The 13q− Deletion Syndrome

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Clinical data are available for 23 cases with the karyotype 46 Dq− or 46 Dr. Seventeen cases have been published, data on four were obtained through personal communication, and two have been studied in our laboratory. Autoradiographic identification of a ring D chromosome has previously been made in six of these cases. In five, the ring originated from a chromosome 13 (Bloom et al 1967; Gerald et al. 1967; Hollowell et al. 1967; Niebuhr and Mikkelsen, personal communication [1968]; and Stimson and Hecht, personal communication [1968]). In one case (Sparkes et al. 1967), the ring apparently originated from a chromosome 14. The authors postulated that their case represented a unique syndrome characterized by absent thumbs and suggested that the same syndrome was present in two cases reported earlier, each having an unidentified ring D chromosome (Bain and Gauld 1963; Adams 1965). The patient studied by Sparkes et al. (1967) had microcephaly, hypertelorism, a broad nasal bridge, ocular dystrophy, and large ears. These features are characteristic of the "Dr" syndrome which Lejeune et al. (1968) proposed as antithetical to trisomy 13.

Congenital anomalies typical of the "Dr" syndrome have also been observed in individuals with a Dq− chromosome. Three of these patients had hypoplastic thumbs (Thompson and Lyons 1965; Laurent et al. 1967; Hollowell, personal communication [1969]). The Dq− chromosome originated from a chromosome 13 in two cases where terminal labeling studies were performed (Gey, personal communication [1968]; Hollowell, personal communication [1969]).

The present paper concerns two clinically similar cases which resemble others with the karyotype 46 Dq− or 46 Dr. Case 1 has a small deletion from a distal segment of the long arm of a D-group chromosome, and case 2 has a ring D chromosome. We have identified each abnormal chromosome autoradiographically as a chromosome 13. We postulate that our cases illustrate one distinct chromosome-deletion syndrome caused by partial monosomy for a distal segment of the long arm of chromosome 13.

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CASE REPORTS

Case 1 was an 18-month-old white male with severe psychomotor retardation. He was the first child of a 42-year-old father and 27-year-old mother. The gestation period was 44 weeks. He weighed 2,300 g, was apneic and flaccid, and required resuscitation at birth. The head was asymmetric with flattening of the top of the left parietal bone and overriding sagittal sutures. The anterior fontanel was soft and flat, and measured 1.5 by 1.5 cm. Microcephaly, trigonocephaly, hypertelorism, a broad nasal bridge, and micrognathia were present (fig. 1). The eyes were microphthalmic, with ptosis, antimongoloid palpebral fissures, epicanthus, and bilateral inferior colobomata of the irises. The ears were simple, low set, and malrotated. The neck was short and webbed, the palate normal. There was marked pectus excavatus and a systolic murmur.

Both humeri were normal, but there was bilateral fusion of the proximal radius and ulna. The middle phalanx of both fifth digits was hypoplastic. The entire right thumb and distal phalanx of the left thumb were absent.

At 31 days the weight was 2,500 g, head circumference 33 cm, length 41 cm, and chest circumference 29 cm. Repeated blood counts and urinalyses were normal. Platelets were normal. Spinal fluid, serum electrolytes, total protein, albumin, glucose, bilirubin, serum alkaline phosphatase, protein-bound iodine, and blood urea nitrogen were normal. His fetal hemoglobin decreased from 4.2% at four months to 3.1% at seven months. X rays of the chest and lower extremities were normal. An intravenous pyelogram, flat plate of the abdomen, electrocardiogram, and electroencephalogram all revealed no abnormalities. At 18 months the teeth showed enamel defects, and the front upper incisors protruded at an angle to each other. Death occurred at 20 months. No further details are available.

Case 2 was a 21-month-old white male with severe psychomotor retardation. He was the first child of a 27-year-old mother and a 29-year-old father, this mating having produced no offspring during the preceding seven years. He was born after 42 weeks gestation, weighing 2,750 g, with a head circumference of 31 cm and height of 50.5 cm. There was microcephaly, trigonocephaly, a broad prominent nasal bridge, hypertelorism, antimongoloid slant to the palpebral fissures, a short webbed neck, slight micrognathia, and a low-set malrotated right ear (fig. 2). A meningocele at the junction of the parietal and occipital bones had a diameter of .75 in, with the skin intact. A skull X ray showed a very small anterior fontanel and open coronal sutures. There was a bifid scrotum with palpable testes, perineal hypospadius and chordee, and imperforate anus with anoperineal fistula. At two days an EEG showed no major asymmetries. An ECG showed right sinus arrhythmia and right axis deviation. A heart murmur was first noted at one week of age. At birth, serum electrolytes were within the normal ranges.

X rays showed deformities of the sacrum, coronal cleft vertebrae of the lumbar spine, bilateral coxa valgus, and a right pelvic kidney. The phalanges of the hands and feet were longer than normal, the fingers hyperextendable. There was mild talipes equinovarus, the second and fourth toes overrode the third toe, and the nail on the left great toe was hypoplastic.

At 21 months the weight was 11.2 kg and the head circumference 46 cm. The teeth

showed enamel defects, and the front upper incisors protruded at an angle to each other. Since birth the facial asymmetry had become more severe, with marked hypoplasia of the left side. There was good head control, but he rarely turned over and could not crawl, sit, or stand.

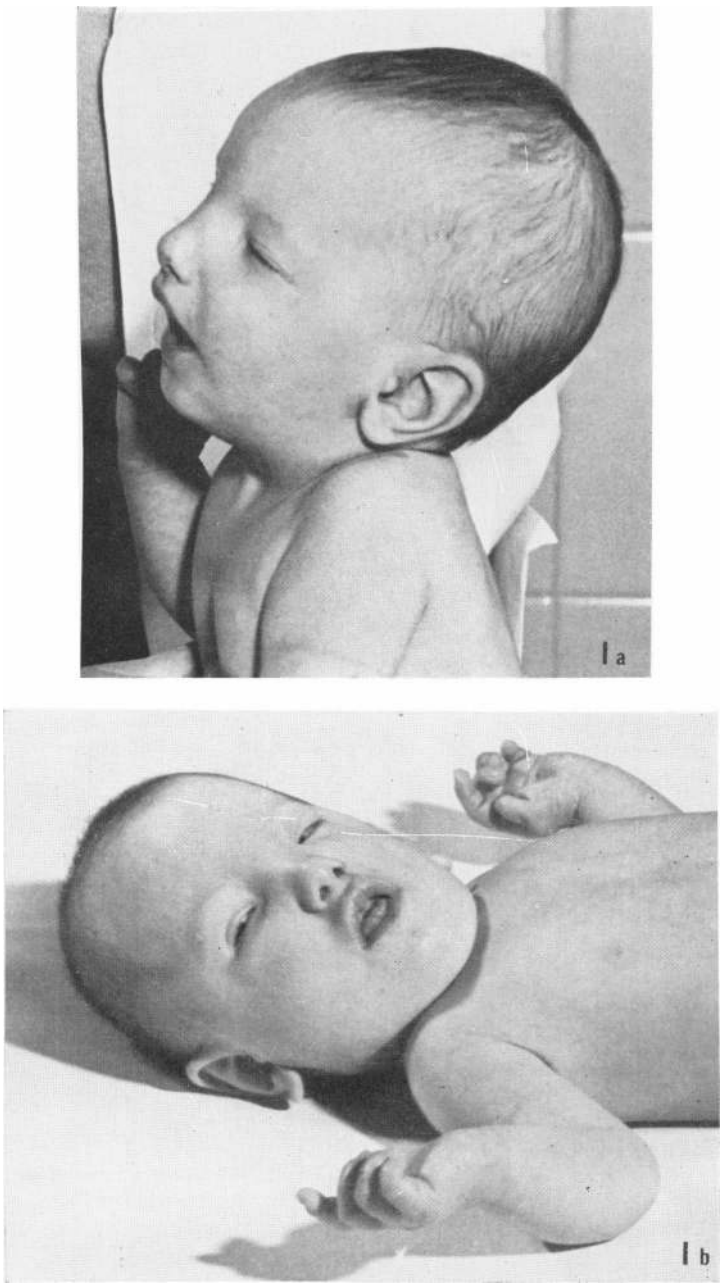

FIG. 1.—Case 1. 46,13q−

GENETIC MARKER STUDIES

Case 1 is heterozygous at the Rh, MN, Duffy, and Gm loci. He and his father have type 2-2 haptoglobin, while his mother's haptoglobin is 2-1. He and his mother are type O, while his father is type A. He is P-negative, both parents are P-positive. He has Gc type 1-1; both parents are type 2-1. Tests for Kell, Lewis, Lutheran, Inv, phosphoglucomutase, adenylate kinase, transferrin, and hemoglobin were uninformative.

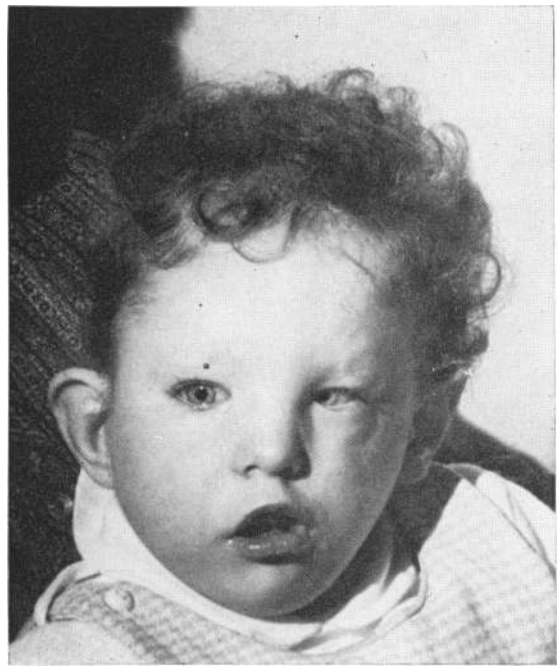

FIG. 2.—Case 2. 46,13r

Case 2 is heterozygous for MN, Duffy, and phosphoglucomutase. He and his mother have type 2-2 haptoglobin, while his father's haptoglobin is 2-1. He is Rh-negative, Jk(a−), and Gc type 2-2, while both parents are Rh-positive, Jk(a+), and Gc 2-1. Gm and Inv were not done; ABO, P, Kell, Lutheran, transferrin, and Ag were uninformative.

CYTOGENETIC STUDIES

The sex chromatin of the proband and parents in both cases was consistent with the apparent sex.

Karyotype analysis for case 1 was carried out with cultured cells from three separate blood samples and a skin biopsy. The karyotype was 46,XY,Dq−. Measurements showed that 20% of the long arm had been deleted. The deleted chromosome was identified autoradiographically as a chromosome 13 (fig. 3; see Appendix). A blood sample from each parent cultured in the usual manner showed normal karyotypes, normal count distribution, and no increase in break frequencies.

Leukocytes from case 2 were cultured from two separate blood samples. The karyo-

type was 46,XY,Dr. The ring chromosome was identified as a chromosome 13 by autoradiography (fig. 3; see Appendix). The karyotype of the father was normal. A successful culture was not obtained from the mother.

DISCUSSION

Clinical data on our cases 1 and 2, together with comparable observations on 21 other 46 Dq− and 46 Dr patients, are listed and given a case number in table 1. Re-

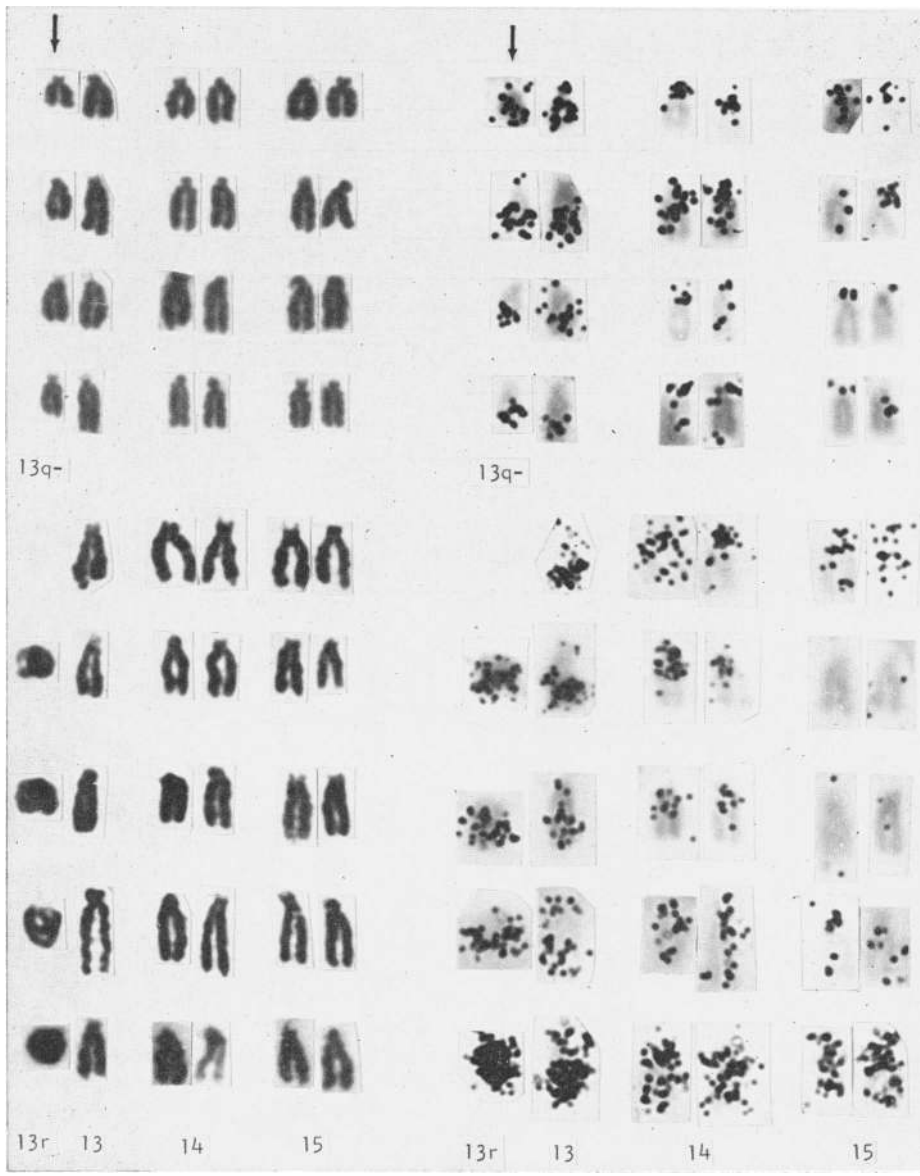

FIG. 3.—Partial karyotypes showing terminally labeled D-group chromosomes from case 1 and case 2.

TABLE 1

CLINICAL FEATURES COMMON TO 46 Dq− AND 46 Dr CASES

FEATURES	CASES*																					
	Cases with a Dq− Chromosome							Cases with a Ring D Chromosome														
	1	3	4	5	6	7	8	9	2	10	11	12	13	14	15	16	17	18	19	20	21	22	23
Autoradiographic identification	13	13	13					14	13	13	13	13	13	13									
Psychomotor retardation	+	+			+		+	+	+	+	+	+	+		+	+	+	+	+	+	+	+	+
Microcephaly	+				+		+	+	+	+	+	+	+		+	+	+	+	+		+	+	
Trigonocephaly (arrhinencephaly)	+		+		+	+	+	+	+	+					+	+	+	+		(+)	+	+	
Broad prominent nasal bridge	+						+	+	+	+		+		+	+	+	+						
Hypertelorism	+			+		+	+	+	+	+					+	+	+	+	+				
Microphthalmus	+				+	+	+	0	+	0					+				+	+			
Epicanthus	+			+				+	+	0		+	+			+	+	+					+
Ptosis	+	+					+	+	+	0								+					
Coloboma of the iris	+		+		+		+	+	0	0						+							
Retinoblastoma	0	+			+			0	0	0													
Micrognathia	+			+	+	+	+	+	+	+								+					+
Protruding upper incisors (maxilla)	+	(+)						0	+	+					+	+	+						
Short neck (lateral neck folds)	+(+)					+(+)		+(+)	+(+)	(+)	+(+)												
Low set (malformed) ears	+	+		+(+)	(+)			+	+	0					(+)	(+)	(+)	(+)			+	+	(+)
Large (malrotated) ears	+(+)							+(+)	+(+)	0	+	+			+	+	+	+(+)	+				+
Facial asymmetry	+							0	+	+		+		+									
Congenital heart disease (murmur)	+			+		(+)	+	+(+)	(+)	0		+	(+)			(+)		+		+			
Imperforate anus	0		+				+	+	+	0	+												
Perineal fistula	0		+				+	+	+	0													
Hypospadias (epispadias)	(+)		+		+	♀	♀	+	+	♀	+	♀	♀							♀		+	
Undescended testes (bifid scrotum)	+	+	(+)		+			+(+)	(+)	♀	+			+	+	+					+		
Pelvic girdle anomalies	?				+		+	+	+	0	+				+	+		+			+		+
Foot and toe anomalies	+			+	+		+	+	+	+			+		+	+	+	+	+	+			
Thumb absent (hypoplastic)	+(+)		(++)		(++)		(++)	++	0	0					+	+		++		+(+)			
Fifth finger short	+			+			+	+	0	0			+								+		
Fourth finger short	0			+				0	0	0													

* 1: Our case 1; 2: our case 2; 3: Gey (1967, personal communication [1968]); 4: Hollowell (personal communication [1969]); 5: Mikelsaar (1967); 6: Thompson and Lyons (1965); 7: Case GH 051267/4308, Taylor (1968); 8: Laurent et al. (1967); 9: Sparkes et al. (1967); 10: Stimson and Hecht (personal communication [1968]); 11: Reisman et al. (1965); Bloom et al. (1967); 12: Hollowell et al. (1967); 13: Gerald et al. (1967); 14: Neibuhr and Mikkelsen (personal communication [1968]); 15, 16, 17: Cases 1, 2, 3, Lejeune et al. (1968); 18: Adams (1965); 19: Oishi (personal communication [1968]); 20: Bain and Gauld (1963); 21: Jacobsen (1966); 22: Teplitz et al. (1967); 23: Wang et al. (1962).

gardless of the specific D-group chromosome abnormality, whether a ring or a deletion involving only part of the long arm, salient clinical features repeat from case to case. The descriptions are most complete for cases 1, 2, 8, 9, and 10. The facial dysmorphy is unique. The patients have microcephaly, trigonocephaly, micrognathia, large malformed ears, and a wide prominent nasal bridge together with hypertelorism, microphthalmia, and ptosis. Facial asymmetry was very striking in our cases 1 and 2 and was noted also in cases 10, 12, and 14. Case 2 developed much of his asymmetry postnatally. Increasing hypoplasia of the left maxillary alveolar ridge with age was observed in case 12. The right ear alone was malrotated in cases 2, 9, and 18.

A protruding maxilla was noted in case 3, while the upper incisors protruded and were set at an oblique angle to each other in cases 1 and 2. Identical tooth eruption was described in cases 15, 16, and 17 and called typical of the "Dr" syndrome by Lejeune et al. (1968). Stimson and Hecht described case 10 as having "buck teeth." She had lateral neck folds, while cases 1, 2, 7, 9, and 11 had a short webbed neck.

Eye morphology and function were affected adversely in many of the patients. Eight cases had microphthalmia, four had a cataract, six had coloboma of the iris, and one of the latter also had coloboma of the retina. Retinoblastoma developed in three cases with a Dq− chromosome. In one of these cases, no congenital anomalies were observed (Lele et al. 1963).

Absent or cryptorchid testes were common among these patients. Our case 1 had epispadias, while varied degrees of hypospadias occurred in cases 2, 4, 6, 9, 11, and 22. Pelvic-girdle and lower-spine anomalies were seen in X rays of 10 patients. An imperforate anus and perineal fistula were present in cases 2, 4, 8, and 9. The anus was imperforate in case 11.

Thumb aplasia was singled out by Sparkes et al. (1967) as the most striking manifestation of a unique "14r" syndrome. Thumb anomalies have been observed in seven of the 46 Dr and 46 Dq− cases, including our case 1, which clearly has a deletion involving chromosome 13. Congenital heart disease occurred in seven cases. The association of congenital heart disease with limb anomalies is common to many genetically determined conditions, for example, the Holt-Oram syndrome (Zetterqvist 1963), a similar dominant disease (Lewis et al. 1965), and Fanconi's pancytopenia (McDonald and Goldschmidt 1960). Their joint occurrence in patients with partial deletion of a D-group chromosome may point to a disturbance of a common embryological mechanism rather than indicate that a specific gene locus has been deleted.

It has been suggested that the structural locus for the haptoglobin gene is located near one end of chromosome 13 (Bloom et al. 1967; Gerald et al. 1967). Cases 11 and 13, each with a ring 13 chromosome, showed anomalous haptoglobin inheritance. Neither of our cases was heterozygous at this locus, although each had one heterozygous parent.

Because of the clinical similarity among the five most completely described 46 Dq− and 46 Dr cases, we postulate that these represent one distinct chromosome-deletion syndrome. Characteristic features of this syndrome are microcephaly, trigonocephaly, a broad prominent nasal bridge, hypertelorism, microphthalmus, epicanthus, ptosis, protruding upper incisors, micrognathia, a short neck with lateral neck folds, large prominent low-set ears, with possible facial asymmetry, imperforate anus or perineal

fistula, and possible hypoplastic or absent thumbs. Since both a ring D chromosome and a Dq− chromosome may be presumed to have lost a distal segment of the long arm, both types of case with this syndrome may be monosomic for the same segment of one D-group chromosome. Autoradiographic studies on nine cases have shown this to be a chromosome 13.

SUMMARY

The present paper reports almost identical clinical findings in two patients; the first has a deletion from the distal segment of the long arm of a chromosome 13, the second has a ring chromosome 13. Strong similarity between our cases and the most completely described cases with Dq− or Dr chromosomes studied in other laboratories has led us to postulate the existence of a distinct chromosome 13q− (including 13r) deletion syndrome.

In the presence of partial monosomy for a distal segment of chromosome 13, the facial dysmorphy is unique: microcephaly, trigonocephaly, protruding upper incisors, micrognathia, large malformed ears, a broad prominent nasal bridge, hypertelorism, and microphthalmus are present. Marked facial asymmetry has also been observed. In some cases there are hypoplastic or absent thumbs; in some there is an imperforate anus and perineal fistula.

ACKNOWLEDGMENTS

We wish to thank Dr. Jerry Jacobs for referring case 2 to us and Dr. H. Ranney for hemoglobin studies on case 1.

APPENDIX

AUTORADIOGRAPHIC STUDIES

Case 1

Cultured leukocytes from case 1 were terminally labeled. Tritiated thymidine (specific activity 1.9 c/millimole) was added at approximately 72 hr incubation at a final concentration of 0.5 μc/milliliter to the first culture, and 0.1 μc/milliliter to the second culture. Colcemid was added 4.5 hr later, and the cultures terminated 6.5 hr after the initial addition of the labeled thymidine. Slides were prepared and stained with Giemsa. Cells were photographed; restained with aceto-orcein; coated with AR 10 stripping film, using the technique of Schmid and Carnes (1965); and exposed in the dark at 4° C for 7, 10, or 27 days.

The autoradiographic label over the D-group chromosomes was analyzed by grain-count distribution in a total of 74 labeled cells from the two cultures (table A1). At the microscope, counts were made over each D-group chromosome: "P" over the short arm and proximal half of the long arm, and "D" over the distal half of the long arm. The grain counts were then standardized by multiplying the ratio of the mean grain count over the D-group chromosomes in all labeled cells to the count in each individual cell.

In order to distinguish chromosomes 13, which terminate DNA synthesis latest in the distal portion of the long arm, from chromosomes 14 and 15, in which this region is not late replicating, the five normal D-group chromosomes were ranked within each cell by the grain count over their distal segment. The frequency distribution of counts for each rank was graphed (fig. 4). There was a sharp distinction between the distribution for one chromosome with the heaviest distal label and four with lighter distal labeling. Consequently, the deleted chromosome was identified by exclusion as a chromosome 13.

Assuming that homologous chromosomes in the D-group replicate synchronously, the Wilcoxon matched-pairs signed-ranks test was used to test the significance of the differences in grain count between the proximal half segments of the chromosomes matched by pattern

TABLE A1

AUTORADIOGRAPHIC DATA, CASE 1

	First Culture		Second Culture	Total
^{3}H-thymidine deoxyriboside final concentration	0.5 μc/ml		0.1 μc/ml	
Length of exposure	7 days	10 days	27 days	
Cells photographed	23	40	57	120
Cells labeled	22	38	55	115
D-group grain count >28	22	36	16	74
Cells scored by pattern	20	31	16	67
Pattern over the Dq− chromosome:				
D_1	11	27	15	53
D_1 or D_2	4	3	1	8
D_2	0	0	0	0
D_2 or D_3	2	0	0	2
D_3	3	1	0	4

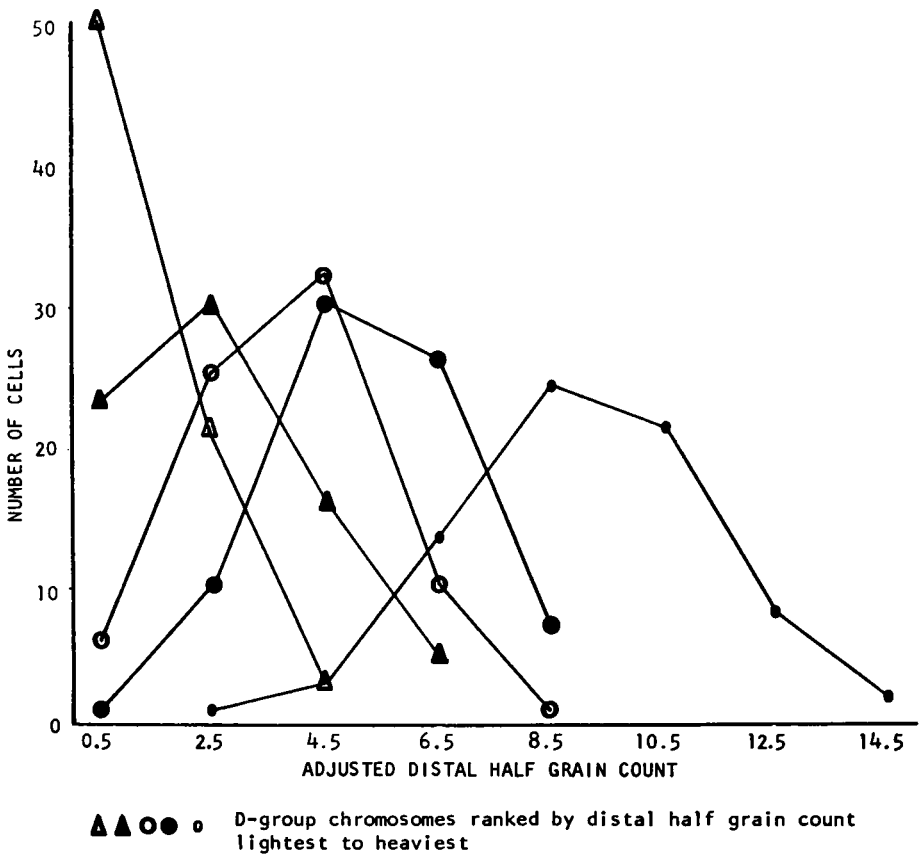

FIG. 4.—Case 1. Distribution of adjusted distal half-grain counts over the five normal D-group chromosomes.

(Giannelli and Howlett 1966). This test was used on the data from all 31 cells informative by pattern from the slides exposed 10 days. The grain counts over the D-group chromosomes are listed in table A2. Chromosomes 14 were randomly assigned to the classes D_2 and D_2' and chromosomes 15 to D_3 and D_3'.

TABLE A2

GRAIN COUNTS AND PATTERNS OVER THE D-GROUP CHROMOSOMES IN 31 CELLS FROM CASE 1

Cell No.	Chromosome											
	Dq−		13			14				15			
			Pattern D_1			Pattern D_2		Pattern D_2'		Pattern D_3		Pattern D_3'	
	Segment		Segment			Segment		Segment		Segment		Segment	
	P*	D†	P	D‡		P	D	P	D	P	D	P	D
				.30	.20								
1	9	14	7	12	3	15	9	14	4	9	4	10	3
2	6	13	6	15	4	17	7	17	9	11	10	13	6
3	4	4	2	4	1	4	0	3	1	1	2	2	0
4	8	10	11	8	5	13	9	9	6	6	7	4	2
5	4	7	5	6	3	6	4	10	0	6	5	5	6
6	1	14	8	8	5	8	3	8	7	4	5	9	1
7	2	8	2	5	5	8	1	7	2	3	3	0	1
8	4	5	2	5	4	5	3	5	3	6	0	5	3
9	6	3	6	4	4	9	3	8	2	6	5	3	5
10	1	4	2	8	3	8	5	4	6	4	1	4	1
11	2	3	5	2	3	4	1	4	2	4	0	1	0
12	6	8	4	3	3	8	5	8	4	7	6	6	4
13	8	13	7	9	4	10	10	10	8	11	5	7	6
14	6	10	7	7	3	9	8	6	7	6	7	6	6
15	2	4	3	6	1	5	4	7	6	4	0	4	2
16	5	5	2	4	3	4	2	8	1	2	3	4	2
17	7	3	4	5	3	9	1	3	7	4	6	7	1
18	0	6	2	5	5	5	4	6	4	0	0	0	2
19	4	5	6	3	5	9	3	8	4	6	0	7	2
20	4	6	3	4	2	5	4	3	3	0	1	3	1
21	1	5	4	4	3	3	3	6	0	1	0	2	1
22	6	12	10	11	7	10	14	12	5	10	2	6	8
23	2	6	7	1	5	2	6	4	3	4	5	3	4
24	1	12	0	8	3	10	5	10	0	5	5	2	6
25	7	8	4	5	3	8	6	7	6	2	4	6	1
26	1	5	1	5	3	8	3	6	3	3	6	4	0
27	5	14	7	9	4	11	7	9	10	10	5	7	6
28	1	6	2	8	1	5	7	6	6	5	1	4	2
29	3	7	6	5	3	7	4	7	4	6	2	6	2
30	5	8	7	7	3	7	7	8	4	8	3	5	4
31	2	9	6	7	2	7	7	8	2	4	7	4	8
Subtotal	123	237	148	193	106	239	155	231	129	158	110	149	96
Total	360		447			394		360		268		245	

* P = proximal half of chromosome (including centromere and short arm).
† D = distal half of chromosome.
‡ D.20 = distal 20% of chromosome; D.30 = distal 30% of chromosome.

There was no significant difference in grain count between the proximal segments of the two chromosomes 14, indicating that these presumptive homologues were well matched. There was a significant difference in count over the proximal half of the deleted chromosome and one randomly chosen chromosome 14, indicating that these segments were nonhomologous and that the deleted chromosome was probably not a chromosome 14. There was no significant difference in grain count between the proximal half of the deleted chromosome and that of the normal chromosome 13, confirming that these segments were homologous. The distal half of the deleted chromosome had 18% fewer grains than the distal half of the normal chromosome 13. This difference was significant at the 5% level with the Wilcoxon test. However, the deleted chromosome was about 20% shorter than its presumed homologue, suggesting that the deletion was from the late-replicating segment.

TABLE A3

AUTORADIOGRAPHIC DATA, CASE 2

^{3}H-thymidine deoxyriboside final concentration	0.5 μc/milliliter
Length of exposure	8 days
Cells photographed	32
Cells labeled	31
Cells scored by grain count	31
Cells scored by pattern	30

PATTERN FREQUENCY OVER THE 5 NORMAL D-GROUP CHROMOSOMES

FREQUENCY OF PATTERN PER CELL					NO. OF CELLS
D_1	D_1 or D_2	D_2	D_2 or D_3	D_3	
1	.....	2	.....	2	26
1	1	1	.....	2	2
1	.....	1	2	1	2
					30

In spite of the deletion from the late-replicating segment, the pattern over the deleted chromosome was consistent with that observed over the long arm of the normal chromosome 13 in 79% of the 67 cells informative by pattern (table A2). In none of the cells was the pattern definitely that of a chromosome 14, while in only 3% was it compatible with that of a chromosome 15.

To test the hypothesis that a single break leading to the deletion from the distal segment was not involved, the grain count over the distal 20% of the long arm of each normal chromosome 13 was subtracted from the count over the distal half of the same chromosome (table A2). The resulting total grain count of 341 was not significantly different from the deleted chromosome total of 360 grains. We conclude that a terminal deletion cannot be ruled out.

Grain patterns over the five normal D-group chromosomes in the 16 labeled cells from the second culture were analyzed separately. In 15 cells, there were definitely one chromosome 13, two chromosomes 14, and two chromosomes 15. In these cells the deleted chromosome could be identified by exclusion alone as a chromosome 13.

Case 2

Tritiated thymidine (specific activity 1.9 c/millimole) was added after approximately 72 hr incubation to a final concentration of .5 μc/milliliter. Colcemid was added 4.5 hr later and the culture terminated 6.5 hr after the initial addition of labeled thymidine. Slides

were prepared and stained with Giemsa. Cells were photographed, then restained with aceto-orcein, coated with AR 10 stripping film, and exposed in the dark at 4° C for eight days.

Five normal D-group chromosomes in 30 labeled cells were identified by pattern (Giannelli and Howlett 1966). In 26 cells, there was definitely only one chromosome 13, two chromosomes 14, and two chromosomes 15 (table A3). By exclusion then, the ring chromosome arose from a chromosome 13. Further evidence that this was a ring 13 chromosome came from quantitative evaluation of the grain counts from all 30 labeled cells. Grain counts for the ring and the five normal D-group chromosomes are listed in table A4. The chromosomes 14

TABLE A4

CASE 2. GRAIN COUNTS AND PATTERN OVER THE RING AND SEGMENTS OF THE D-GROUP CHROMOSOMES

CELL NO.	CHROMOSOME										
	Ring	13		14				15			
		Pattern D_1		Pattern D_2		Pattern D_2'		Pattern D_3		Pattern D_3'	
		Segment		Segment		Segment		Segment		Segment	
		P*	D†	P	D	P	D	P	D	P	D
1	22	3	10	11	9	6	6	6	3	3	2
2	27	9	15	9	6	8	7	4	3	8	6
3	22	9	10	15	6	10	6	11	3	5	0
4	.....	9	12	14	5	13	9	6	6	10	5
5	41	17	24	19	12	15	13	13	13	13	7
6	21	10	12	9	6	9	9	4	3	9	2
7	11	3	5	7	3	6	3	4	3	3	2
8	20	8	13	11	5	13	8	9	4	10	5
9	35	13	21	13	11	12	9	12	9	12	7
10	25	5	16	8	4	7	9	7	3	6	5
11	16	6	10	9	8	7	7	10	0	3	1
12	18	7	12	5	7	10	5	5	4	4	6
13	21	11	10	13	6	10	5	7	5	3	1
14	.....	8	22	11	11	16	10	11	6	16	3
15	33	10	15	15	8	13	10	11	6	1	1
16	40	5	17	14	2	13	9	2	2	3	2
17	18	12	20	19	10	11	8	12	6	7	2
18	37	16	29	27	13	22	19	11	9	15	18
19	33	6	12	2	2	3	1	1	1	0	0
20	35	8	10	17	2	15	5	4	4	10	8
21	24	12	16	12	11	16	4	10	7	8	2
22	37	15	19	16	8	16	11	4	5	10	4
23	42	20	20	24	19	25	18	21	15	17	9
24	31	20	29	26	11	26	9	15	11	22	12
25	.....	13	23	12	10	15	11	13	7	6	6
26	40	17	28	16	13	13	10	7	3	6	5
27	29	16	34	21	18	27	17	9	7	9	5
28	34	13	20	13	9	13	9	4	6	6	3
29	33	16	19	12	7	14	7	5	1	6	2
30	30	10	18	11	9	11	9	10	2	9	0
Subtotal	.....	327	521	411	251	395	263	248	157	240	131
Total	775	848		662		658		405		371	

* P = proximal half of chromosome (including centromere and short arm).

† D = distal half of chromosome.

were randomly assigned to classes D_2 and D_2' and chromosomes 15 to D_3 and D_3'. The ring chromosome was missing from three cells. The ring chromosome averaged 28.1 grains in 27 cells, while the normal chromosome 13 averaged 28.3 grains in 30 cells.

To obtain sharper discrimination among the five normal D-group chromosomes, these were ranked by the grain count of their distal half without regard to pattern identification. The grain counts were standardized as in case 1, and the frequency distribution for each rank was graphed. The histogram obtained was very similar to that of case 1 (fig. 5). There

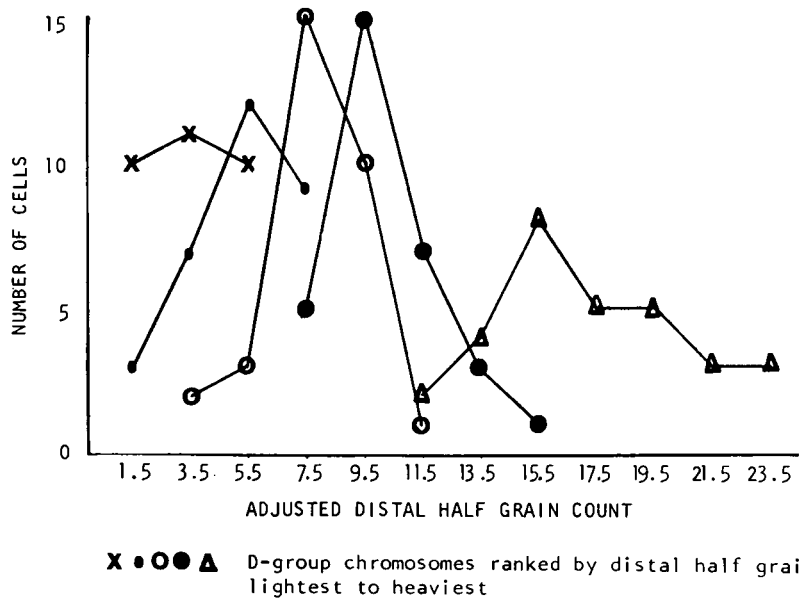

FIG. 5.—Case 2. Distribution of adjusted distal half-grain counts over the five normal D-group chromosomes.

was a distinct separation between the first, or heaviest-ranking, chromosome and the four chromosomes with a lighter distal label. Since there was only one distribution typical of a chromosome 13, we conclude that the ring chromosome originated from the missing chromosome 13.

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