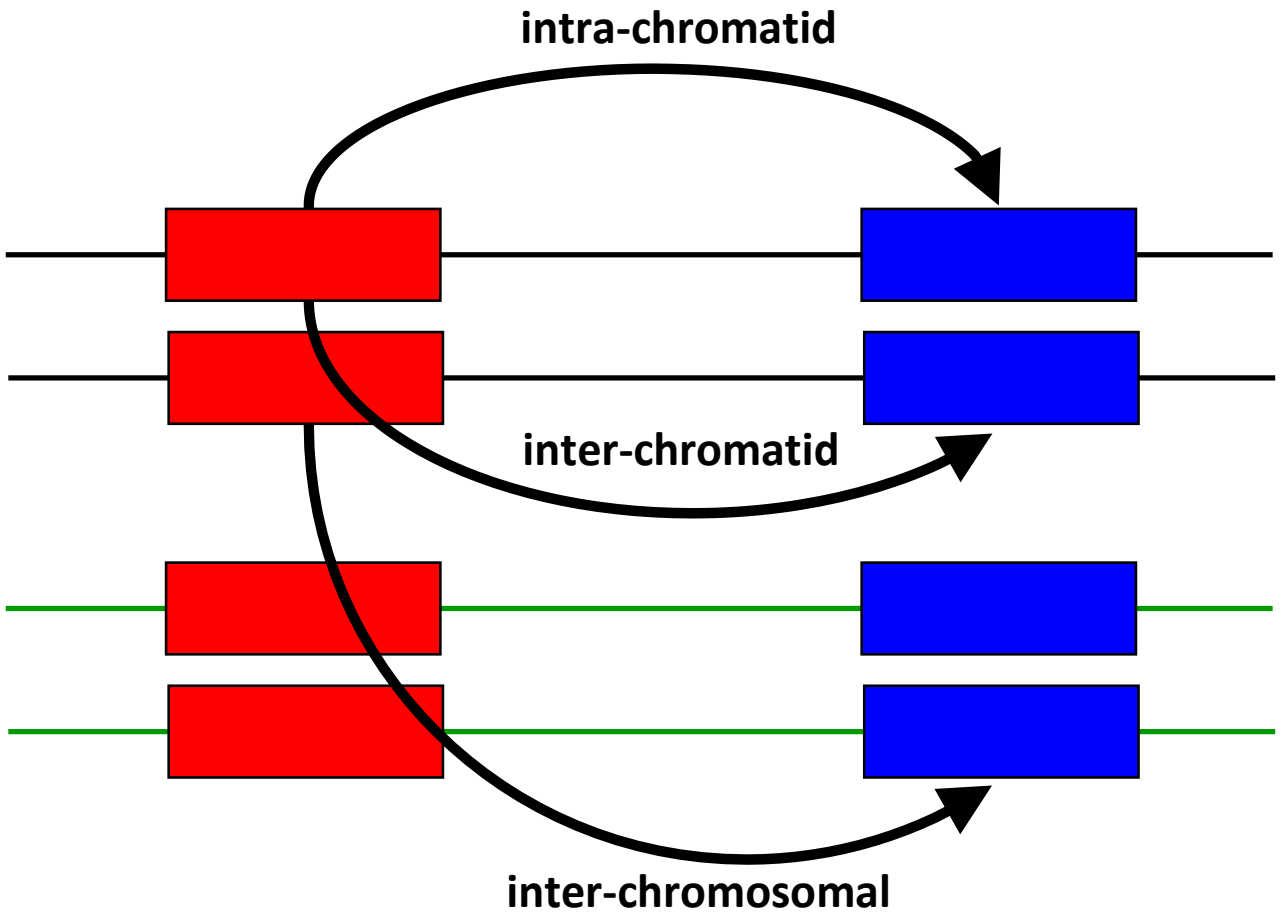
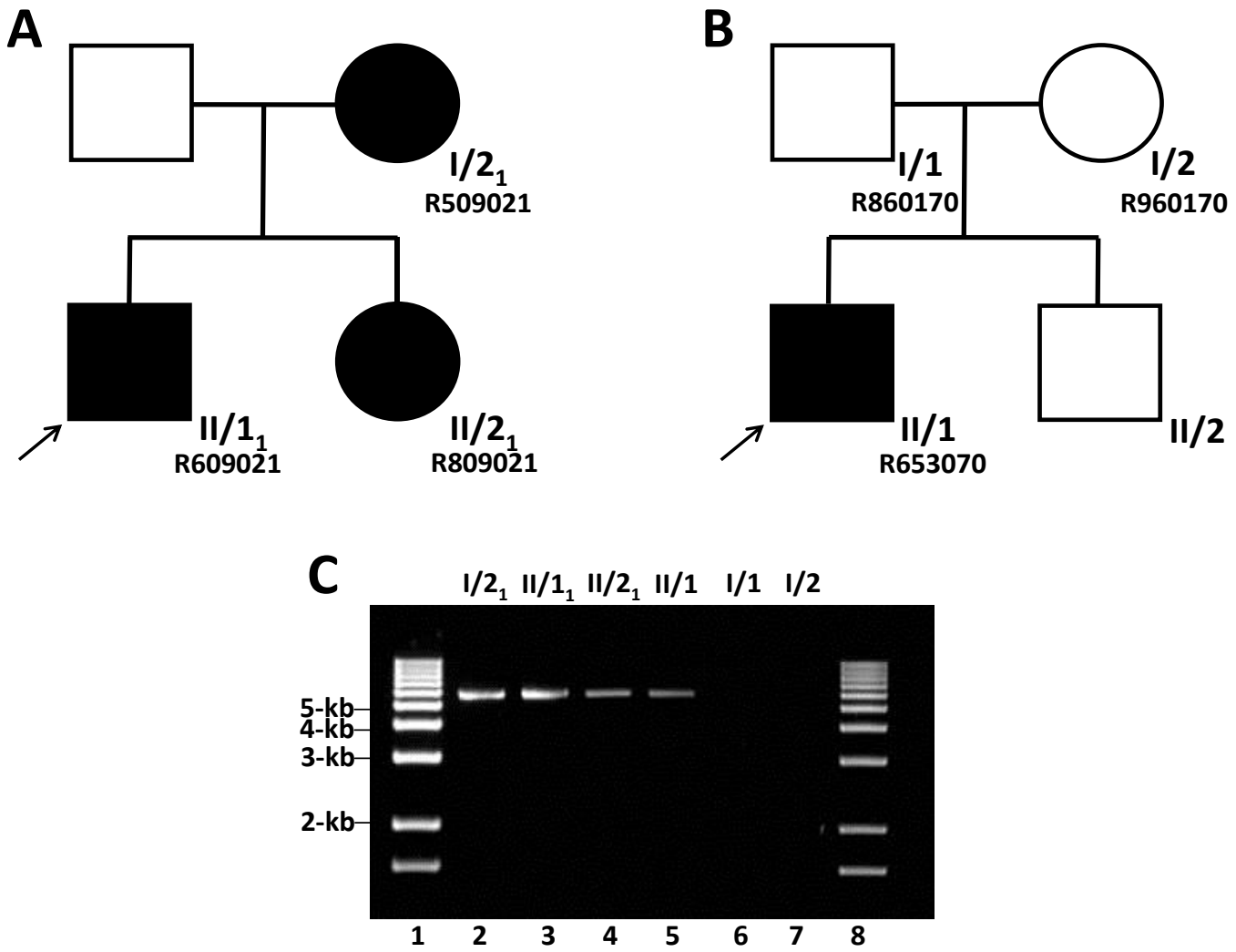


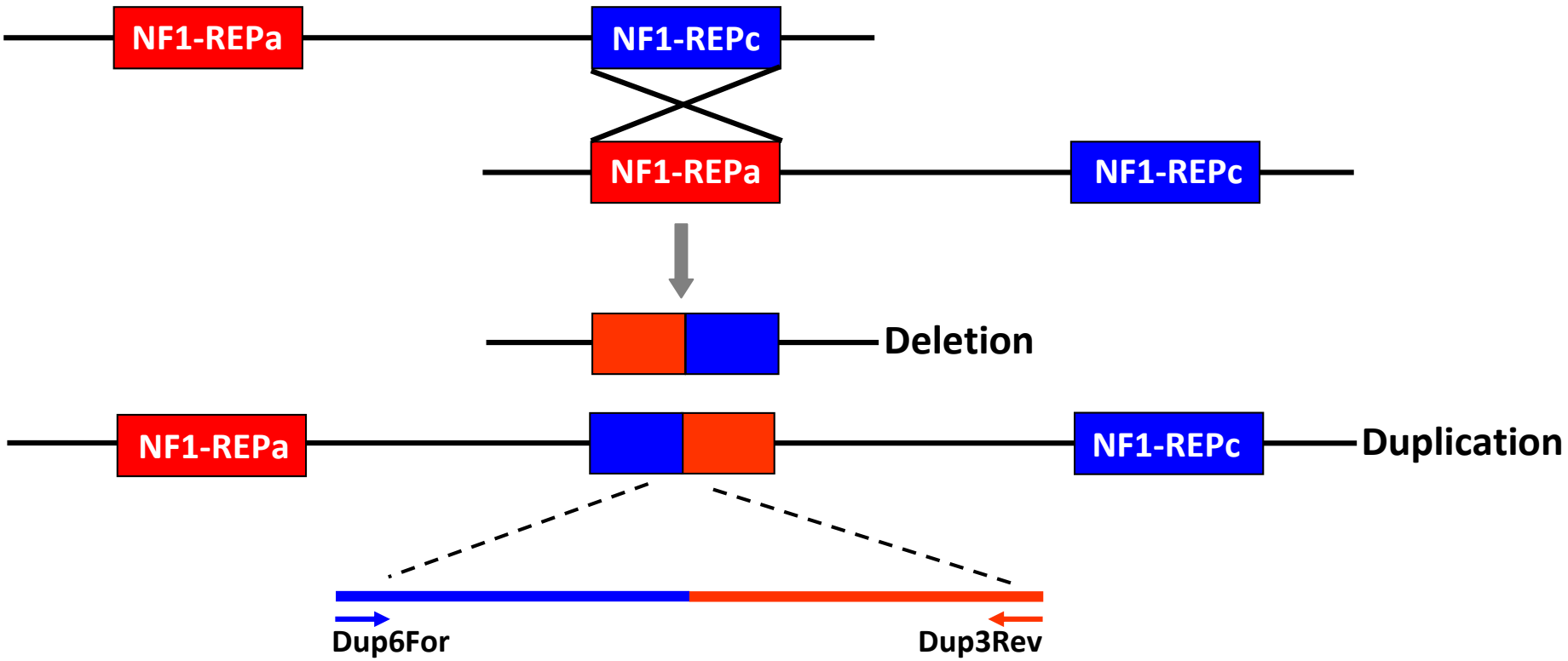
Supp. Figure S1. Different possible types of NAHR between NF1-REPa (red) and NF-REPc (blue) which represent nonallelic but highly similar sequences. The NAHR events are indicated by black arrows. The horizontal black lines represent sister chromatids of one chromosome 17 and the green lines the sister chromatids of the other chromosome 17.


A
I/2$_1$
R509021
II/1$_1$
R609021
II/2$_1$
R809021
B
I/1
R860170
I/2
R960170
II/1
R653070
II/2
C
I/2$_1$ II/1$_1$ II/2$_1$ II/1 I/1 I/2
5-kb
4-kb
3-kb
2-kb
1 2 3 4 5 6 7 8

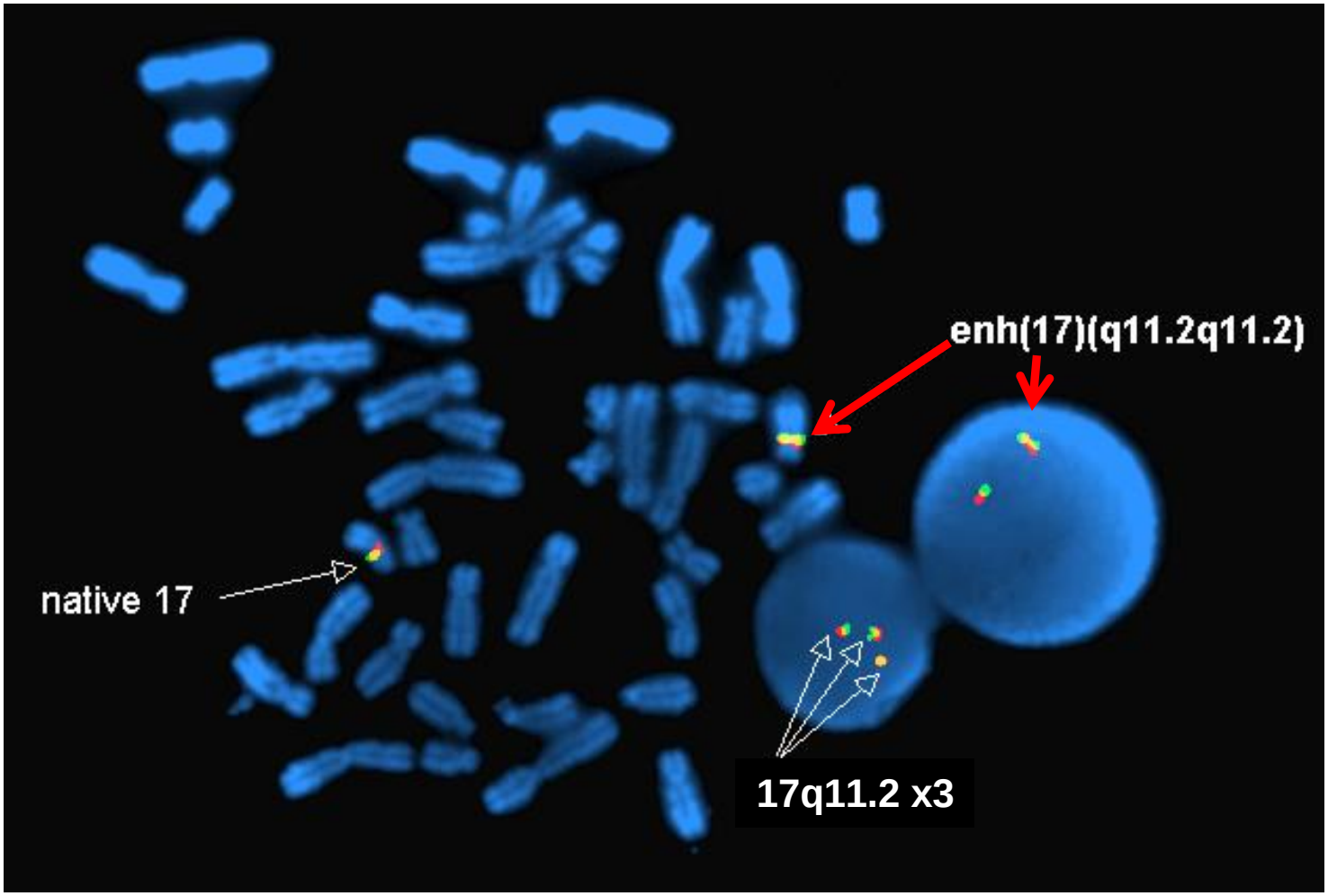
Supp. Figure S2. The large *NF1* duplications were detected by breakpoint-spanning PCR in the index patients R609021 and R653070. (**A**) Patient 609021 is a familial case since his mother and sister also harboured the duplication. (**B**) By contrast, patient R653070 most likely represents a sporadic patient whose parents did not harbour the deletion in their germ lines as ascertained by MLPA and breakpoint-spanning PCR performed on blood cell DNA. Also his brother (II/2) did not harbour the duplication as determined by MLPA. (**C**) Separation of the PCR-amplified 5.8-kb breakpoint-spanning fragments on an agarose gel. The breakpoint-spanning PCR was performed with a forward primer located in NF1-REPc and a reverse primer located in NF1-REPa. Size markers are indicated in lanes 1 and 8. The PCR products amplified from the duplication carriers are visible in lanes 2-5.

Marker	Position of marker	R653070			R960170			R860170	
D17S1873	27457470 - 27457612		137	135	121	137		135	119
D17S975	28099987 - 28100245		256	260	252	256		260	260
D17S1532	28683996 - 28684165		170	170	174	170		170	164
D17S1307	29473353 - 29473561	210	206	210	210	206		210	210
NF1.PCR3	29589841 - 29590220	388	374	390	388	374		390	386
D17S2237	29617725 - 29618128	400	392	404	400	392		404	400
IVS27TG24.8	29640672 - 29640901	272	278	272	272	278		272	272
IVS27AC28.4	29644294 - 29644500	205	203	211	205	203		211	209
D17S1166	29649022 - 29649173	142	162	142	142	162		142	142
IVS38GT53.0	29668522 - 29668693	186	176	186	186	176		186	186
3NF1	29919355 - 29919599	250	238	236	250	238		236	250
D17S1800	29936829 - 29937098	278	274	272	278	274		272	284
D17S1880	31013893 - 31014084		194	182	194	188		182	174
D17S1293	32560166 - 32560449		278	274	278	258		274	266
D17S907	33866310 - 33866639		286	326	286	318		326	290
D17S1788	36086054 - 36086207		154	158	154	156		158	156
D17S1809	62697450 - 62697596		134	140	134	144		140	142

Supp. Figure S3. Microsatellite marker analysis performed in order to determine the origin of the *NF1* duplication detected in patient R653070. The size of the respective marker alleles is indicated in bp. The coloured rectangles represent the inferred and estimated haplotypes. R960170 is the mother of patient R653070 whereas proband R860170 is his father. The grey rectangle demarcates the markers located within the *NF1* gene region. These markers are deleted on one chromosome 17 in patients with type-1 *NF1* deletions. In patient R653070, a duplication of these markers was observed. Comparing the haplotypes of the family members, we conclude that the *NF1* duplication occurred in the maternal germline by inter-chromosomal or inter-chromatidal NAHR.

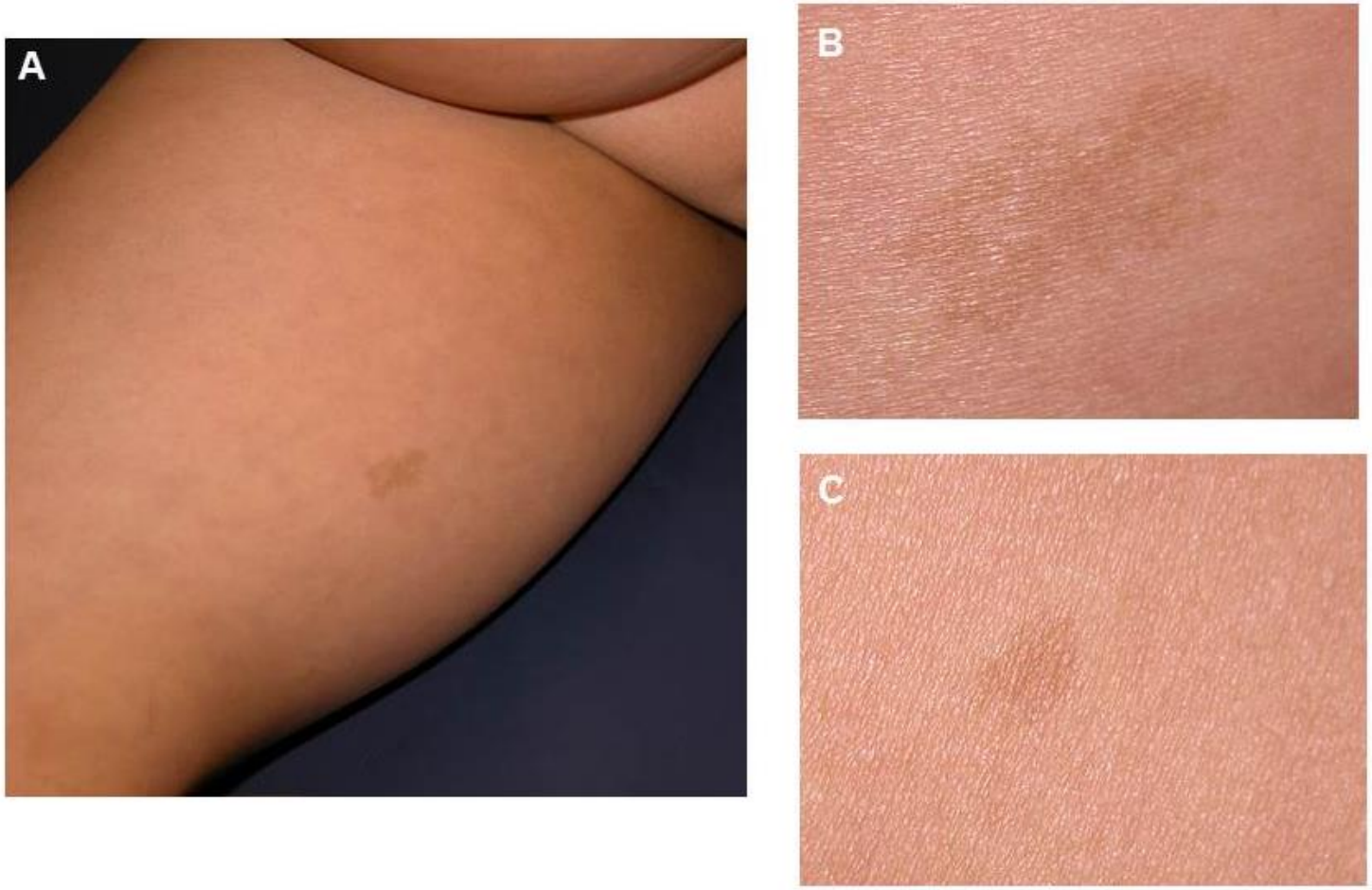


Supp. Figure S4. NAHR between the low-copy repeats NF1-REPa and NF1-REPc, either by an inter-chromosomal or an inter-chromatidal mechanism, gives rise to an *NF1* deletion and its reciprocal *NF1* duplication. The duplication breakpoint-spanning PCR fragment of 5.8-kb is amplified with primers Dup6For and Dup3rev whose relative locations are indicated.

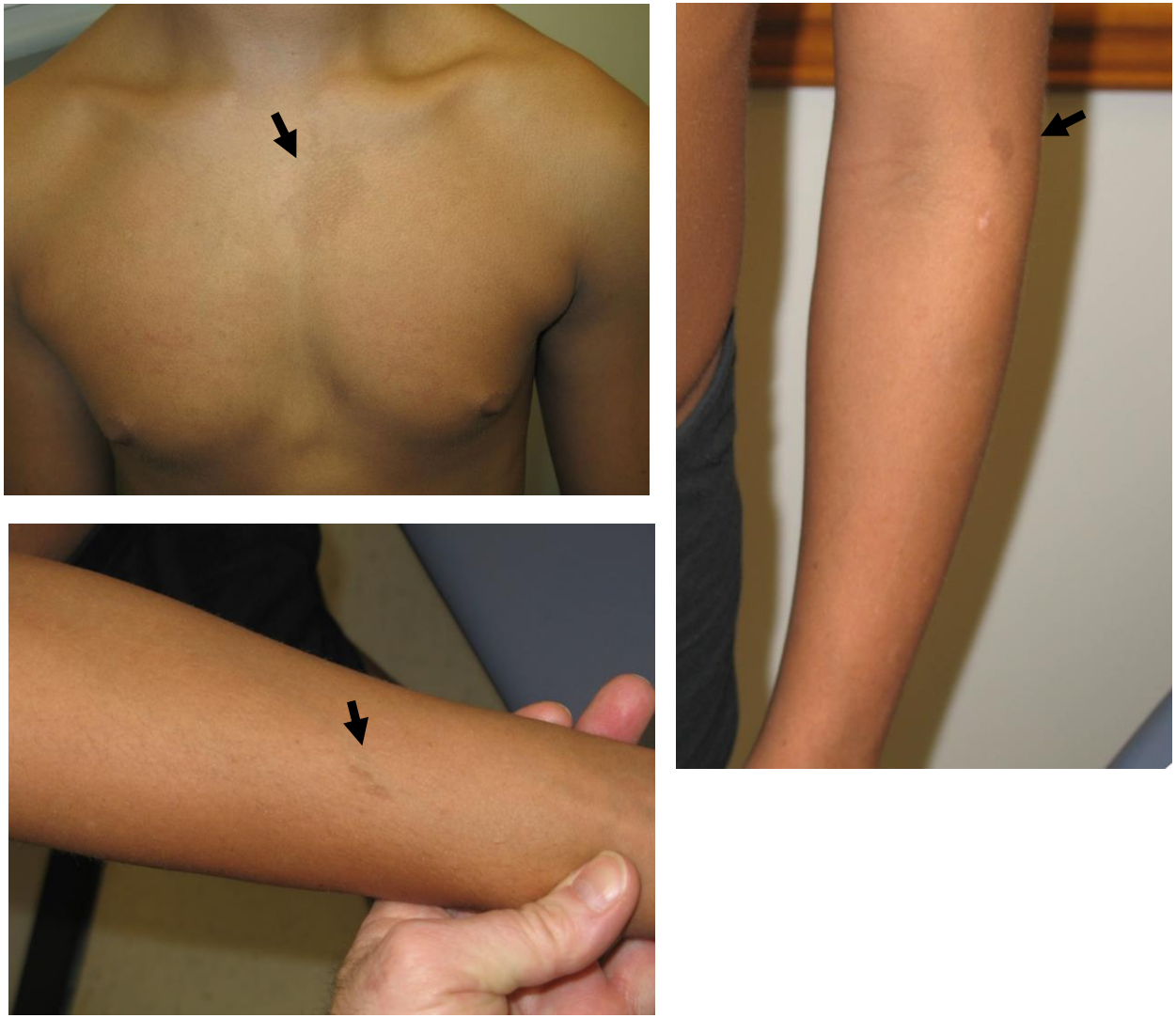


Supp. Figure S5. FISH confirmed the in-tandem *NF1* duplication in patient R653070. FISH was performed using probes RP5-1002G3 (visible as red fluorescent signals) and RP5-926B9 (green). FISH probe RP5-1002G3 is located in the telomeric region of the *NF1* gene, whereas FISH probe RP5-926B9 spans the centromeric part of the *NF1* gene. Thus, two red/green fusion FISH signals are indicative of one copy of the *NF1*

gene. However, in the patient, three red/green fusion FISH signals were detected, examples of which are shown in the lower interphase nucleus (three arrows). On the metaphase chromosomes and in many other interphase nuclei, two of the red/green fusion FISH signals were adjacent to one another giving rise to an enhanced (enh) FISH signal (marked by red arrows).



Supp. Figure S6. Café au lait spots observed in patient R609021. The café au lait spot located on the thigh seen in (A) is enlarged in (B) in order to show its irregular borders and the nonhomogeneous pigmentation. The café au lait spot in (C) is located on the abdomen.



Supp. Figure S7. Café au lait spots observed in patient R653070.

Supp. Table S1. Results of the MLPA analysis using the P122-C2 kit (MRC Holland, The Netherlands)

Probe designation	Probe position on chromosome 17 (hg19)	P1 R609021	P2 R509021	P3 R809021	P4 R653070
TRAF4 9176-L9350	27,074,291-27,074,314	0.99	0.99	0.95	0.92
TRAF4 8620-L8632	27,075,052-27,075,075	1.07	1.15	1.08	1.55
SSH2 9635-L9920	27,963,580-27,963,603	1.05	1	1.01	0.94
SSH2 9634-L9919	28,022,495-28,022,518	1.05	1.09	1.09	1.22
BLMH 9627-L9912	28,599,612-28,599,635	1.08	1.08	1.09	1.14
BLMH 9626-L9911	28,618,478-28,618,501	1	0.97	0.99	0.9
CPD 9628-L9913	28,770,910-28,770,933	1.06	1.02	1.01	0.94
CPD 9629-L9914	28,789,420-28,789,443	1.02	0.98	1	0.94
SUZ12p 11798-L12590	29,058,391-29,058,414	**1.38**	**1.38**	**1.41**	**1.46**
SUZ12p 11801-L12592	29,085,145-29,085,168	**1.45**	**1.43**	**1.42**	**1.54**
CRLF3 3780-L3289	29,124,380-29,124,403	**1.38**	**1.45**	**1.42**	**1.42**
ATAD5 3781-L3290	29,162,044-29,162,067	**1.45**	**1.54**	**1.48**	**1.73**
CENTA2 3782-L3291	29,253,873-29,253,896	**1.52**	**1.55**	**1.53**	**1.88**
RNF135 3783-L3292	29,311,688-29,311,711	**1.45**	**1.47**	**1.47**	**1.63**
NF1 Ex. 1 2491-L1922	29,421,598-29,421,621	**1.41**	**1.4**	**1.44**	**1.29**
NF1 Ex. 17 2507-L1938	29,552,202-29,552,225	**1.44**	**1.49**	**1.46**	**1.84**
NF1 Ex. 30 2512-L1943	29,576,023-29,576,046	**1.33**	**1.29**	**1.34**	**1.35**
NF1 Ex. 49 2525-L1956	29,676,152-29,676,175	**1.58**	**1.54**	**1.52**	**1.43**
NF1 Ex. 57 5220-L3309	29,687,576-29,687,599	**1.39**	**1.39**	**1.36**	**1.33**
UTP6 3785-L3294	30,202,348-30,202,371	**1.5**	**1.52**	**1.49**	**1.72**
SUZ12-10 3786-L3295	30,315,410-30,315,433	**1.47**	**1.48**	**1.45**	**1.53**
LRRC37B 3787-L3296	30,348,569-30,348,592	**1.47**	**1.56**	**1.48**	**1.72**
ZNF207 9637-L9949	30,693,753-30,693,776	1.1	1.07	1.06	1.08
PSMD11 9632-L9917	30,773,979-30,774,002	1.01	0.99	0.99	0.9
PSMD11 9633-L9918	30,796,071-30,796,094	1.08	1.13	1.06	1.22
MYO1D 9631-L9916	31,094,710-31,094,733	1.04	1.06	1.04	1.25
MYO1D 9630-L9915	31,107,652-31,107,675	1.1	1.06	1.1	1.17

Supp. Table S2. Sequences and locations of the primers used to amplify the 5811-bp *NF1* duplication breakpoint-spanning PCR product

Primer	5'→3' sequence	Position of the primer in [a]	
		NF1-REPa	NF1-REPc
Dup6for	AATGAAGACAAGGGCTCACTGCATC		30,408,259-30,408,283
Dup3rev	AAGGGGCTCGGAGAAGTTAAGG	28,997,938-28,997,959	

a: chromosomal positions are indicated according to human genome assembly hg19

Supp. Table S3. Cycle conditions for the PCR performed with primers Dup6for and Dup3rev in order to amplify the *NF1* duplication breakpoint-spanning fragment

Step	Description	Temperature [°C]	Time [min]
1	initial denaturation	94	10 min
2	denaturation	94	30 sec
3	primer annealing	60	30 sec
4	elongation	68	4 min plus 30 sec
5	final elongation	68	5 min

Steps 2-4 were repeated 35 times.

Supp. Table S4. Primers used to sequence the *NF1*duplication breakpoint-spanning PCR product of 5811-bp amplified with primers Dup6for and Dup3rev

Primer	5'→ 3'sequence	Position[a] of primer on chromosome 17 in		Annealing temperature
		NF1-REPa	NF1-REPc	
2290 hin	TCAACCTCCCAGGCTCCCGAA	28,992,825-28,992,845	30,408,907-30,408,951	62°C
2314rev	TTTGCACGTGTGACCTTCCACA	28,996,756-28,996,777	30,412,872-30,412,893	62°C
Typ1_1rev	CCAGGCCCCCTCTTCCTAGA	28,996,360-28,996,379	30,412,476-30,412,495	62°C
Typ1_2for	AGGCTGGTCTCCAACTCCTT	28,993,753-28,993,772	30,409,862-30,409,881	58°C
Typ1_5for	ATCTGCCTTCCAAGAACTGC	28,996,422-28,996,441	30,412,538-30,412,557	58°C
Typ1_6rev	TCTAGGTTTTCTGAGGTCACACC	28,993,959-28,993,981	30,410,068-30,410,090	58°C
Typ1_7rev	GCAGGAGGATAGTTTGAACCAG	28,992,946-28,992,967	30,409,052-30,409,073	58°C
Typ1_8rev	TTCCCCAGCCGGAGAGAG	28,995,165-28,995,182	30,411,276-30,411,293	60°C
Typ1_9rev	GGGACAGTTGAGGGGGTACT	28,995,515-28,995,534	30,411,630-30,411,649	58°C
Typ1_5rev	AAGTTAAGGGCCAGGCACA	28,993,673-28,993,692	30,409,781-30,409,799	58°C

a: chromosomal positions are indicated according to the human genome assembly hg19

Supp. Table S5. Sequence analysis of the *NF1* duplication breakpoint-spanning PCR products which were amplified from genomic DNA of patients R609021 and R653070

PSV	Position of PSV in		Nucleotides at PSV sites in		PSVs identified with the breakpoint-spanning PCR products in patients	
	NF1-REPc (hg19)	NF1-REPa (hg19)	NF1-REPa	NF1-REPc	R609021[a]	R653070
1	30,408,920-30,408,943	28,992,837-28,992,838	–	CTCAAGTGATCC TCCCACCTCAGC	CTCAAGTGATCCTCC CACCTCAGC	CTCAAGTGATCCTCC CACCTCAGC
2	30,409,076	28,992,970	T	C	C	C
3	30,409,221	28,993,115	A	G	G	G
4	30,409,276	28,993,170	A	G	G	G
5	30,409,478	28,993,372	C	T	T	T
6	30,409,490-30,409,491	28,993,382-28,993,383	–	TC	TC	TC
7	30,409,720	28,993,613	T	G	G	G
8	30,409,728	28,993,621	A	G	G	G
9	30,409,735	28,993,628	T	C	C	C
10	30,409,736	28,993,629	G	A	A	A
11	30,409,738	28,993,631	C	T	T	T
12	30,409,743	28,993,636	C	T		
13	30,409,744	28,993,637	A	G		
14	30,409,841-30,409,842	28,993,733-28,993,734	–	AG	AG	AG
15	30,409,888	28,993,779	T	A	A	A
16	30,409,952	28,993,843	G	T	T	T
17	30,410,094	28,993,986	T	–	–	–
18	30,410,767	28,994,656	C	A	A	C*
19	30,411,033	28,994,921	C	A	A	C*
20	30,411,096	28,994,985	C	–	–	–
21	30,412,077	28,995,962	T	C	C	C
22	30,412,090	28,995,975	T	G	G	G
23	30,412,107	28,995,992	G	A	G*	G*
24	30,412,191	28,996,075	–	A	A	A
25	30,412,617	28,996,501	A	G	A	G
26	30,412,702	28,996,586	C	T	C	T
27	30,412,772	28,996,656	A	G	A	A
28	30,412,799	28,996,683	C	T	C	C
29	30,412,871	28,996,755	A	C	A	A

–: Allele harbouring the deletion of this indel variant
a: The same PSVs as observed in patient R609021 were also detected in his mother and sister who also harbour the *NF1* duplication.

The analysis of paralogous sequence variants (PSVs) indicated the regions of strand exchange between NF1-REPa (red) and NF1-REPc (blue). The regions of strand exchange during nonallelic homologous recombination (NAHR) giving rise to these *NF1* duplications are located within the PRS2 recombination hotspot. This hotspot also harbours the regions of strand exchange of type-1 *NF1* deletions [De Raedt et al., 2006; Bengesser et al., 2014]. PSVs 1-29 indicated in this table are located in a 4-kb region within the NF1-REPs which harbours the PRS2 hotspot. In both patients with type-1 *NF1* duplications (R609021 and R653070), gene conversion was detected in sequences flanking the regions of strand exchange. The corresponding PSVs that we infer have been affected by gene conversion are marked by asterisks.

Supp. Table S6. Location of the regions of strand exchange during NAHR causing type-1 *NF1* deletions and their reciprocal duplications

PSV	Position of PSV in		Nucleotides at PSV sites observed in breakpoint-spanning PCR fragments of patients with type-1 *NF1* deletions and duplications																			
	NF1-REPa	NF1-REPc	DEL 1	DEL 2	DEL 3	DEL 4	DEL 5	DEL 6	DEL 7	DEL 8	DEL 9	DEL 10	DEL 11	DEL 12	DEL 13	DEL 14	DEL 15	DEL 16	DEL 17	DEL 18	DEL 19	DEL 20	DUP R609021	DUP R653070
1	28,992,837-28,992,838	30,408,920-30,408,943																						
2	28,992,970	30,409,076																						
3	28,993,115	30,409,221																						
4	28,993,170	30,409,276																						
5	28,993,372	30,409,478																						
6	28,993,382-28,993,383	30,409,490-30,409,491																						
7	28,993,613	30,409,720																						
8	28,993,621	30,409,728																						
9	28,993,628	30,409,735																						
10	28,993,629	30,409,736																						
11	28,993,631	30,409,738																						
12	28,993,636	30,409,743																						
13	28,993,637	30,409,744																						
14	28,993,733-28,993,734	30,409,841-30,409,842																						
15	28,993,779	30,409,888																						
16	28,993,843	30,409,952																						
17	28,993,986	30,410,094																						
18	28,994,656	30,410,767																						•
19	28,994,921	30,411,033																						•
20	28,994,985	30,411,096																						
21	28,995,962	30,412,077																						
22	28,995,975	30,412,090																						
23	28,995,992	30,412,107																					•	•
24	28,996,075	30,412,191																						
25	28,996,501	30,412,617																						
26	28,996,586	30,412,702																						
27	28,996,656	30,412,772																						
28	28,996,683	30,412,799																						
29	28,996,755	30,412,871																						

PSV positions are indicated according to hg19. Sequence analysis of the breakpoint-spanning PCR products revealed the indicated PSV pattern (blue: NF1-REPc-specific PSVs; red: NF1-REPa-specific PSVs). The regions of strand exchange, indicated by a green horizontal line, are located between PSVs derived from NF1-REPa (red) and NF1-REPc (blue).

PSVs 1-29 indicated in this table are located in a 4-kb region within the NF1-REPs which harbour the PRS2 hotspot. The region of strand exchange identified by the analysis of the 20 type-1 *NF1* deletions has been previously reported [Bengesser et al., 2014]. The regions of strand exchange of the *NF1* duplications in patients R609021 and R653070 are located telomeric to the regions of strand exchange identified in type-1 *NF1* deletions. In both patients with type-1 *NF1* duplications, gene conversion was detected in sequences flanking the regions of strand exchange. The corresponding PSVs that we infer have been affected by gene conversion are marked by black circles.

Supp. Table S7. Summary of all *NF1* duplication carriers reported previously and in this study including the clinical features observed

Patient	Age at diagnosis [a]	Sex	Dup. type	Inheritance	Growth	Neuro-development	Neurology	Dysmorphic features	Hair	Tooth anomalies	Other	Reference
1	13 y	F	1 or 2	unknown	short stature, microcephaly	DD	seizures	not specified	not specified	not specified	subject 1 carried another copy-number variant, a BP1-BP2 15q11.2 deletion proximal to the Prader–Willi/ Angelman syndrome critical region. This deletion may predispose to neuro-cognitive disabilities, developmental delay and seizures	Moles et al. [2012]
2	4 y	M	1 or 2	paternal	macrocephaly	severe DD, ID	seizures	present, high palate and ankyloglossia	not present	not specified	pectus excavatum, adducted thumbs, hypospadias, chronic lung disease	Moles et al. [2012]
3	unknown	F	1 or 2	unknown	normal	normal	unknown	absent	unknown	unknown	healthy father of patient 2	Moles et al. [2012]
4	11 y	M	1, 2 or 3	unknown	failure to thrive	severe DD	seizures	present	not present	not specified	dizziness, mitral valve prolapse, sib with similar phenotype (not tested)	Moles et al. [2012]
5	newborn	M	3	*de novo*	not specified	not specified	polymicro-gyria	bilateral CL/P, left iris coloboma	not specified	not specified		Moles et al. [2012]
6	2 m	M	1 or 2	unknown	not specified	not specified	not specified	present	not specified	not specified		Moles et al. [2012]
7	21 y	M	1 or 2	unknown	normal, small hands and feet	DD, ID	DTR3+	absent	not present	not present	diabetes, hypertension, hypercholesteremia, GERD	Moles et al. [2012]
8	3 y	M	1 or 2	maternal	height >97th PC, weight >95th PC, head circumference 70th PC	DD, speech delay	normal MRI	flat midface, short palpebral fissures, short nose with mildly broad and flattened tip, smallmouth, unilateral lacrimal pit	not present	small lower teeth, widely spaced	irregular linear hypopigmented skin lesion, one faint CALS, strong adult body odour, unilateral microphthalmia	Moles et al. [2012]
9	unknown	M	1 or 2	unknown	not specified	language delay	not specified	flat midface, short palpebral fissures, short nose with mildly broad and flattened tip, congenital unilateral microphthalmia	not specified	not specified	affected mother of patient 8	Moles et al. [2012]
10-17											family of 8 *NF1* duplication carriers, two individuals were not clinically affected	Grisart et al. [2008]
10 (III.3)	37 y	M	1	paternal, father carrier	short stature (10th PC), microcephaly (5th PC)	DD, ID		mild facial dysmorphic features, long midface, malar hypoplasia, nasal deformation	early onset baldness at the age of 14-15 years	dental enamel hypoplasia	brother of patient 11, large testes	Grisart et al. [2008]
11 (III.4)	38 y	M	1	paternal, father carrier	short stature, microcephaly	DD, ID	increased lower limb reflexes	mild facial dysmorphic features, long midface, malar hypoplasia, nasal deformation	premature balding	dental enamel hypoplasia	brother of patient 10, large testes	Grisart et al. [2008]
12 (III.9)	unknown	M	1	paternal, father carrier	unknown	unknown	unknown	unknown	unknown	unknown	half-brother of patients 10 and 11	Grisart et al. [2008]
13 (II.6)	unknown	M	1	maternal, mother	unknown	ID	unknown	unknown	premature balding since the age of 18	dental enamel hypoplasia	father of patients 10, 11, and 12	Grisart et al. [2008]

Patient	Age at diagnosis [a]	Sex	Dup. type	Inheritance	Growth	Neuro-development	Neurology	Dysmorphic features	Hair	Tooth anomalies	Other	Reference
				carrier					years			
14 (II.9)	unknown	M	1	maternal, mother carrier	short stature, microcephaly	ID	unknown	unknown	premature balding	dental enamel hypoplasia	half-brother of patient 13	Grisart et al. [2008]
15 (1.4)	unknown	F	1	unknown	unknown	ID	unknown	unknown	unknown	unknown	mother of patients 13 and 14, grandmother of patients 10, 11 and 12	Grisart et al. [2008]
16 (III.6)	unknown	F	1	paternal, father carrier	not specified	not specified	not specified	not specified	not specified	not specified	healthy half-sister of patients 10 and 11, daughter of patient 13	Grisart et al. [2008]
17 (III.8)	unknown	F	1	paternal, father carrier	not specified	not specified	not specified	not specified	not specified	not specified	healthy half-sister of patients 10 and 11, daughter of patient 13	Grisart et al. [2008]
18	unknown	F	1 or 2	unknown	failure to thrive, microcephaly	DD, ID	not specified	not specified	not specified	not specified		Lu et al. [2007]
R609021	6.4 y	M	1	maternal	short stature, no macrocephaly	DD, ID	not specified	short midface, bulging forehead with marked frontal bossing	not specified	teeth and dental enamel anomalies were not detected	6 CALS; autism spectrum disorder, hyperactivity and poor verbal communication skills; brother of patient R509021	this study
R809021	5.1 y	F	1	maternal	short stature, no macrocephaly	not specified	not specified	short philtrum	not specified	not specified	hyperactivity, autism spectrum disorder, sister of patient R809021	this study
R509021	28 y	F	1	unknown	short stature, no macrocephaly	ID, learning disabilities	not specified	big forehead, short philtrum	not specified	not specified	mother of patients R809021 and R509021	this study
R653070	11 y	M	1	*de novo*	no short stature, no microcephaly or macrocephaly	ID (IQ75), learning disabilities	not specified	not specified	not specified	not specified	10 CALS, Lisch nodules detected in both eyes, younger brother with developmental delay/autism spectrum disorder	this study

a: age at diagnosis, years, months
CALS: café au lait spots
CL/P: cleft lip and palate
DD: developmental delay
ID: intellectual disability
DTRs 3+: deep tendon reflexes
F: female
GERD: gastroesophageal reflux disease
M: male
MRI; Magnetic resonance imaging
PC: percentile

Supp. Table S8. Diagnostic criteria for neurofibromatosis 1 (NF1)[a]

• 6 or more café au lait spots (0.5 cm in children or 1.5 cm in adults)
• 2 or more cutaneous/subcutaneous neurofibromas or one plexiform neurofibroma
• Axillary or inguinal freckling
• Optic pathway glioma
• 2 or more Lisch nodules (iris hamartomas seen on slit lamp examination)
• Bony dysplasia (sphenoid wing dysplasia, bowing of long bone pseudarthrosis)
• First degree relative with NF1

a: according to the National Institutes of Health Consensus Development Conference Statement: Neurofibromatosis Arch Neurol Chicago, 1988;45:575–578.

Supp. Text S1

Fluorescence in situ hybridization (FISH) was performed on 200 cultured blood cells using probes RP5-1002G3 (visible as red fluorescent signals) and RP5-926B9 (green). FISH probe RP5-1002G3 is located in the telomeric region of the *NF1* gene, whereas FISH probe RP5-926B9 spans the centromeric portion of the *NF1* gene [De Raedt et al., 2004]. Thus, two red/green fusion FISH signals should be obtained from an interphase nucleus with two normal chromosomes 17. By contrast, three red/green fusion FISH signals would be expected if the *NF1* gene region is duplicated.

Supp. Reference

De Raedt T, Brems H, Lopez-Correa C, Vermeesch JR, Marynen P, Legius E. 2004. Genomic organization and evolution of the *NF1* microdeletion region. Genomics 84:346-360.

Supp. Text S2

Clinical investigation

Index patient R609021

The male patient R609021 was 6 years and 4 months old at the time of investigation. He exhibited clinical signs of autism spectrum disorder including hyperactivity and poor verbal communication skills. Dysmorphic facial features, a bulging forehead (marked frontal bossing), midface hypoplasia as well as bluish sclerae were also noted. His body height was 114.7 cm (25th PC), his weight was 23 kg (50-75th PC) and his head circumference was 53 cm (75-90th PC). He had five CALS which were considered to be atypical with irregular borders: one café au lait spot located on his left abdomen (size: 4 x 2 cm); one café au lait spot on his right iliac spine (2 x 2 cm); one café au lait spot on his back (0.8 x 0.2 cm); 2 CALS on his right leg (0.6 x 0.5 mm and 1.1 x 0.5 cm in size) (Supp. Figure S6). He also exhibited a single freckle in each inguinal region. On his back, a patch of Mongolian spots regressing in the lumbosacral area were observed as well as a hypopigmented spot of 1.2 x 0.5 cm in size on the right side of his chest. The following finger and palmar dermatoglyphic patterns were noted: intermediate axial triradius with transverse mainline endings as well as a central pocket loop in the right 4th and 5th fingers. The teeth of the patient were normal; dental enamel anomalies were not observed.

Patient R809021

Patient R609021 had a younger sister (R809021) who was 5 years and one month old at the time of clinical investigation. Her body height was 105 cm (10-25th PC), her weight was 18 kg (25-50th PC) and her head circumference was 49.8 cm (25-50th PC). Patient R809021 exhibited hyperactivity and clinical signs of an autism spectrum disorder. Furthermore, bluish sclerae and a short philtrum were noted. Café au lait spots were not observed but two small freckles were present. Analysis of the finger and palmar dermatoglyphics indicated a tendency to central pocket loop in both 4th fingers and in the right 5th finger as well as intermediate axial triradius of the left palm.

Patient R509021

The mother (R509021) of patients R609021 and R809021 was 28 years old at the time of clinical investigation. She exhibited short stature with a body height of 150 cm (<5th PC). Her weight was 51 kg (25-50th PC) and her head circumference was 55 cm (50th PC). She had a big forehead, a short philtrum, and multiple freckles (also seen in her father) not restricted to inguinal and axillary regions. She reported that when she was pregnant with her son, patient R609021, she developed

gestational diabetes lasting until the end of the pregnancy as well as edema. She was also diagnosed with pre-eclampsia.

Neither of the duplication carriers of this family (family 1 in Supp. Figure S2) exhibited neurofibromas.

Index patient R653070

Physical examination at the age of 13 years revealed 10 CALS but no neurofibromas or axillary/ inguinal freckling. His height was 171.5 cm (90^{th} PC), weight 59.5 kg (80^{th} PC) and his head circumference was 56.5 cm (75^{th} to 90^{th} PC).