UNEXPECTED FINDINGS WITH THE NEW CHROMOSOME BANDING TECHNIQUES IN A PATIENT FORMERLY DIAGNOSED AS HAVING G-DELETION SYNDROME II

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A girl is described, who, upon her first admission to our pediatric department in 1965, was supposed to have G-deletion syndrome II on the base of the chromosome findings in routine orcein-stained preparations and her clinical aspect. When, however, in 1972 a Q- and G-banding analysis was performed, the patient seemed to be a t(14q+; 22q—) translocation carrier.

These findings are discussed in relation to the known heterogeneity of the clinical picture of patients previously reported as having a G-deletion syndrome and in whom no banding studies were done.

Finally, the need for more extensive studies with the different techniques is stressed, particularly, in cases of Gq— chromosomes, of which the familial occurrence is suggestive of a hidden reciprocal translocation.

INTRODUCTION

Chromosome aberrations can be recognized more accurately by means of the modern banding techniques than with the classic staining techniques. For instance, a 54-year-old mentally retarded woman, originally considered as having a presumptive monosomy 21 on the base of a routine chromosome analysis, was found with the new banding techniques to have a 4q/21q translocation (Dutrillaux et al. 1973). A more or less similar case will be discussed here. A girl, first diagnosed as having G-deletion syndrome II on the base of routine orcein-stained mitoses, was found later on, with the banding techniques, to have a t(14q+; 22q—) translocation.

CASE REPORT

The proband, born on 1st May 1965, was the second child of a nonconsanguineous mating; the father was 38 and the mother 31 years old at the time of the child's birth. The pregnancy and confinement were normal. The parents and the older child, a 9-year-old girl, are healthy. Before the birth of the patient, the mother had two spontaneous abortions, both at 2½ months. The family history is negative, no other instances of mental retardation or congenital malformations are known.

In the neonatal period there were no feeding difficulties or other complications. The psychomotor development was said to have been normal up to the age of 4 months: the child was very active and lively, smiled at her parents and could hold her head upright. Afterwards, however, the patient became more and more apathetic. She no longer smiled at her parents and she was very flabby, lying down passively without moving her arms and legs. At the age of 4½ months, for the first time the patient had seizures, which occurred as follows: usually before falling asleep, the child became

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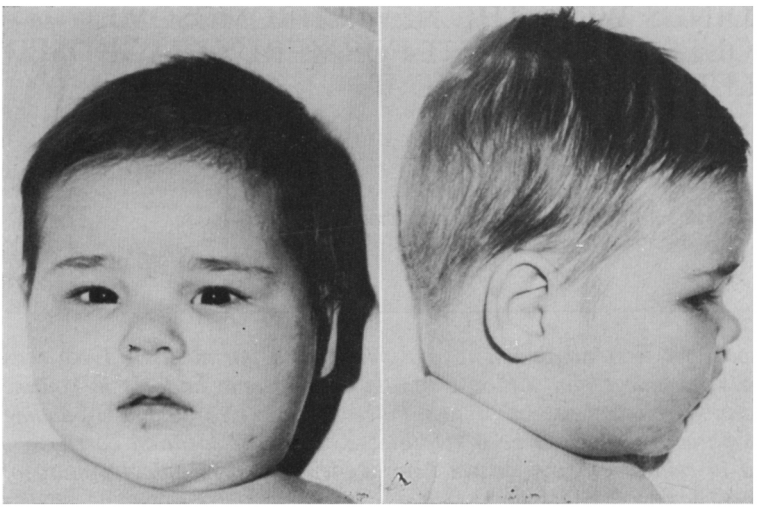

Fig. 1. The patient: frontal and lateral view.

pale, her eyes turned away, she showed convulsive movements of the arms and legs and she was unconscious for several minutes. She had these seizures at regular intervals of 15 minutes until she was asleep.

Because of her psychomotor retardation and the seizures the patient was admitted to our department on 23rd November 1965.

The physical examination revealed a well-developed, dull-appearing girl (Fig. 1). Her weight was 8850 g (P90), her length 76 cm (> P97) and head circumference 44 cm (P50). The psychomotor retardation was obvious. The patient was very quiet and apathetic, she could not hold her head upright, nor sit unsupported. She didn't smile and, although she followed with her eyes, she showed little interest in the things around her. When lying down, a spontaneous flexion of the arms and an extension and abduction of the legs were seen. Also, she exhibited intermittent choreoathetotic movements with symmetrical rotation of the wrists and with subtile movements of the fingers, alternating with flexion and extension of the legs. Only minor congenital anomalies were seen. There was a high forehead, a wide-open fontanel and a flat occiput. The hair implantation was normal. The face was expressionless and exhibited noncontinuous eyebrows, a slight hypertelorism, an epicanthal fold of the right eye, a broad and flat nasal bridge, and a slight prognathism. The ears were soft, but had a normal implantation. The neck was short and thick with an additional transverse fold. The chest was flat with wide-set nipples. The hands showed tapering fingers with cushions at the proximal phalangeal joints, while no such cushions were seen on the feet. A supplementary fold of the buttocks was present.

The clinical investigation of the cardiovascular and urogenital systems revealed no anomalies. The lung auscultation and abdominal examination were normal. Neither liver, nor spleen was palpable. The tendon reflexes were normal and symmetrical, and the Babinsky reflex was absent.

Central Investigations

The ophthalmological investigation revealed a slight internal strabismus. X-rays of the skull were normal. The electroencephalogram was intermittently normal and pathological and indicative of diffuse disturbances in the electrogenesis. A pneumoencephalography revealed signs of a diffuse, cerebral atrophy, while an electromyogram was indicative of a diffuse neurogenic affection.

Laboratory Findings

Normal values for glucose and albumine and normal cell count were found in the cerebrospinal fluid. A total blood count was normal. The bone marrow revealed a hypoplasia of the normoblasts.

TABLE 1

DERMATOGLYPHIC ANALYSIS OF THE PROBAND, HER PARENTS AND SISTER

Finger pattern	Left hand V	IV	III	II	I	Right hand I	II	III	IV	V	TFRC
Proband	U 7/0	U 18/0	U 13/0	R 0/12	W 25/23	W 17/22	R 0/13	U 9/0	W 23/7	U 18/0	160
Father	U 20/0	W 26/5	U 21/0	W 13/22	W 24/22	W 23/20	R 0/23	W 20/18	W 28/23	U 19/0	226
Mother	U 18/0	W 21/8	U 7/0	U 8/0	U 10/0	U 10/0	U 3/0	R 0/2	U 17/0	U 18/0	114
Sister	U 24/0	W 39/18	W 31/19	W 15/23	W 26/15	W 27/19	W 13/24	W 21/18	W 29/22	U 17/0	260

Palm pattern		Palm formula	atd angle	ab count
Proband	L	9 . 7 . 5″ . 4 — t → 13 — A^u . 0 . 0 . 0 . L	42	38
	R	11 . 9 . 7 . 5′ — t → 13 — A^u . 0 . 0 . L . 0	44	32
Father	L	11 . 7 . 7 . 3 — t → 13 — A^u . 0 . 0 . 0 . L	38	46
	R	11 . 9 . 7 . 4 — t → 13 — A^u . 0 . 0 . L . 0	39	42
Mother	L	9 . 7 . 5″ . 1 — t → 11 — A^u . 0 . 0 . 0 . L	41	49
	R	11 . 9 . 7 . 4 — t → 13 — A^u . 0 . 0 . L . 0	42	41
Sister	L	7 . 5″ . 5″ . 3 — t → 13 — A^u . 0 . 0 . 0 . L	37	43
	R	11 . 7 . 7 . 4 — t → 13 — A^u . 0 . 0 . 0 . L	38	43

Proband: left and right hallucal area = large distal loop.

The serological determinations of calcium, phosphor, alkaline phosphatase, lipids, cholesterol, electrolytes ad aminoacids were normal. The urinanalysis and urinary metabolic screening tests were normal.

The Dermal Ridge System

Table 1 summarizes the dermatoglyphic findings in the proband, her parents and sister. Except for a high total finger ridge count in the patient, her father and sister, no peculiar features were noted. The dermal creases in the proband and her relatives were normal.

Chromosome Studies

Chromosome preparations were obtained from short-term peripheral blood cultures, established in December 1965. A total of 45 mitoses was counted. There were 46 chromosomes of which two in the G group were abnormal (Fig. 2). The first abnormal chromosome was a variant with an elongated short arm. In less contracted mitoses this seemed to be due to enlarged and more isopyknotic satellites. The second abnormal chromosome showed a distinct deletion of the long arm. It was

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comparable to the Philadelphia chromosome in chronic myelogenous leukemia. At that time no other abnormal chromosomes were seen. The mother and sister had normal karyotypes. In the father, however, the same G variant as in the patient was seen.

A second blood sample was obtained from the patient in January 1972 and an analysis of the

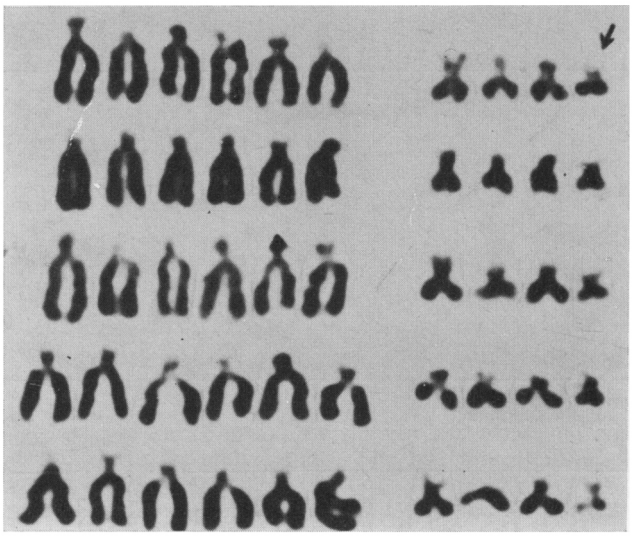

Fig. 2. Partial karyotype of the D and G group chromosomes of the patient. The 22q- chromosome is indicated by an arrow.

autoradiographic labelling pattern, as well as an analysis of the G-banding (Seabright 1971) and Q-banding patterns (Caspersson et al. 1970*b*) were performed.

In the autoradiographic analysis only 20 mitoses with an adequate labelling were found. In fifteen of them the deleted and one of the normal G chromosomes were not, or only slightly, labelled, whereas the variant and the other normal G chromosomes were clearly more heavily labelled. Consequently, the deleted and the variant G chromosomes were identified respectively as a number 22 and 21. This was confirmed by the G-banding analysis (Fig. 3).

Surprisingly, however, in the G-banded mitoses a third abnormal chromosome was found. This was a chromosome 14, of which the lightly stained distal region q32 was enlarged. This abnormal chromosome 14 was not recognized in the routine orcein-stained preparations, probably because the elongated chromosome 14 visually did not significantly exceed the normal chromosomes 13 in length. Instead of a partial monosomy 22, as was first supposed, the patient had a balanced translocation, which according to the banding patterns most probably was a t(14; 22) (q32; q11) reciprocal

Fig. 3. The normal and abnormal chromosomes 14 and 22 of six G-banded mitoses of the patient.

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translocation. The Q-banding analysis confirmed the foregoing findings and, in addition, showed the enlarged satellites of the 21 variant to be intensely fluorescent.

Length and surface measurements of the long arms of the normal and abnormal D and G chromosomes (Table 2) on photographs of orcein-stained mitoses (final enlargement 3300 ×) were carried out using a Zeiss Microvideomat (the qualitative and quantitative analyser of the televised image).

TABLE 2

LONG-ARM MEASUREMENTS OF THE CHROMOSOMES IN THE G GROUP [a] AND THE D GROUP [b]

Chromosome	21_N	21_{VAR}	22_N	22q—	13_N [c]	14_N	14q+	15_N [c]
Length [d]	6.0 ± 0.8	6.5 ± 1.0	6.7 ± 0.9	4.6 ± 0.5	19.4 ± 2.7	18.5 ± 2.3	21.5 ± 3.0	16.8 ± 2.2
Surface [d]	5.6 ± 1.1	5.8 ± 0.9	6.1 ± 1.1	3.4 ± 0.7	19.4 ± 3.6	19.3 ± 4.0	22.1 ± 2.5	18.0 ± 2.8

[a] Measurements on 49 mitoses.
[b] Measurements on 28 mitoses.
[c] Mean of the two homologous chromosomes.
[d] Values of the measurements of length and surface area expressed in percentage of the sum of all length or surface measurements respectively.

In the D group the chromosomes were classified, in descending order of long arm length, as the 14q+ chromosome, the two chromosomes 13, the one normal chromosome 14 and the two chromosomes 15. Between the different normal chromosomes 13, 14 and 15, no significant differences in length and surface area were noted. The presumed 14q+ chromosome, however, was significantly larger in length and surface area than the other normal D chromosome, e.g., in respect to the larger chromosomes 13 (length, $P < 0.05$; surface area, $P < 0.02$). In the G group, the smaller in length of the two normal chromosomes was considered as the number 21 and the larger as the number 22. In respect of the long arm of the normal 22, the long arm of the 22q— shows a deletion of 31.3% in length and a deletion of 44.3% in surface area. In respect of the long arm of the normal chromosome 14 there was a 16.2% and 14.5% increase in length and surface area respectively of the long arm of the 14q+ chromosome. Finally, there is a fairly good correlation in length and surface area between the deleted segment of the 22q— and the extra segment of the 14q+ chromosome [length: 22 N—22(q—) = 2.1 and 14(q+)—14 N = 2.9; surface: 22 N—22(q—) = 2.7 and 14(q+)—14 N = = 2.8].

DISCUSSION

In 1970 Warren and Rimoin reviewed the published cases of syndromes associated with a long-arm-deleted G chromosome or a ring chromosome G. They concluded that were two distinct syndromes, i.e., the G deletion syndromes I and II, which they postulated to be due to a deleted or ring 21, respectively 22, chromosome. This was later confirmed (Crandall et al. 1972).

The clinical aspect of patients with the G-deletion syndrome II is rather inconspicuous and variable. According to Warren and Rimoin (1970) the more frequent features are mental retardation, hypotonia, epicanthus and syndactylism of toes, while the infrequent findings are ptosis, bifid uvula, microcephaly, high-arched palate, large or low-set ears, and clinodactyly.

Patients with the G-deletion syndrome I or " antimongolism " are generally more severely affected. The more consistent findings include mental retardation, microcephaly, hypertonia, antimongoloid slant, prominent nasal bridge, high-arched palate, micrognathia, large or low-set ears, skeletal malformations and growth retardation. The more variable findings include nail anomalies, hypospadias, inguinal hernia, cryptorchidism and pyloric stenosis.

At the time of Warren and Rimoin's publication, our patient was supposed to have the G-deletion syndrome II. This was based on the chromosome findings in routine orcein-stained preparations, as well as on the patient's clinical aspect. Indeed, our patient had mental and motor retardation, hypotonia and epicanthus in common with the G-deletion syndrome II. It was thought she exhibited a mild form of this syndrome.

When, however, in 1972 a G- and Q-banding analysis was performed in the proband, surprisingly a balanced t(14; 22) translocation was found. It therefore seemed that the patient's phenotype was most probably not the result of a chromosome imbalance, even though a small deletion and/or duplication of 14q or 22q chromosome material, according to the concept of *aneusomie de recombinaison* (Lejeune and Berger 1965), cannot be ruled out completely. In this respect, the patient's psychomotor and neurological evolution, and her somatic measurements, more likely indicate a non chromosomal affection.

Our, case, as well as the case of Dutrillaux et al. (1973), underlines the need of an accurate chromosome analysis, especially when small deletions or duplications, such as Gq- chromosomes seem to be the only anomaly in routine stained preparations. On the other hand they also underline the superiority of the modern staining techniques over the classical ones in analysing abnormal karyotypes. In the orcein-stained preparations of our patient, the presumed 14q+ chromosome was significantly larger than the other normal D chromosomes, as shown by the length and surface measurements. Nevertheless, the abnormal 14q+ chromosome was not recognized simply de visu being unaware of the banding results. This may be mainly attributed to the impossibility of identifying the D chromosomes individually in classically stained mitoses.

As stressed by Grosse et al. (1971), the patients with a partial or complete G monosomy, previously reported in the literature without banding studies, show a very heterogeneous clinical picture. Banding studies in these patients will probably partially reduce this heterogeneity, since other cases may turn out, in fact, to be translocation heterozygotes or even more complex chromosome aberrations, such as, for example, a ring chromosome derived from a G/G centric fusion (Orye and Craen 1974). Very important are the modern banding studies, viewed from the point of genetic counseling, in case of Gq- or other small deletions, present in normal and abnormal family member and thus suggestive of a familial reciprocal translocation. Examples of such cases are those of Ricci et al. (1970) and Day and Miles (1965). Eventually, the use of the different banding techniques is recommended in order to establish a possible, hidden translocation. Indeed, in the case of Dutrillaux et al. (1973) the 4q/21q translocation was detected only with che R-banding and not with the G- or Q-banding techniques. More or less similar cases may be those reported by Caspersson et al. (1970*a*) and Punnett et al. (1973). These authors both described a patient with a partial trisomy 22 (47,XY,(22q—)+), of which the mother also showed the 22q— chromosome (46,XX,22q—). A G- and/or Q-banding analysis, however, revealed no translocation heterozygosity in the mother. An R or even a T-banding analysis may be more appropriate in these families, as, for instance, is demostrated in the recognition of the t(9q+; 22q—) translocation in chronic myelogenous leukemia (Van den Berghe 1973).

A final remark concerns the chromosome findings in our patient and in chronic myelogenous leukemia (Rowley 1973). Both are very similar and it would be very interesting to follow up our patient to see if she, later on, will develop chronic myelogenous leukemia.

REFERENCES

Caspersson T., Hulten M., Lindsten J., Zech L. 1970*a*. Distinction between extra G-like chromosomes by quinacrine mustard fluorescence. Exp. Cell Res., 63: 240.

Caspersson T., Zech L., Johansson C., Modest E.J. 1970*b*. Identification of human chromosomes by DNA-binding fluorescent agents. Chromosoma, 30: 215.

Crandall B.F., Weber F., Muller H.M., Burwell J.K. 1972. Identification of 21R and 22R chromosomes by quinacrine fluorescence. Clin. Genet., 3: 264.

Day R.W., Miles C.P. 1965. Familial Down's syndrome with undetected translocation. J. Pediatr., 67: 399.

Dutrillaux B., Jonasson J., Lauren K., Lejeune J., Lindsten J., Petersen G.B., Saldana-Garcia P. 1973. An unbalanced 4q/21q translocation identified by the R but not by the G and Q chromosome banding techniques. Ann. Genet., 16: 11.

Grosse K.P., Böwing B., Hoffengärtner F., Reinhardt G., Sitzmann C.F. 1971. Ring G-Chromosom. Zwei kasuistische Beiträge und Literaturübersicht. Humangenetik, 12: 142.

Lejeune J., Berger R. 1965. Sur deux observations familiales de translocations complexes. Ann. Genet., 8: 21.

Orye E., Craen M. 1974. A t(21q21q) ring chromosome. Hum. Hered, 24: 253.

Punnett H.H., Kistenmacher M.L., Toro-Sola M.A., Kohn G. 1973. Quinacrine fluorescence and Giemsa banding in trisomy 22. Theoret. Appl. Genet., 43, 134.

Ricci N., Dallapiccola B., Preto G. 1970. Familial transmission of a Gq-(Ph^1-like) chromosome. Ann. Genet., 13: 263.

Rowley J.D. 1973. A new consistent chromosomal abnormality in chronic myelogenous leukemia identified with quinacrine fluorescence and Giemsa staining. Nature (Lond.), 243: 290.

Seabright M. 1971. A rapid banding technique for human chromosomes. Lancet, 2: 971.

Van den Berghe H. 1973. The Ph^1-chromosome: translocation to chromosome 9. Lancet, 2: 1030.

Warren R.J., Rimoin D.L. 1970. The G deletion syndromes. J. Pediatr., 77: 658.

RIASSUNTO

Inattesi Risultati Ottenuti con le Nuove Tecniche di Bandeggio in una Paziente Inizialmente Diagnosticata come Affetta da Sindrome II di Delezione G

Viene descritto un caso di una ragazza che, al momento della sua prima ammissione in clinica nel 1965, venne ritenuta affetta da sindrome II di delezione G sulla base del suo aspetto clinico e dei risultati dell'analisi cromosomica standard con colorazione all'orceina. Quando, tuttavia, nel 1972 venne effettuata un'analisi a bande QG, la paziente si è rivelata come una portatrice di traslocazione t(14q +; 22q —).
Tali risultati sono discussi in relazione alla nota eterogeneità del quadro clinico di pazienti precedentemente descritti come affetti da sindrome di delezione G ed in cui non siano stati effettuati studi con la tecnica delle bande.
Viene infine sottolineata la necessità di studi piú estesi con le diverse tecniche, particolarmente in casi con cariotipo Gq—, la cui ricorrenza familiare può suggerire una traslocazione reciproca nascosta.

RÉSUMÉ

Résultats Inattendus Obtenus grâce aux Nouvelles Techniques de Bandage chez une Patiente Diagnostiqée Initialement comme Atteinte de Syndrome II de Délétion G

L'on décrit ici le cas d'une jeune fille, qui, au moment de sa première admission en clinique en 1965, est atteinte de syndrome II de délétion G, jugement fondé sur son aspect clinique et sur les résultats de l'analyse chromosomique standard avec coloration à l'orcéine. Lorsqu'en 1972 une analyse à bandes QG fut effectuée, la patiente se révéla conductrice de translocation t(14q +; 22q —).
On discute ces résultats en se référant à l'hétérogénéité connue du tableau clinique de patients décrits précédemment comme atteints du syndrome de délétion G et chez lesquels n'a été effectuée aucune étude avec la technique des bandes.
On souligne la nécessité d'études plus étendues avec les différentes techniques, particulièrement dans des cas avec caryotype Gq—, dont la récurrence familiale peut suggérer une translocation réciproque cachée.

ZUSAMMENFASSUNG

Ueberraschende Ergebnisse mit den neuen QG-Bandtechniken bei einer zuerst als Fall von G-Delektionssyndrom II diagnostizierten Patientin

Ein Mädchen wurde bei ihrer ersten Hospitalisierung 1965 auf Grund des klinischen Befunds und der Ergebnisse der Standard-Chromosomanalyse mittels Orzeinfärbung als Fall von G-Delektionssyndrom II diagnostiziert. 1972, bei Anwendung der QG-Bandanalyse stellte es sich hingegen heraus, dass Pat. Trägerin einer t(14q +; 22q —)-Translokation ist.
Es folgt eine Erörterung dieser Diagnosen angesichts der bekannten Heterogenität des klinischen Bildes bei Patienten, die früher, vor Anwendung der QG-Bandtechnik, als Fälle von G-Delektions-Syndrom beschrieben worden waren.
Es wird daher die Notwendigkeit eingehenderer Untersuchungen mit verschiedenen Techniken betont, besonders bei Fällen von Gq-Karyotyp, die bei familiärem Auftreten an eine versteckte reziproke Translokation denken lassen.

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