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ARTICLE

Towards mapping phenotypical traits in 18p− syndrome by array-based comparative genomic hybridisation and fluorescent *in situ* hybridisation

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Molecular karyotyping holds the promise of improving genotype–phenotype correlations for frequent chromosome conditions such as the 18p− syndrome. In spite of more than 150 reported cases with deletions in 18p, no reliable phenotype map for the characteristic clinical findings such as mental retardation, post-natal growth retardation and typical facial features has been established yet. Here, we report on four patients with partial monosomy 18p of different sizes owing to unbalanced translocations that were thoroughly characterised clinically and by molecular karyotyping. One patient had a terminal deletion of 1.6 Mb in 18p and a trisomy of 8q24.23-qter as determined by array-based comparative genomic hybridisation and large insert clone fluorescent *in situ* hybridisation. In two sibs and a fourth patient, cytogenetic and molecular-cytogenetic analyses showed the terminal deletions in 18p (8.0 and 13.84 Mb, respectively) to be accompanied by partial trisomies of 20p. Literature analyses of typical phenotypic features of 18p−, 8q+ and 20p+ syndromes allowed the attribution of clinical findings in our patients to the respective chromosomal aberration. Based on these data, we propose a phenotype map for several clinical features of the 18p− syndrome: Round face was tentatively mapped to the distal 1.6 Mb of 18p; post-natal growth retardation and seizures to the distal 8 Mb and ptosis and short neck to the proximal half of 18p.
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Introduction

The 18p− syndrome, first described by de Grouchy *et al*,[1] is one of the most frequent autosomal terminal deletion syndromes with more than 150 reported cases.[2,3] Monosomies of the entire short arm as well as partial monosomies 18p have been reported.[4] Frequent clinical features of 18p− syndrome include moderate to severe mental retardation, post-natal growth retardation, a round face, downturned corners of the mouth and dysplastic ears. Microcephaly, epicanthic folds, ptosis, hypertelorism, micrognathia, dental anomalies, short neck and pterygium colli are found less frequently. Malformations such as congenital heart defects or brain malformations mostly of the holoprosencephaly spectrum have also been reported.[2,5–7]

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In spite of the syndrome's frequency, no reliable phenotype map has been established. The fact that most of the 18p pure deletions involve the entire short arm[2] is the main reason for this. Moreover, the fact that partial deletions are essentially secondary to unbalanced translocations so that the phenotype is influenced by the concomitant trisomy may explain the phenotypic variability reported in the 18p− syndrome. Other factors influencing this variability may include age differences of the patients, inhomogeneous clinical classification, incomplete penetrance of the trait, undetected mosaicism and the uncovering of a recessive trait by the deletion. Obviously the definition of the exact deletion breakpoint may improve genotype/phenotype correlations. To our knowledge, only four cases with partial terminal monosomies of 18p have been reported with exact breakpoint analyses and detailed phenotypic data. One of them had an associated partial trisomy[8] and three had rather large deletions which hampered the attribution of phenotypical traits to different regions of 18p.[7]

Here, we report on four patients, two of which are sibs, with partial monosomies 18p of different sizes ranging from 1.6 to 13.8 Mb as demonstrated by breakpoint analyses. The molecular karyotypes detected were correlated with the patients' phenotypes obtained by thorough clinical investigations. Based on the data from our patients and the literature, we propose a tentative phenotype map for deletions of 18p.

Case reports

Case 1

The girl is the second of two children and was born after an uncomplicated pregnancy and delivery (42nd week of gestation). Birth measurements were normal (weight 3270 g/−0.7 SD, length 52 cm/−0.5 SD and occipital-frontal circumference (OFC) 36 cm/+0.7 SD). At the age of 6 months, motor development delay was noted by a paediatrician and treated with physiotherapy. With 11 months she started to crawl. Upon neuropaediatric evaluation at the age of 15 months, her fine motor skills, understanding of speech and social behaviour appeared normal. Her gross motor skills were delayed by 3–4 months. She could walk without assistance at the age of $1\frac{10}{12}$ years and speak at the age of 3 years. She had two febrile convulsions at the ages of $2\frac{6}{12}$ and $3\frac{6}{12}$ years. Upon clinical examination at the age of $3\frac{4}{12}$ years, she had short stature (length 87 cm/−3.4 SD, weight 14 kg/−0.5 SD and OFC 50 cm/+0.2 SD). Hypertelorism, slightly up-slanting palpebral fissures, a slightly up-turned and broad nose, slight micrognathia, a thin upper lip as well as a round face with full cheeks were noted (Figure 1, Table 2). There was no

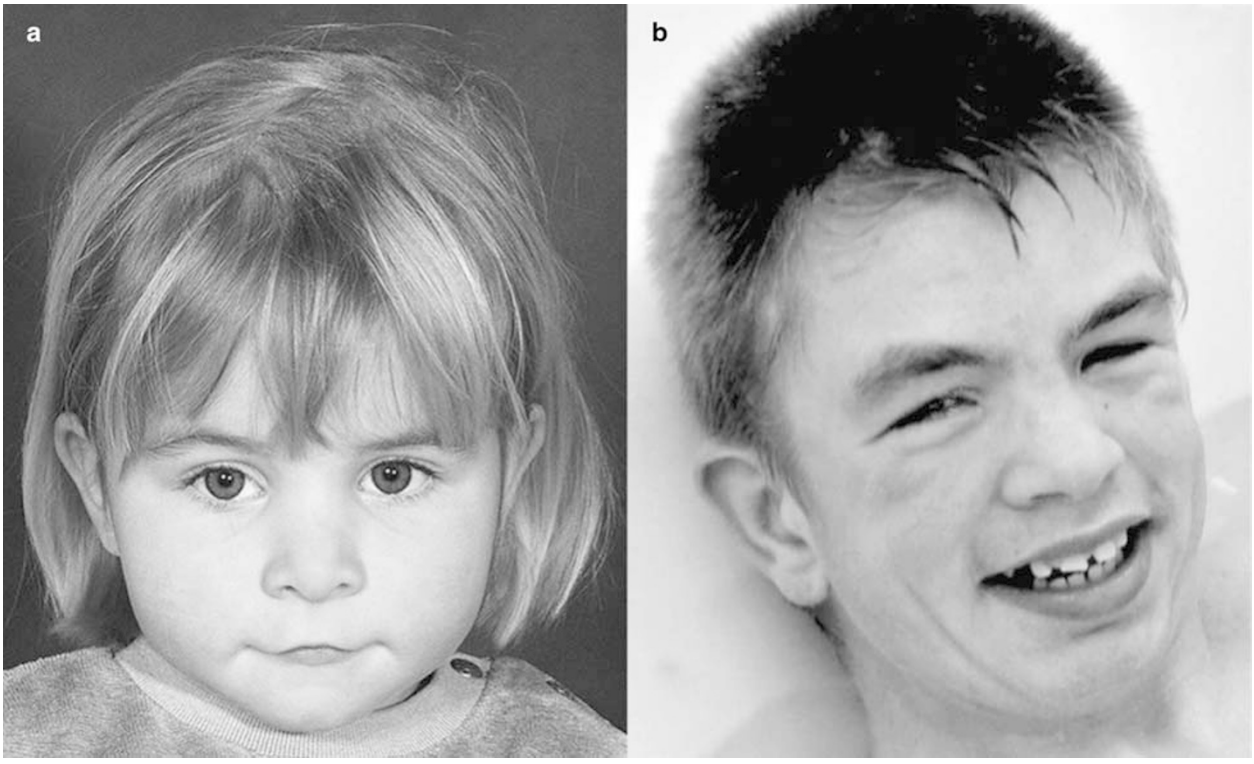

Figure 1 Facial appearances of case 1 and case 4. (**a**) Case 1 at age $3\frac{4}{12}$ years. (**b**) Case 4 at age 13. Permissions to publish pictures of cases 2 and 3 could not be obtained.

evidence for malformations. Her psycho-motor development was mildly retarded with a delay of 6–8 months, especially concerning speech and gross motor skills. After starting to attend a kindergarten with special assistance, her speech development improved. Repeated electroencephalograms gave normal results.

Case 2

The girl is the second of three children. She was born after an uncomplicated pregnancy and delivery (41st week of gestation). Her birth measurements were normal (weight 3040 g/−0.8 SD, length 51 cm/−0.7 SD and OFC 37 cm/ +1.6 SD). Her right hip was dislocated and her left hip was dysplastic. At the age of 2 months, she had become macrocephalic and a hydrocephalus internus with enlarged lateral and third ventricles and decreased brain volume mainly of the right cerebral hemisphere was diagnosed by cranial computed tomography (CCT). After a ventriculoperitoneal shunt operation, she developed a cerebral haemorrhage affecting the ventricles and the left temporo-parietal parenchyma. Recurrent cerebral seizures commenced 4 days after surgery. At the age of 3 months, inguinal hernia was noted and corrected surgically. Her psychomotor development was delayed. She was able to sit at the age of 2 years and to walk without support between 4 and 6 years of age. Speech development started with $2\frac{6}{12}$ years. Upon clinical examination at $8\frac{3}{12}$ years, she showed muscular hypotonia, ataxia and tetraparesis. At the age of $9\frac{10}{12}$ years, mental retardation was diagnosed by short non-verbal intelligence testing. Electroencephalography showed markedly slow basal activity with sharp-slow-wave-complexes confirming epileptic potential despite continuous antiepileptic medication. She used a wheel chair since age 20 but was able to walk freely for several steps. Upon clinical evaluation at the age of 22 years, she was mildly mentally retarded. Her height was 155 cm (−1.9 SD), her weight was 40 kg (−2.2 SD) and her OFC 56 cm (+0.8 SD). Her epilepsy was well controlled by medication. She had convergent strabismus, a broad nose, a thin upper lip, irregular dentition, a long face (like her father), slight bilateral syndactyly of the second and third toes and brachydactyly of both fifth fingers.

Case 3

The younger brother of case 2 was born after an uncomplicated pregnancy and delivery (42nd week of gestation). At birth, his weight was 2800 g (−2.3 SD), his length 50 cm (−1.3 SD) and his OFC 35 cm (−0.4 SD). He had muscular hypotonia as a child. At the age of 4 months, hydrocephalus internus with enlargement of the lateral, third and fourth ventricles and diffuse brain atrophy was diagnosed by CCT. At the age of 5 months, hemiparesis was noted. After several cerebral seizures starting at the age of $1\frac{6}{12}$ years, he is continuously treated with carbamazepine, which controls the epilepsy well. He was able to sit at $1\frac{6}{12}$ years and to walk at 5 years but showed no speech development. Upon clinical evaluation at the age of 16 years, he was severely mentally retarded with a speech development limited to vocalisation. His growth was severely retarded (height 142 cm/−4.2 SD, weight 30 kg/ −4.0 SD and OFC 50 cm/−4.8 SD). He had severe scoliosis, a long face (like his father), a broad nose and a prominent lower lip.

Case 4

This patient was first reported in 2003.[9] The boy is the first of two sons and was born at 34 weeks of gestation by caesarean section owing to breech presentation. The pregnancy had been complicated by an oligohydramnios caused by foetal renal dysplasia. At birth, measurements were normal (length 47 cm/±0 SD and weight 2510 g/±0 SD). Tetralogy of Fallot was diagnosed and bilateral renal dysplasia was confirmed. He was able to sit at the age of 12 months. At the age of $2\frac{6}{12}$ years, he was able to walk without support and spoke his first word. His measurements were normal at this age (height 93 cm/±0 SD, weight 13 kg/−0.5 SD and OFC 48 cm/+1.3 SD). He presented with dysmorphic features such as prominent forehead, telecanthus and hypertelorism, short nose, downturned corners of the mouth, widely spaced teeth and simply modelled helices of low-set ears. His hair was coarse and thick, he had a long neck with low posterior hairline and his voice was hoarse. He had small, widely spaced nipples and under-riding third toes. At the age of 7 years, dialysis was required because of renal insufficiency. Renal transplantation was performed at the age of $7\frac{6}{12}$ years. At re-examination at the age of $8\frac{6}{12}$ years, his body measurements were still normal (height 127 cm/−0.9 SD, weight 25 kg/−0.7 SD and OFC 53 cm/ +0.5 SD). At this time, he presented with a flat round face with full cheeks, periorbital fullness, hypertelorism, ptosis, a short base of the nose, micrognathia and a low posterior hairline. Both third toes were hypoplastic and under-riding. His facial appearance at the age of 13 years is shown in Figure 1.

Methods

Conventional cytogenetics and subtelomeric FISH

Conventional cytogenetic investigation of metaphases prepared from peripheral blood lymphocytes was performed according to standard procedures using GTG banding. Subtelomere fluorescent *in situ* hybridisation (FISH) with the ToTelVysion Probe Panel (Vysis, Downers Grove, IL, USA) was performed according to manufacturer's instructions.

Array-based CGH

Genomic DNA from peripheral blood of case 1 was analysed by array-based comparative genomic hybridisation (CGH). The genomic DNA array used comprises more

than 8000 large insert clones. Except for the addition of 2000 region specific clones from the RPCI (RZPD, Berlin, Germany) and CalTech (Invitrogen, Karlsruhe, Germany) BAC libraries, the array has been published previously.[10] Array assembly, hybridisation and analysis were essentially performed as described previously.[10]

Breakpoint analyses

For the exact determination of genomic aberration sizes, BAC and PAC clones were selected from the University of California Santa Cruz (UCSC) and Ensembl Genome Browsers. The BACs and PACs used as FISH probes for the 18p, 20p and 8q breakpoint analyses are given in Table 1. The DNA was isolated by midi-preparation and labelled by standard nick translation reactions either directly with FITC-dUTP (Roche, Mannheim, Germany), Cy3-dUTP (Amersham, Braunschweig, Germany) and DEAC-dCTP (Perkin Elmer, Rodgau-Jügesheim, Germany) or indirectly with biotin-dCTP (Invitrogen) and digoxigenin-11-dUTP (Roche). FISH was performed according to standard protocols and analysed using an Axioplan 2 imaging fluorescence microscope (Zeiss, Jena, Germany) using a Sensys CCD camera (Photometrics, Tucson, AZ, USA) and Cytovision software (Applied Imaging, Newcastle upon Tyne, UK).

If breakpoint-spanning clones were identified, imbalance sizes are given as the distance of the genomic midpoint of this clone from the corresponding telomere ± half of the clone size. If the breakpoint was located between two clones, imbalance sizes are given as the distance of the middle of the two genomic midpoints from the corresponding telomere ± half of the distance between the midpoints.

Table 1 FISH probes utilised in and results of 18p deletion mapping (left) and mapping of partial trisomies 8q (case 1) and 20p (cases 2–4) (right)

			Signal number						
18p BAC Clone ID	*Distance clone midpoint-telomere (Mb)*	*Chromosomal band*	*Case 1*	*Cases 2+3*	*Case 4*	*8q BAC Clone ID*	*Distance clone midpoint-telomere (Mb)*	*Chromosomal band*	*Signal number Case 1*
324G2	0.25	18p11.32	1	ND	ND	526P7	5.91	8q24.23/q24.3	3
769O8	0.71	18p11.32	1	ND	ND	172M18	6.89	8q24.23	3
1152E8	0.82	18p11.32	1	ND	ND	92G1	7.17	8q24.23	3
1018M4	0.98	18p11.32	1	ND	ND	644K4	7.48	8q24.23	2
1094D2	1.14	18p11.32	1	ND	ND	122H7	7.82	8q24.23	2
703C2	1.31	18p11.32	1	ND	ND	356M23	8.39	8q24.23	2
737E6	1.45	18p11.32	1	ND	ND	343P9	9.71	8q24.22/q24.23	2
598O4	1.51	18p11.32	1	ND	ND	45B19	10.62	8q24.22	2
702B13	1.60	18p11.32	2*	ND	ND	20p BAC and PAC Clone ID	Distance midpoint-telomere (Mb)	Chromosomal band	Signal number Cases 2+3
291G24	1.64	18p11.32	2	ND	ND				Case 4
78H1	2.22	18p11.32	2	1	1	153M12	0.99	20p13	3 3
113J12	3.51	18p11.31	ND	1	ND	26F18	3.16	20p13	ND 3
102E12	4.54	18p11.31	2	1	1	686P6	5.06	20p13/p12.3	3 3
956P8	5.49	18p11.31	ND	1	ND	1012M14	5.47	20p12.3	3 ND
96L16	6.39	18p11.31	2	1	1	1023O16	5.85	20p12.3	3 ND
1058N6	7.43	18p11.23	ND	1	ND	649H22	6.22	20p12.3	2* 3
125I18	7.56	18p11.23	2	1	ND	859D4	6.66	20p12.3	2 ND
939O4	7.70	18p11.23	ND	1	ND	997K18	8.14	20p12.3	2 3
22C7	7.86	18p11.23	ND	1	ND	91I2	9.97	20p12.2	ND 3
931H21	8.00	18p11.23	ND	2*	1	44H15	12.14	20p12.1	ND 2
878H17	8.33	18p11.23	2	2	ND	665P7	14.16	20p12.1	ND 2
644G14	8.48	18p11.23/p11.22	ND	2	ND	622P4	16.23	20p12.1	ND 2
56P20	8.60	18p11.22	ND	2	ND	383L5	18.21	20p11.23	ND 2
918F20	8.74	18p11.22	ND	2	ND	907N8	20.71	20p11.23	ND 2
775C11	9.00	18p11.22	ND	2	ND	135E6	23.16	20p11.21	ND 2
295A3	10.46	18p11.22	2	2	1	161K13	25.58	20p11.21	ND 2
500G20	11.95	18p11.21	2	2	1				
107F13	12.82	18p11.21	ND	ND	1				
463M18	13.51	18p11.21	ND	ND	1				
984A4	14.17	18p11.21	ND	ND	2				
54M22	14.88	18p11.21	ND	ND	2				

FISH results are given by the respective signal number: 1: monosomy, 2: disomy, 2*: breakpoint spanning clone (diminished signal intensity), 3: trisomy, ND: not determined. Monosomic and trisomic clones are colour-coded in dark grey and breakpoint-spanning clones in light grey. All clones are from the RCPI-11 human BAC library except for clones 859D4 and 997K18 that are PACs from the RCPI-5 library. Localisations according to UCSC Genome Browser May 2004 Freeze.

Results

Case 1

Conventional and subtelomere analysis GTG-banding revealed additional material of unknown origin located terminally on the short arm of chromosome 18. FISH-analysis with the subtelomeric probe for 18p (ToTelVysion Probe panel, Vysis) demonstrated this probe to be deleted in the derivative chromosome 18.

Array-based CGH The array-CGH analysis detected a terminal gain on 8q with a size of 6.8–8.3 Mb in addition to confirming a terminal deletion on 18p with a size of 0.71–1.55 Mb (Figure 2). The breakpoint for the 18p-monosomy was localised between clones RP11-769O8 (loss) and RP11-291G24 (normal ratio). The 8q breakpoint was localised between clones RP11-172M18 and RP11-356M23.

Breakpoint analysis The breakpoints were further defined by FISH with panels of BAC probes (Table 1). Clone RP11-702B13 showed a reduced signal intensity on the derivative chromosome 18 (Figure 3a) demonstrating RP11-702B13 to be a breakpoint spanning clone and the deletion size to be 1.60 ± 0.08 Mb. FISH localised the 8q breakpoint between clones RP11-92G1 and RP11-644K4 and determined the size of the trisomic segment to be 7.32 ± 0.16 Mb (Table 1).

Cases 2 and 3

Conventional cytogenetic and subtelomere analyses - Cytogenetic analysis identified a derivative short arm of chromosome 18 in cases 2 and 3. FISH with a subtelomeric probe for 18p (Vysis) showed this probe to be deleted in the derivative chromosome 18 in cases 2 and 3. Subsequent screening with the ToTelVysion Probe Panel found a third signal for the 20p subtelomeric probe on the derivative 18p identifying an unbalanced translocation between the short arms of chromosomes 18 and 20 in both cases. Subsequent FISH analyses of the family revealed a balanced subtelomeric translocation 18p/20p in the sibs' mother.

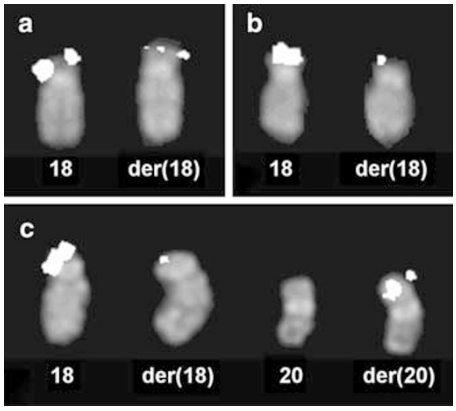

Figure 3 FISH results of 18p breakpoint determining clones in cases 1, 2 and 3. (**a**) Case 1: 18p breakpoint spanning clone RP11-702B13 showing diminished signal intensity on the derivative chromosome 18. The normal chromosome 18 was identified by FISH probe RP11-703C2 from the region deleted in the der(18) (not shown in this black and white image). (**b** and **c**) Case 2 and mother of cases 2 and 3: 18p breakpoint spanning clone RP11-931H21. (**b**) Unbalanced translocation (case 2): Diminished signal intensity on the derivative chromosome 18. (**c**) Balanced translocation (mother of cases 2 and 3): Split signal on both derivative chromosomes 18 and 20.

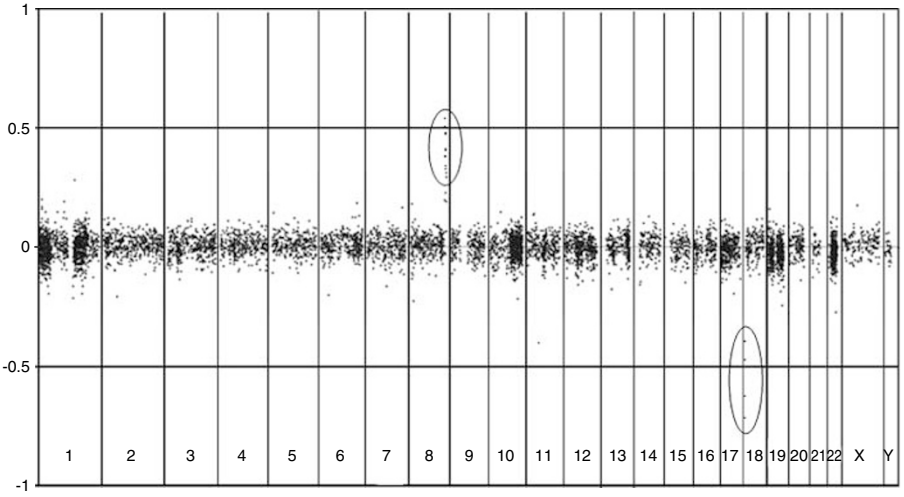

Figure 2 Array-CGH analysis of DNA from peripheral blood of case 1 identifying a gain of distal 8q and confirming the loss of terminal 18p. In the test/reference fluorescence intensity ratio plot of the whole genome, the BAC, PAC and cosmid clones are sorted according to their genomic location along the chromosomes with the short arm on the left and the long arm on the right. Gaps were introduced to mark the border between two chromosomes. Other gaps correspond to heterochromatin. The profile shows a loss of the three most distal clones in subband 18p11.32 (below 7 SD from the mean) and a gain of 13 clones in subbands 8q24.23–q24.3 (above 7 SD from the mean).

Breakpoint analysis The breakpoints were characterised by hybridisation of 18p and 20p FISH probe panels onto metaphases of unbalanced cases 2 and/or 3 and their balanced mother (Table 1). For BAC RP11-931H21 (18p11.23), a split signal was detected on the mother's der(18) and der(20) (Figure 3c) and diminished signal intensities on the der(18) of cases 2 and 3 (Figure 3b). Thus, RP11-931H21 was shown to span the 18p breakpoint located in 18p11.23. The deletion size on 18p was determined to be 8.0 ± 0.1 Mb in cases 2 and 3. For BAC RP11-649H22 (20p12.3), a split signal on the mother's der(20) and der(18) was detected and a diminished signal intensity on the der(18) in cases 2 and 3. The 20p breakpoint was thus shown to be spanned by clone RP11-649H22 and the size of the partial trisomy in cases 2 and 3 to be 6.2 ± 0.1 Mb.

Case 4

Cytogenetic analysis Neonatally and at the age of $2\frac{6}{12}$ years, normal male karyotypes were diagnosed with a structural resolution of 370 and 400 bands per haploid genome, respectively. At the age of $8\frac{6}{12}$ years, conventional chromosomal analysis at 460 bands per haploid genome revealed a derivative chromosome 18 suggestive of an unbalanced translocation. By analyses of parental chromosomes, a balanced translocation between 18p and 20p was found in the father and a normal karyotype in the mother. Thus, case 4 was found to have a paternally derived unbalanced translocation der(18)t(18;20) with cytogenetically determined breakpoints in 18p11.2 and 20p12.3.

Breakpoint analysis FISH analysis with BAC/PAC probe panels was performed to determine the 18p and 20p breakpoints in case 4 (Table 1). The 18p breakpoint was localised in 18p11.21 between clones RP11-463M18 (monosomic) and RP11-984A4 (disomic). The deletion size was determined to be 13.84 ± 0.33 Mb. The 20p breakpoint was localised between clones RP11-91I2 (trisomic, 20p12.2) and RP11-44H15 (disomic, 20p12.1) and the trisomy size was determined to be 11.06 ± 1.09 Mb.

Discussion

Here, we report on four patients with differently sized partial monosomies of 18p and accompanying partial trisomies of 8q (case 1) and 20p (cases 2–4) as identified by array-based CGH and FISH-analysis with BAC and PAC probes. The deletions were determined to encompass the terminal 1.6 Mb (breakpoint in 18p11.32, case 1), 8.0 Mb (breakpoint in 18p11.23, familial cases 2 and 3) and 13.84 Mb (breakpoint in 18p11.21, case 4) of 18p, respectively.

It was the aim of this study to correlate the chromosomal aberrations present in the studied patients with the patients' clinical features in order to start the compilation of a phenotype map in 18p− syndrome. As our patients' phenotypes were caused by aberrations of two different chromosomal regions, particular caution was exercised when trying to distinguish the clinical effects of the two genomic alterations. Thus, we compared the phenotypic findings in our patients with those described in the literature for patients with partial deletions of 18p[2,5,10–20] and patients with partial trisomies of 8q[2,11,12] or 20p.[13–19] These data are summarised in Table 2. Although a synergistic effect of both defects, that is, the partial monosomy and the partial trisomy, in the determination of the phenotype cannot be ruled out entirely, we attributed clinical features to the deletion of 18p if they were present in at least one of our cases and have not been reported for the respective patient's partial trisomy.

On the basis of our analyses, we delineated a tentative phenotype map for partial monosomies of the short arm of chromosome 18 (Figure 4). The round face of case 1 has not been described for partial trisomies of distal 8q so that it was assigned to a deletion of the distal 1.6 Mb of 18p. Likewise, we attributed two symptoms, namely post-natal growth retardation and seizures, to a deletion of the distal half of 18p because they were present in patients 2 and 3, but not in patients with partial trisomy 20p. Thus, we suggest that haploinsufficiency of genes in the distal 8 Mb of 18p may cause post-natal growth retardation and seizures. This hypothesis is supported by two published patients with short stature and slightly larger terminal 18p deletions of 10.2–10.7 Mb and 10.7–11.8 Mb, respectively.[7] USP14 (ubiquitin-specific protease 14), located at 18p11.32, may be an interesting gene in the context of growth retardation and seizures as mice with a mutation resulting in reduced protein expression were growth-retarded[20] and the protein has been implicated in regulating synaptic activity in mammals.[33] A second possible candidate gene for seizures, DLGAP1 (discs large-associated protein 1) located at 18p11.31, is part of the postsynaptic density in neuronal cells.[34] The hydrocephalus internus with brain atrophy present in cases 2 and 3 points to the localisation of a responsible haploinsufficient gene in the distal 8 Mb of 18p. This gene is most likely different from the TGIF gene located at 18p11.31. Mutations in TGIF cause alterations from the holoprosencephaly spectrum including specific brain malformations, which are found in at least 10% of patients with 18p− syndrome.[2] However, cases 2 and 3 showed no symptoms of holoprosencephaly. Hip dislocation is not included in our phenotype map although it is present in case 2 as it is common in the general population and is found in only 10% of 18p− patients.[5]

We attributed known clinical features of 18p− syndrome to the proximal half of 18p if they were absent in our three patients in whom the proximal 7.4 Mb of 18p were not deleted (cases 1–3). This approach is not as straightforward because the absence of a feature may also be due to reduced

Table 2 Clinical features of the four patients with partial monosomy 18p and partial trisomies 8q or 20p presented here in comparison to the typical phenotype of 18p−, 20p+ and 8q+ syndromes

Feature	*18p− syndrome*	*Case 1 ($3\frac{1}{2}$ years)*	*Case 2 (22 years)*	*Case 3 (16 years)*	*Case 4 ($6\frac{1}{2}$ years)*	*Partial trisomy 20p*[a]	*Partial trisomy 8q21–q24 - qter*[2,11,12]
		Cytogenetic breakpoint and size of partial monosomy 18p					
		18p11.32; 1.6 Mb	*18p11.23; 8.0 Mb*	*18p11.23; 8.0 Mb*	*18p11.21; 13.8 Mb*		
		Cytogenetic breakpoint and size of accompanying partial trisomy					
		8q24.23; 7.5 Mb	*20p12.3; 6.2 Mb*	*20p12.3; 6.2 Mb*	*20p12.1–0.2; 11.1 Mb*		
Anthropometric data							
Post-natal growth retardation	85%[5]	+	(+)	+	−	−	+
Normal birth measurements	>70%[5]	+	(+)	−	+	+	+
Microcephaly	29%[21]	−	−	+	−	+	−
Malformations							
Brain malformation	+[2]	−	+	+	−	−	−
Congenital heart defects	10%[21]	−	−	−	+	+	+
Dysplastic kidney	3%[5]	−	−	−	+	(+)	+
Genital malformation/ hypoplasia	18%[5]	−	−	−	−	(+)	+
Inguinal hernia	7%[b]	−	+	−	−	+	−
Vertebral malformations	+[2]	−	−	+	−	+	+
Other anomalies							
Mental retardation/ developmental delay	100%[5]	+	+	+	+	+	+
Muscular hypotonia	−[2]	−	+	+	−	+	+
Seizures	+[6,22,23]	(+)	+	+	−	−	+
Hip dislocation	10%[5]	−	+	−	−	−	−
Dental anomalies	29%[21]	−	+	−	(+)	+	+
Craniofacial anomalies							
Flat occiput	−[2]	−	−	−	−	(+)	+
Brachycephaly	14%[b]	−	−	−	−	(+)	−
Prominent forehead	21%[b]	−	−	−	+	(+)	+
Flat face	−[2]	−	−	−	+	+	−
Round face	50%[b]	+	(−)	(−)	+	+	−
Up-slanting palpebral fissures	+[2]	(+)	−	−	−	+	+
Epicanthic folds	40%[21]	−	(−)	(−)	−	+	−
Ptosis	38%[21]	−	−	−	+	−	−
Hypertelorism/ Telecanthus	41%[21]	+	−	−	+	+	+
Up-turned nose	−[2]	(+)	−	−	+	+	−
Flat nose	+[2]	−	−	−	−	+	−
Broad nose	+[2]	(+)	+	+	−	+	+
Full cheeks	−[2]	+	(−)	(−)	+	+	+

Table 2 (*Continued*)

Feature	*18p− syndrome*	*Case 1 ($3\frac{1}{2}$ years)*	*Case 2 (22 years)*	*Case 3 (16 years)*	*Case 4 ($6\frac{1}{2}$ years)*	*Partial trisomy 20p*[a]	*Partial trisomy 8q21–q24 - qter*[2,11,12]
		Cytogenetic breakpoint and size of partial monosomy 18p					
		18p11.32; 1.6 Mb	*18p11.23; 8.0 Mb*	*18p11.23; 8.0 Mb*	*18p11.21; 13.8 Mb*		
		Cytogenetic breakpoint and size of accompanying partial trisomy					
		8q24.23; 7.5 Mb	*20p12.3; 6.2 Mb*	*20p12.3; 6.2 Mb*	*20p12.1–0.2; 11.1 Mb*		
Down-turned corners of mouth	57%[b]	−	−	−	+	−	+
Micrognathia	35%[5]	(+)	−	−	+	+	+
Low-set ears	−[2]	−	−	−	+	+	+
Dysplastic ears	53%[b]	−	−	−	+	+	+
Low posterior hairline	+[2]	−	−	−	+	(+)	+
Anomalies of neck/trunk							
Short neck	42%[5]	−	−	−	−	(+)	+
Pterygium colli	25%[5]	−	−	−	−	(+)	−
Widely spaced hypoplastic nipples	+[2]	−	−	−	+	+	+
Anomalies of extremities							
Brachydactyly	+[2]	−	+	−	−	+	−
Clinodactyly	21%[21]	−	−	−	−	+	+
Hands/feet short and broad	21%[b]	−	−	−	−	−	−
Syndactyly, for example, second/third toe	13%[5]	−	+	−	−	+	−
Other anomalies of the toes	−[2]	−	−	−	(+)	+	+

−: absent/not detected; (−) absent, absence possibly owing to age at description (+): slightly/seldom present; +: present.
[a]Analysis of 72 patients from eight publications[2,13–19]
[b]Analysis of 75 patients from 10 publications[5,24–32]

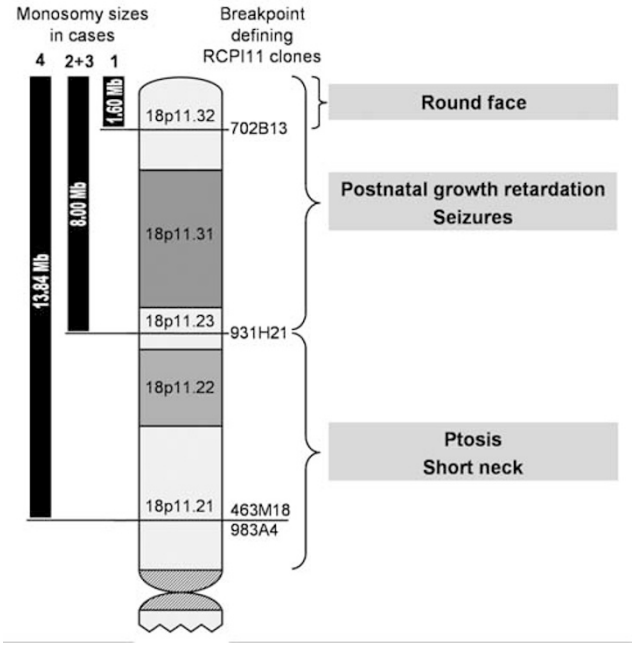

Figure 4 Proposed phenotypic map for deletions of 18p based on the findings in our patients and analysis of published cases.[2,5,10,13–21,24,26–29] The partial monosomies of cases 1-4 are indicated by horizontal bars (breakpoints) and vertical bars (sizes of deletions). Round face is present in case 1 and has not been described for partial trisomy 8q. Post-natal growth retardation and seizures are 18p− syndrome symptoms that are present in case 2 and/or 3 and have not been described for partial trisomy 20p. Thus, we attribute these symptoms to haploinsufficiency of genes located in the distal 8 Mb of 18p. In contrast, we propose that genes in the proximal half of 18p are responsible for two dysmorphisms of 18p− syndrome that have been described with penetrances of 38–42% in the literature but are absent in cases 1–3 (ptosis and short neck).

penetrance. Consequently, we only included those features absent in cases 1–3 for which the respective penetrance was known to be above 35%. Penetrance data were either gathered from reviews[5,21] or from our own analysis of 75 published patients.[5,24–32] Interestingly, two features of the typical facial appearance of 18p− syndrome met the criteria of absence in cases 1–3 and of penetrances above 35%. Thus, we propose that ptosis and short neck are caused by haploinsufficiency of genes located in the proximal half of 18p. This reasoning is supported by two reports indicating that neither of these traits were present in a patient with monosomy of the distal 8.3–8.4 Mb of 18p[8] and that both traits were found in two patients with deletions larger than 10 Mb.[7]

In summary, we present a first tentative genotype–phenotype map for patients with an 18p− syndrome. This study contributes towards achieving more accurate phenotype predictions in patients with monosomy of 18p necessary for precise genetic counselling. Additionally, the attribution of clinical features to different parts of the short arm of chromosome 18 may eventually help to identify genes responsible for specific symptoms of this frequent deletion syndrome.

Electronic database information

URLs for data presented in this report are as follows: University of California Santa Cruz (UCSC) Genome Browser, http://genome.ucsc.edu, assembly May 2004; Ensembl Genome Browser, http://www.ensembl.org/Homo_sapiens/

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