Pitt-Rogers-Danks syndrome and Wolf-Hirschhorn syndrome are caused by a deletion in the same region on chromosome 4p16.3

Sarina G Kant, Arie Van Haeringen, Egbert Bakker, Ingrid Stec, Dian Donnai, Paul Mollevanger, Geoffrey C Beverstock, Mirjam C Lindeman-Kusse, Gert-Jan B Van Ommen

Department of Clinical Genetics, University Hospital Leiden K5-R, PO Box 9600, 2300 RC Leiden, The Netherlands
S G Kant
A Van Haeringen

Department of Clinical Cytogenetics, University Hospital Leiden, Leiden, The Netherlands
P Mollevanger
G C Beverstock

Department of Human Genetics, Leiden University, Leiden, The Netherlands
E Bakker
I Stec
G-J B Van Ommen

De Hartekamp and De Hondseberg, Institutions for the Mentally Disabled, Heemstede and Oisterwijk, The Netherlands
M C Lindeman-Kusse

Department of Medical Genetics, St Mary's Hospital, Manchester, UK
D Donnai

Correspondence to: Mrs Kant.
Received 24 October 1996
Revised version accepted for publication 30 January 1997

Abstract
Recently, a deletion of chromosome 4pter was found in three patients with Pitt-Rogers-Danks syndrome. We investigated two of these patients, by means of DNA and FISH studies, together with two additional patients with Pitt-Rogers-Danks syndrome, to determine the critical region of the deletion in these patients and to compare this with the critical region in Wolf-Hirschhorn syndrome.
All four patients showed terminal deletions of chromosome 4p of different sizes. One of them appeared to have an unbalanced karyotype caused by a cryptic translocation t(4;8) in the mother, resulting in a deletion of chromosome 4pter and a duplication of chromosome 8pter. The localisation of the Wolf-Hirschhorn critical region has been confined to approximately 1 Mb between D4S43 and D4S115. Our study shows that the deletions in four patients with the Pitt-Rogers-Danks syndrome overlap the Wolf-Hirschhorn critical region and extend beyond this in both directions. This study, combined with the fact that our third patient, who was previously described as a Pitt-Rogers-Danks patient, but who now more closely resembles a Wolf-Hirschhorn patient, makes it likely that Pitt-Rogers-Danks and Wolf-Hirschhorn syndromes are different clinical phenotypes resulting from a deletion in the same microscopic region on chromosome 4p16.
(J Med Genet 1997;34:569-572)

Keywords: Pitt-Rogers-Danks syndrome; Wolf-Hirschhorn syndrome; 4p deletion; 4p-

Pitt-Rogers-Danks syndrome (PRDS) is a rare syndrome characterised by prenatal and post-natal growth retardation, microcephaly, prominent eyes, short philtrum, a large mouth, and mental retardation.
The first four patients were described in 1984 by Pitt *et al.*¹ who suggested this condition might be autosomal recessive. Subsequently, Donnai,² Oorthuys and Bleeker-Wagemakers,³ and Lizcano-Gil *et al.*⁴ all presented additional cases with PRDS. No chromosomal abnormalities were found. However, recently, we and others^{5,6} described four patients with PRDS and a deletion of chromosome 4pter. Additionally, Clemens *et al.*⁷ described a 4pter deletion in the two sisters originally described by Pitt *et al.*¹ Retrospective cytogenetic investigation of the patient of Oorthuys and Bleeker-Wagemakers³ showed a hitherto unnoticed 4pter deletion.
A partial deletion of the short arm of chromosome 4 is also found in patients with Wolf-Hirschhorn syndrome (WHS). This syndrome is characterised by mental and growth retardation and a distinct pattern of dysmorphic features.⁷ Other authors have already suggested that one of the cases described by Pitt *et al.*¹ had facial features suggestive of Wolf-Hirschhorn syndrome.⁸ The WHS critical region is localised between D4S43 and D4S115 and is about 1 Mb in size.^{9,10}
We further investigated the two patients described previously by us⁶ (patients 1 and 2), the patient described by Oorthuys and Bleeker-Wagemakers³ (patient 3), and a new patient (patient 4) with PRDS to determine the critical region of the deletion in these patients and to compare this region with the WHS critical region.
Case reports
Three unrelated previously reported patients and a new patient with PRDS and a chromosome 4p deletion were studied. The clinical features of the patients are summarised in table 1.
The first patient is a white girl born in 1982. She has clinical features of PRDS, which have previously been documented.⁶ Her karyotype is

Table 1 Clinical features in WHS and our patients

	WHS	Patient 1	Patient 2	Patient 3	Patient 4
Low birth weight	+	+	+	+	+
Short stature	+	+	+	+	+
Mental retardation	+	+	+	+	+
Epilepsy	+	+	+	-	+
Microcephaly	+	+	+	+	+
Hypertelorism	+	+	+	+	+
Prominent glabella	+	-	-	-	-
Beaked nose	+	+	+	+	+
Short philtrum	+	+	+	+	+
Maxillary hypoplasia	-	+	+	+	+
Wide mouth	-	+	+	+	+
Cleft lip/palate	+	-	-	-	-
Micrognathia	+	-	-	-	-
Ears low set/simple	+	-	-	+	+
Cardiac malformation	+	-	-	+	-
Urogenital malformation	+	-	-	-	+

46,XX,del(4)(p16.3). Both parents have normal chromosomes.

The second patient, a white male born in 1951, has the typical clinical features of PRDS, which have also been extensively documented by the previous authors.⁶ His karyotype is 46,XY,del(4)(p16.3). The father has normal chromosomes. The mother was dead.

The third patient is a white girl born in 1976 (fig 1). Her clinical features have been fully described by Oorthuys and Bleeker-Wagemakers.³ Chromosome studies at that time appeared to be normal. Fluorescence in situ hybridisation (FISH) analysis now shows a partial deletion of chromosome 4p and a partial trisomy of chromosome 8p as a result of an unbalanced cryptic translocation. Her karyotype is 46,XX,der(4),t(4;8)(p16.3;p23.1). The mother was a carrier of the balanced form of this translocation. The father had normal chromosomes.

The fourth patient, reported by Donnai,¹¹ was born at term in 1980 weighing 2500 g. She fed poorly and gained weight slowly. Investigations showed the presence of only one kidney. At 13 months she began to have seizures, usually with fever, and anticonvulsant medication was begun. Her developmental milestones were slow; she walked at 2 years, spoke in sentences at 5 years, and could read simple words and write her name at 11 years.

When examined at 11 years 9 months she was small (height, weight, and head circumference below the 3rd centile). She had a triangular face with prominent eyes, a beaked nose, and a short philtrum. Her ears were simple with unfolded helices. Her labia majora were hypoplastic and on her right palm there was an extra transverse crease.

Her referral diagnosis was PRDS, but her small size and facial features were reminiscent of WHS. Fluorescent in situ hybridisation using Oncor WHS cosmid probe (D4Z1) confirmed a deletion of 4p16.3-pter.

Materials and methods

The deletion was studied using standard FISH techniques as described by Lichter *et al.*¹² The FISH probes used are listed in table 2. In addition

Table 2 Results of FISH and DNA analysis

Locus	Probe	Probe type	Patient			
			1	2	3	4
D4F26	pC847.351	F	-	-	-	-
D4S90	190H5	F	-	-	ND	ND
D4S43	c39	D	-	?	-	?
D4S136	C80	F	-	-	ND	ND
D4S136	cyc8.91	F	-	ND	ND	ND
D4S127	p363	D	?	?	-	?
D4S126	c102	D	-	?	-	?
D4S81	pRB1.6	S	+	-	-	-
D4S10	C5.5	F	+	+	ND	ND
D4S10	H5.52	S	+	+	-	ND
D4S10	pTV20	S	+	+	-	ND
D4S10	p5.5	S	+	+	-	ND
D4S10	F5.53	S	ND	ND	ND	-
D4S62		S	ND	ND	ND	+
D4S394	037yg1	D	ND	ND	-	?
D4S1582	283zf5	D	ND	ND	+	+

The probes, FISH (F) as well as Southern blot (S) and dinucleotide repeat (D), are listed in order from the most distal on chromosome 4p (D4F26, 150 kb from the telomere) to the most proximal. ND not determined, ? results inconclusive, - deletion present, + no deletion present.

tion the deletion was analysed using primer sets for dinucleotide repeats from distal 4p loci and by Southern blot hybridisation with markers coding for distal 4p loci. The probe names used are listed in table 2. All the probes have been precisely localised on a long range restriction map of chromosome 4p.¹³⁻¹⁵ Appropriate informed consent was obtained from all four patients and their parents concerning the FISH and DNA analysis.

Results

The results of both the FISH and DNA analyses are listed in table 2. Whenever a probe was tested that was inconclusive, this is indicated in the table by a question mark. In patient 1 this occurred on locus D4S127, because the mother is homozygous for this locus and the father heterozygous with one allele of the same size as the mother. Patient 1 is therefore either homozygous for this same polymorphism or has a deletion at this locus. In patient 2, three probes were inconclusive (D4S43, D4S127, and D4S126) because the father is homozygous for these loci and the mother was not available for analysis. Patient 2 is thus either homozygous for these polymorphisms or has a deletion of these loci. In patient 4, four probes were inconclusive (D4S43, D4S127, D4S126, and D4S394) because the mother is homozygous for these loci and the father was not available for analysis. Patient 4 is, like patient 2, thus either homozygous for these polymorphisms or has a deletion of these loci. However, a plausible explanation is that in all patients the deletion is present in the loci concerned, except for locus D4S394 in patient 4, because they are flanked on both sides by a deleted locus.

The most proximal deleted locus on chromosome 4p in patient 1 is D4S126, in patient 2 D4S81, in patient 3 D4S394, and in patient 4 D4S10. The most distal locus tested on chromosome 4p (D4F26, 150 kb from the telomere) is deleted in all four patients. Examples of two different loci are given in figs 2 and 3. In fig 2, the FISH signal of probe pC847.351 (D4F26) is shown in patient 3 on one of the

Figure 1 (A) Front view and (B) side view of patient 3 aged 19 years.

Figure 2 FISH signal of probe pC847.351 (D4F26) in patient 3.

Figure 3 Southern blot of the pRB1.6 (D4S81) polymorphism in patients 1, 2, and 3 (P1, P2, P3) and their parents (F=father, M=mother, C=control). A deletion is present in patients 2 and 3, but not in patient 1.

Figure 4 Physical map of chromosome 4p. The bars below the map illustrate the WHS critical region and the deletions found in the four patients.

chromosome 4 homologues. Figure 3 shows a Southern blot of the pRB1.6 polymorphism (D4S81) for patients 1, 2, and 3 and their parents. In patients 2 and 3 a deletion is present while in patient 1 no deletion is present. From this we conclude that the minimal deleted region in the four patients extends from D4S126 to the telomere.

Patient 1 has received the deleted chromosome from her father. Patient 3 obtained the deleted translocation chromosome from her mother. In patients 2 and 4 it was not possible to determine the parent of origin, because only one parent of each patient was available for analysis.

Discussion

PRDS is a rare condition and fewer than 20 patients have been recorded.¹⁻⁶ Recently, in six of these patients, a deletion 4p has been found.^{5,6} We have reinvestigated the patient of Oorthuys and Bleeker-Wagemakers,³ in whom a deletion 4p was also found, caused by an unbalanced cryptic translocation (4;8). We analysed the patients of Lindeman-Kusse *et al*⁶ and Oorthuys and Bleeker-Wagemakers³ and a new patient to determine if the critical region of the deletion was the same as in the WHS.

This study clearly shows that PRDS is caused by a deletion overlapping the critical region of WHS (fig 4). Interestingly, the phenotype in PRDS is not quite the same as the WHS phenotype. In general it appears that not only are the clinical abnormalities in WHS more severe than in PRDS, but also that the prognosis of WHS seems to be worse than in PRDS.

Several different explanations for this phenotypic difference can be proposed. The influence of imprinting is not very likely. In two of our patients with PRDS the parent of origin is known, as well as in the two sisters originally described by Pitt *et al*.⁵ In three of these patients the deletion is paternally derived and in one maternally derived. In WHS a de novo deletion can also originate from either the mother or the father without any major clinical differences.¹⁵

In patient 3, one might conclude that there is an influence on the phenotype of PRDS of duplication of chromosome 8. However, Tranebjaerg *et al*¹⁷ described two brothers with partial trisomy of chromosome 8p caused by a translocation (8;14)(p23.1;q32.3). These two boys only shared subtle facial dysmorphism and slight mental retardation. The younger boy had delayed speech development but attended normal school. From these data, it appears that the contribution of duplication of 8p to the phenotype in PRDS is minimal.

A contiguous gene syndrome could be held responsible for the difference in phenotype in WHS and PRDS. The expectation would then be that deletions in WHS would be larger than in PRDS, explaining the more severe phenotype in WHS. However, our three patients all have a larger deletion than some patients described as having typical features of WHS.^{9,18}

Patient 3 in our study more closely resembles the WHS phenotype at the age of 19 years than

she did at the age of 8 years.³ This could be because of a changing phenotype in WHS, although many patients with WHS are diagnosed shortly after birth.

In our opinion, WHS and PRDS are two phenotypes caused by a deletion in the same microscopic region on chromosome 4p16. The difference between the two syndromes is that PRDS is diagnosed in those who have a less severe phenotype and present later, and WHS is diagnosed in those who are more severe and thus present early.

Further work, especially in other PRDS patients, is needed to limit the critical region in PRDS and to confirm or disprove that PRDS and WHS are different expressions of the same chromosomal abnormality and possibly of the same gene.

We would like to thank Dr H G Brunner and Dr J J G Hoorweg-Nijman for kindly providing DNA and blood samples from their patients.

- 1 Pitt DB, Rogers JG, Danks DM. Mental retardation, unusual face, and intrauterine growth retardation: a new recessive syndrome? *Am J Med Genet* 1984;19:307-13.
- 2 Donnai D. A further patient with the Pitt-Rogers-Danks syndrome of mental retardation, unusual face, and intrauterine growth retardation. *Am J Med Genet* 1986;24:29-32.
- 3 Oorthuys JWE, Bleeker-Wagemakers EM. A girl with the Pitt-Rogers-Danks syndrome. *Am J Med Genet* 1989;32:140-1.
- 4 Lizcano-Gil LA, Garcia-Cruz D, Garcia-Cruz O, Sanchez-Corona J. Pitt-Rogers-Danks syndrome: further delineation. *Am J Med Genet* 1995;55:420-2.
- 5 Clemens M, Martsof JT, Rogers JG, Mowery-Rushton P, Surti U, McPherson EW. Pitt-Rogers-Danks syndrome: the result of a 4p microdeletion. *Am J Med Genet* 1996;66:95-100.
- 6 Lindeman-Kusse MC, Van Haeringen A, Hoorweg-Nijman JJG, Brunner HG. Cytogenetic abnormalities in two new patients with Pitt-Rogers-Danks phenotype. *Am J Med Genet* 1996;66:104-12.
- 7 Guthrie RD, Aase JM, Asper AC, Smith DW. The 4p-syndrome. *Am J Dis Child* 1971;122:421-5.
- 8 Winter RM, Baraitser M. *Multiple congenital anomalies*. 1st ed. London: Chapman & Hall, 1991:493.
- 9 Gandelman K, Gibson L, Stephen Meyn M, Yang-Feng TL. Molecular definition of the smallest region of overlap in the Wolf-Hirschhorn syndrome. *Am J Hum Genet* 1992;51:571-8.
- 10 Wright TJ, Denison K, Altherr MR. Redefinition and molecular analysis of the critical region for the Wolf-Hirschhorn syndrome locus. *Am J Hum Genet Suppl* 1995;57:A233.
- 11 Donnai D. Editorial comment: Pitt-Rogers-Danks syndrome and Wolf-Hirschhorn syndrome. *Am J Med Genet* 1996;66:101-3.
- 12 Lichter P, Tang EJC, Call K, et al. High resolution mapping of human chromosome 11 by in situ hybridization with cosmid clones. *Science* 1990;247:64-9.
- 13 Altherr MR, Bengtsson U, Elder FF, et al. Molecular confirmation of Wolf-Hirschhorn syndrome with a subtle translocation of chromosome 4. *Am J Hum Genet* 1991;49:1235-42.
- 14 Van Ommen GJB, Buetow KH, Murray JC. Report of the committee on the genetic constitution of chromosome 4. *Genome Priority Reports* 1993;1:221-48.
- 15 Gyapay G, Morissette J, Vignal A, et al. The 1993-94 Génethon human genetic linkage map. *Nat Genet* 1994;7:246-9.
- 16 Dallapiccola B, Mandich P, Bellone E, et al. Parental origin of chromosome 4p deletion in Wolf-Hirschhorn syndrome. *Am J Med Genet* 1993;47:921-4.
- 17 Tranebjaerg L, Mansa B, Steenbuch L. Prometaphase chromosome and immunological studies in different derivatives of balanced t(8;14)(p23;q323). *Clin Genet* 1984;26:276-7.
- 18 McKeown C, Read AP, Dodge A, Strecko O, Mercer A, Harris R. Wolf-Hirschhorn locus is distal to D4S10 on the short arm of chromosome 4. *J Med Genet* 1987;24:410-12.