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Down Syndrome Due to De Novo Robertsonian Translocation t(14q;21q): DNA Polymorphism Analysis Suggests That the Origin of the Extra 21q Is Maternal

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Summary

Down syndrome is rarely due to a de novo Robertsonian translocation t(14q;21q). DNA polymorphisms in eight families with Down syndrome due to de novo t(14q;21q) demonstrated maternal origin of the extra chromosome 21q in all cases. In seven nonmosaic cases the DNA markers showed crossing-over between two maternal chromosomes 21, and in one mosaic case no crossing-over was observed (this case was probably due to an early postzygotic nondisjunction). In the majority of cases (five of six informative families) the proximal marker D21S120 was reduced to homozygosity in the offspring with trisomy 21. The data can be best explained by chromatid translocation in meiosis I and by normal crossover and segregation in meiosis I and meiosis II.

Introduction

Down syndrome due to trisomy 21 is one of the most common chromosomal abnormalities in humans that is associated with mental retardation. A rare form of Down syndrome is due to de novo Robertsonian translocation t(14q;21q) and occurs with a frequency of 1.1%–3.6% of Down syndrome live borns in population studies (Mikkelsen et al. 1976, 1989; Pulliam and Huether 1986). Robertsonian translocations have been suggested to occur during oogonial/spermatogonial mitosis (Ohno et al. 1961) or meiotic prophase I (Mirre et al. 1980; Stahl et al. 1983). Cytogenetic analysis using chromosomal heteromorphisms has suggested that the de novo t(14q;21q) chromosome has originated from the mother in 11 of 12 cases in which the parental origin has been determined (Robinson 1973; Magenis and Chamberlin 1981; Mikkelsen et al. 1989).

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As part of a study of the mechanism and origin of nondisjunction in trisomy 21 (Warren et al. 1987; Antonarakis et al. 1991), we have studied eight families with Down syndrome due to de novo t(14q;21q). Analysis of chromosome 21–specific DNA polymorphisms permitted the determination of the parental origin of the extra chromosome 21q and suggested that in the majority of cases the extra 21q was probably due to chromatid translocation in maternal meiosis I and to normal crossover and segregation in meiosis I and meiosis II.

Material and Methods

Eight families with a proband with Down syndrome due to de novo Robertsonian translocation t(14q;21q) were studied. Cytogenetic analysis of metaphases from lymphocytes included QFQ and/or GTG banding. In one proband (DS2277) the cytogenetic analysis showed 28 metaphases with 46,XX, − 14, + t(14q; 21q) and two metaphases with normal karyotype 46,XX. In the other seven probands no evidence of mosaicism was detected in 15 metaphases examined. The parents of the eight probands had normal karyotypes, establishing the de novo origin of the transloca-

tion t(14q;21q) chromosome in all families. In seven probands the translocation chromosome appeared to be monocentric by cytogenetic analysis using C bands; however, in family DS2277 the translocation chromosome was dicentric.

DNA was isolated from peripheral blood leukocytes as described (Kunkel et al. 1977). Restriction-endonuclease digestions, agarose gel electrophoresis, transfer of DNA fragments to nitrocellulose filters, hybridization with radioactive DNA probes, washing, and autoradiography were performed as described elsewhere (Warren et al. 1987, 1989). The probe-enzyme combinations for the chromosome 21 markers used were (1) D21K9-*Taq*I of locus D21S13, (2) p21-4U-*Msp*I of locus D21S110, (3) pPW236B-*Eco*RI of locus D21S11, (4) Fr8-77-*Eco*RI of locus D21S82, (5) p78/2-8b-*Pst*I of the MX1 gene, (6) pMCT15-*Pst*I of locus D21S113, (7) CRI-L427-*Rsa*I of locus D21S112, and (8) ML18-*Taq*I of the COL6A1 gene (for description of the probes and references, see Warren et al. 1989; Petersen et al. 1991*b*). In addition, six different DNA polymorphisms due to either $(GT)_n$ dinucleotide repeats (Litt and Luty 1989; Tautz 1989; Weber and May 1989; Weber 1990) or a variable poly(A) tract of an Alu repeat (AluVpA) (Economou et al. 1990) were detected by using PCR amplification of genomic DNA (Saiki et al. 1985). The PCR polymorphisms were (1) a $(GT)_n$ repeat of locus D21S120 (Burmeister et al. 1990), (2) a $(GT)_n$ repeat of locus D21S156 (Lewis et al. 1990), (3) a $(GT)_n$ repeat at IVS4 of the HMG14 gene (Petersen et al. 1990), (4) a $(GT)_n$ repeat and (5) an AluVpA at IVS5 of the HMG14 gene (Petersen et al. 1991*a*), and (6) an $(AC)_n$ repeat of locus D21S171 (Petersen et al., in press). The primers used, end labeling of specific primers, PCR programs, PAGE of the amplification products, and autoradiography have been described elsewhere (Burmeister et al. 1990; Lewis et al. 1990; Petersen et al. 1990, 1991*a*, and in press). The order of the loci scored which span the entire long arm of human chromosome 21 has been determined by linkage analysis using the CEPH families (Warren et al. 1989; Petersen et al. 1991*b*) and by pulsed-field gel electrophoresis analysis using somatic cell hybrids (Gardiner et al. 1990). In order to exclude isodisomy for chromosome 14, probe pCMM101 of locus D14S13 was used. This locus recognizes a VNTR polymorphism on chromosomal band 14q32 (Nakamura et al. 1988). The parental origin of the extra 21q was determined by dosage analysis, when two different alleles were present in the proband, or by scoring the polymorphic alleles, when three different alleles were present in the proband (McCormick et al. 1989; Petersen et al. 1991*a*).

Results and Discussion

The parental origin of the additional chromosome 21q was determined in all families by at least two independent DNA polymorphisms (table 1). The origin was maternal in all eight families. Previous cytogenetic analyses of 13 published families with de novo t(14q;21q) determined the origin of the t(14q;21q) to be maternal in 11 cases, and paternal in one case, and unknown in one case. In this latter analysis chromosomal heteromorphisms of the short arm of the free chromosomes 14 and 21 have been compared with those of the parental karyotypes. The origin of the t(14q;21q) has been determined because the free chromosome 14 had the same chromosomal heteromorphism pattern as did one of the parental chromosomes 14 (Robinson 1973; Magenis and Chamberlin 1981; Mikkelsen et al. 1989). However, the reliability of cytogenetic heteromorphisms in determining the parental origin of acrocentric chromosomes has been recently disputed (Antonarakis et al. 1991). Cytogenetic analysis in our cases determined the origin of the free chromosome 14 in only one case (family DS3325). The free chromosome 14 in this case was paternal, and therefore the t(14q;21q) was maternal in origin.

The four possible parental origins of the t(14q;21q) chromosome and of the extra chromosome 21 (or 21q) in de novo t(14;21q) Down syndrome are shown in figure 1. The nonmosaic cases of the present study belong to the categories illustrated in panels *A* and *D* of figure 1, since the possibilities illustrated in *B* and *C* of figure 1 can be excluded because the extra chromosome 21q is not paternal. In order to exclude more complicated models in which both the t(14q;21q) and the free 14 from the same parent, we used D14S13 DNA marker polymorphism analysis. The results show that in all cases (except DS3402, which was not informative), one chromosome 14 was inherited from the father and one was inherited from the mother, thus excluding the possibility of chromosome 14 isodisomy (see table 2, DNA marker D14S13).

The genotypes for all the families are shown in table 2. Given the evidence that the extra 21q was of maternal origin in all cases, the reduction/nonreduction to homozygosity for a given DNA marker heterozygous in the mother was determined (table 2). In one family (DS2277) all seven informative loci showed reduction to homozygosity, giving no evidence of crossing-over

Table 1

Parental Origin of Extra Chromosome 21q, Parental Ages at Birth of Down Syndrome Proband, Sex of Proband, and Loci Informative for Determination of Parental Origin

Family	Origin	Maternal Age (years)	Paternal Age (years)	Sex of Proband	Loci Informative
DS2277	Maternal	30	32	F	D21S82, MX1, D21S171, D21S112, and COL6A1
DS2899	Maternal	29	29	M	D21S13, D21S110, D21S11, D21S156, HMG14, D21S113, D21S112, and COL6A1
DS3202	Maternal	26	30	M	D21S120 and D21S112
DS3325	Maternal	30	34	M	D21S156 and D21S112
DS3380	Maternal	22	23	F	D21S120, D21S156, and HMG14
DS3396	Maternal	37	47	F	D21S13, D21S82, D21S112, and COL6A1
DS3399	Maternal	22	21	M	D21S120, D21S156, D21S171, and D21S112
DS3402	Maternal	22	21	M	D21S120, D21S82, HMG14, and D21S112

between the chromosomes 21 in maternal meiosis. In this case, which is the mosaic case, both the extra 21q and the formation of the Robertsonian translocation chromosome were probably due to an early post-zygotic (mitotic) nondisjunction (fig. 2*A*). Another mechanism, which cannot be excluded by the present data, is a prezygotic origin followed by an early post-zygotic (mitotic) loss of the translocation chromosome, with duplication of the remaining chromosome 14 leading to uniparental isodisomy for chromosome 14 in the 46,XX cell line (fig. 2*B*). Chromosome 14 DNA polymorphic marker D14S13 showed two alleles of equal intensity, one coming from the father and one coming from the mother (table 2). However, with only less than 10% mosaicism for the normal cell line, differences in intensity of the polymorphic alleles cannot be observed. In the seven nonmosaic cases the genotypic data from the scoring of many markers along the entire length of 21q suggest at least one crossover between the two chromosomes 21 during maternal meiosis I. The Robertsonian translocation therefore did not influence the normal pairing and crossing-over between the chromosome 21 homologues in meiosis I.

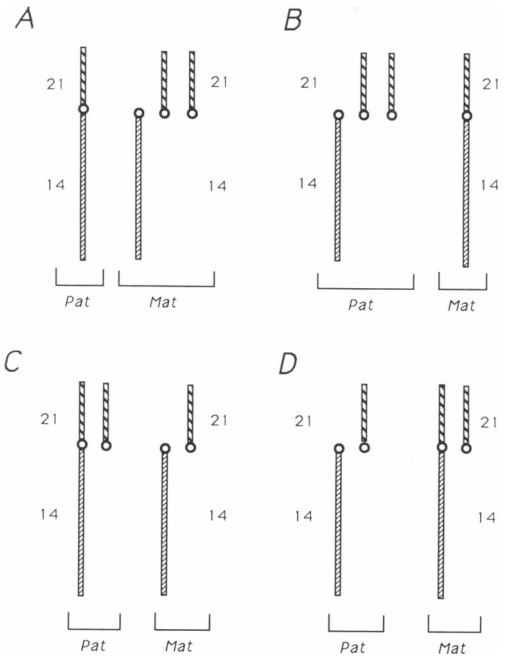

Figure 1 Schematic representation of four possible parental origins of t(14q;21q) and extra chromosome 21 in Down syndrome due to de novo t(14q;21q). Chromosome 21q is shown with heavy stripes, and chromosome 14q is shown with light stripes. In panel *A* the t(14q;21q) is paternal and the extra chromosome 21 is maternal; in panel *B* the t(14q;21q) is maternal and the extra chromosome 21 is paternal; in panel *C* both the t(14q;21q) and the extra chromosome 21 are paternal; in panel *D* both the t(14q;21q) and the extra chromosome 21 are maternal in origin. *Pat* = paternal; *Mat* = maternal.

The most proximal polymorphic DNA marker known on 21q is D21S120 (Burmeister et al. 1990). This marker, which is completely linked to D21S16 and D21S13, maps about 6 cM distal to an alphoid DNA polymorphism specific for the human chromosome 21 centromere (Jabs et al. 1991). In the present study, five of six cases informative for the D21S120 marker showed reduction to homozygosity in the proband, suggesting either a meiosis II error in the majority of cases (even if 6% of these five were misclassified because of crossing-over between the centromere and the D21S120 locus) or a chromatid translocation during meiosis I followed by normal segregation of the translocation products (see fig. 3).

The mean maternal age at birth of the trisomic child

Table 2

Chromosome 21 and Chromosome 14 DNA Polymorphisms in Families with Down Syndrome Due to de novo t(14q;21q) Translocation

Locus[a]	Polymorphisms in Family[b]			
	DS2277	DS2899	DS3202	DS3325
D21S120	11-11-111	23-12-123 NR	33-12-113 M R	23-13-333 R
D21S13	12-12-122	22-11-112 M	12-11-111	12-12-222 R
D21S110	11-12-111 R	22-12-112 M R		11-11-111
D21S11	22-12-222 R	22-11-112 M	12-12-111 R	11-12-111 R
D21S82	23-13-112 M R	22-23-222 R	12-22-122	22-12-122 NR
D21S156	12-12-122	24-13-233 M R	23-12-123 NR	13-24-124 M NR
HMG14:				
$(GT)_n$ IVS4	12-11-111	12-11-112		11-13-113 NR
$(GT)_n$ IVS5	12-11-111	23-11-113 M	12-11-111	12-13-113 NR
AluVpA IVS5	13-11-111	23-11-113 M		12-13-113 NR
MX1	11-22-122 M	22-22-222	12-12-112	11-12-112 NR
D21S113	11-12-111 R	11-22-122 M	11-11-111	12-12-122
D21S171	22-13-233 M R	22-12-222 R	22-12-222 R	22-12-122 NR
D21S112	12-34-133 M R	34-12-223 M R	13-24-234 M NR	12-33-233 M
COL6A1	12-34-144 M R	23-13-112 M R	12-11-111	22-12-122 NR
D14S13	13-24-34	22-11-12	33-12-13	12-23-13
	DS3380	DS3396	DS3399	DS3402
D21S120	23-12-112 M R	11-12-111 R	11-22-122 M	23-14-344 M R
D21S13	11-22-122 M	12-12-112	12-22-222	
D21S110				
D21S11		12-22-222	12-12-112	22-12-222 R
D21S82		33-23-223 M R	22-12-122 NR	22-33-233 M
D21S156	34-12-123 M NR	12-12-112	24-13-134 M NR	12-23-223 NR
HMG14:				
$(GT)_n$ IVS4	22-13-123 M NR	12-23-223 NR	12-12-122	22-11-112 M
$(GT)_n$ IVS5	22-13-123 M NR	12-22-222	23-13-133 NR	23-11-112 M
AluVpA IVS5	33-13-133 NR			
MX1	12-12-112	22-22-222	22-12-122 NR	11-12-112 NR
D21S113	12-12-122	11-11-111	12-12-222 R	11-12-112 NR
D21S171	23-12-123 NR	12-23-	22-12-112 M R	12-12-112
D21S112	12-13-112 R	34-12-123 M NR	22-11-112 M	33-12-123 M NR
COL6A1		23-11-113 M	12-11-111	12-11-112
D14S13	14-23-24	34-12-14	12-34-13	13-12-12

[a] The loci on chromosome 21 are ordered in columns corresponding to their relative chromosomal order based on either linkage analysis or pulsed-field gel electrophoresis. The first locus is the closest to the centromere of chromosome 21, and the last locus is the closest to the telomere of the long arm of human chromosome 21. The only chromosome 14 marker studied, D14S13, is listed at the bottom of each section.

[b] The three parts of the data correspond to father, mother, and proband, respectively. The numbers in the genotypes represent the different alleles at a specific locus. M = maternal origin of the extra 21q as determined by the respective DNA marker. R and NR = reduction to homozygosity or nonreduction to homozygosity, respectively, for a given DNA marker (see text for details).

in the seven nonmosaic cases was 26.8 years (table 1). This is not different from the mean maternal age of 27.7 in 10 maternal cases studied by cytogenetic heteromorphisms (Magenis and Chamberlin 1981; Mikkelsen et al. 1989). The combined mean maternal age from all the above 18 cases is 27.3 years, which is not significantly increased over the mean maternal age in Western societies.

Electron-microscopic studies of oocytes and spermatocytes have shown association of ribosomal genes in the nucleolar fibrillar center, providing a structural basis for the formation of Robertsonian translocations

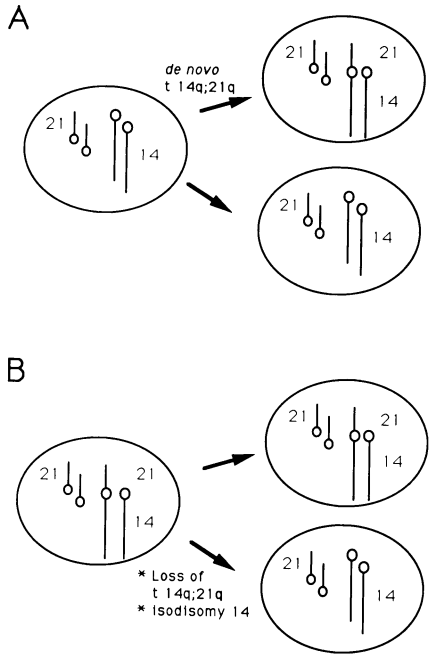

Figure 2 Schematic representation of mosaic t(14q;21q) trisomy 21. In panel *A* the zygote contains two free chromosomes 21 and two free chromosomes 14. Subsequently, in a postzygotic mitotic division, a de novo t(14q;21q) occurs in one cell lineage. In panel *B* the zygote contains two free chromosomes 21, one t(14q; 21q), and one free chromosome 14. Subsequently, in a postzygotic mitotic division, a loss of t(14q;21q) chromosome occurs and duplication of the free chromosome 14 results in isodisomy 14 in that particular cell lineage.

in meiotic prophase I (Mirre et al. 1980; Stahl et al. 1983). These studies demonstrated a close proximity of chromatids in the short-arm region of nonhomologous acrocentrics. A breakage/reunion model either by random or as an orderly, nonrandom process during meiotic pairing and exchange (Hecht and Kimberling 1971) has been suggested as an explanation for the formation of all observed types of Robertsonian translocations. Genetic exchange among ribosomal genes on nonhomologous acrocentric chromosomes has been demonstrated (Arnheim et al. 1980; Schmickel et al. 1985). Alphoid sequences shared among the centromeric regions of acrocentric chromosomes have been isolated (Devilee et al. 1986; Jorgensen et al. 1987, 1988; Choo et al. 1988, 1989), and it has been suggested that homologies are responsible for recombination and Robertsonian translocations (Choo et al. 1988, 1989). Several types of breakpoints have been proposed by molecular cytogenetic studies (Cheung et al. 1990). Structural studies of mitosis have shown that there is a close proximity of the acrocentric chromosomes when they are forming a common nucleolus during mitotic prophase (Ohno et al. 1961), and satellite association has been proposed as a risk factor in meiotic nondisjunction (Hanson and Mikkelsen 1978).

The present study suggests that the extra 21q in the majority of t(14q;21q) Down syndrome cases is maternal in origin. The mosaic case in our study is probably due to an early postzygotic nondisjunction in a mitotic division (fig. 2*A*), which is similar to the second meiotic division. The results of the DNA analysis in the nonmosaic cases of the present study do not allow a distinction between the possibilities illustrated in panels *A* and *D* in figure 1; i.e., do not differentiate between the following alternatives: (1) one chromosome 21 and the t(14q;21q) come from the mother, and (2) two chromosomes 21 are maternal, and the t(14q;21q) is paternal in origin. In figure 1*A* a paternal t(14q;21q) chromosome coincides with (or induces) a maternal meiotic nondisjunction of chromosomes 21. This alternative is possible, as meiosis II in the female finishes only after fertilization. A possible interchromosomal effect has been suggested in a cytogenetic study, in which a high proportion of meiosis II errors were found among cases of free trisomy 21 that have parents with either chromosomal rearrangements or fragile sites (Mikkelsen et al. 1989). However, the accuracy of the interpretation of the cytogenetic heteromorphisms has recently been questioned, since a number of discrepancies have been found between cytogenetic and DNA polymorphism data (Antonarakis et al. 1991). In figure 1*D*, the t(14q;21q) chromosome formation occurs in maternal meiosis. A translocation formation in the second meiotic division would be possible as a chromatid translocation between the chromosomes 14 and 21 (White 1973, pp. 198–230) and followed by nondisjunction of the free 21 chromatid. However, the mechanism illustrated in figure 1*D* can be explained by a chromatid translocation occurring during meiosis I and followed by normal segregation of the translocation products (fig. 3). In this case, reduction of centromeric markers to homozygosity is not indicative of second meiotic nondisjunction, since, as shown in figure 3, nondisjunction is not a feature of this mechanism. This alternative is compatible with all the data of the present study and requires only a single abnormal event, i.e., the chromatid translocation. It is also compatible with the cytogenetic studies where, in almost all cases, the t(14q;21q

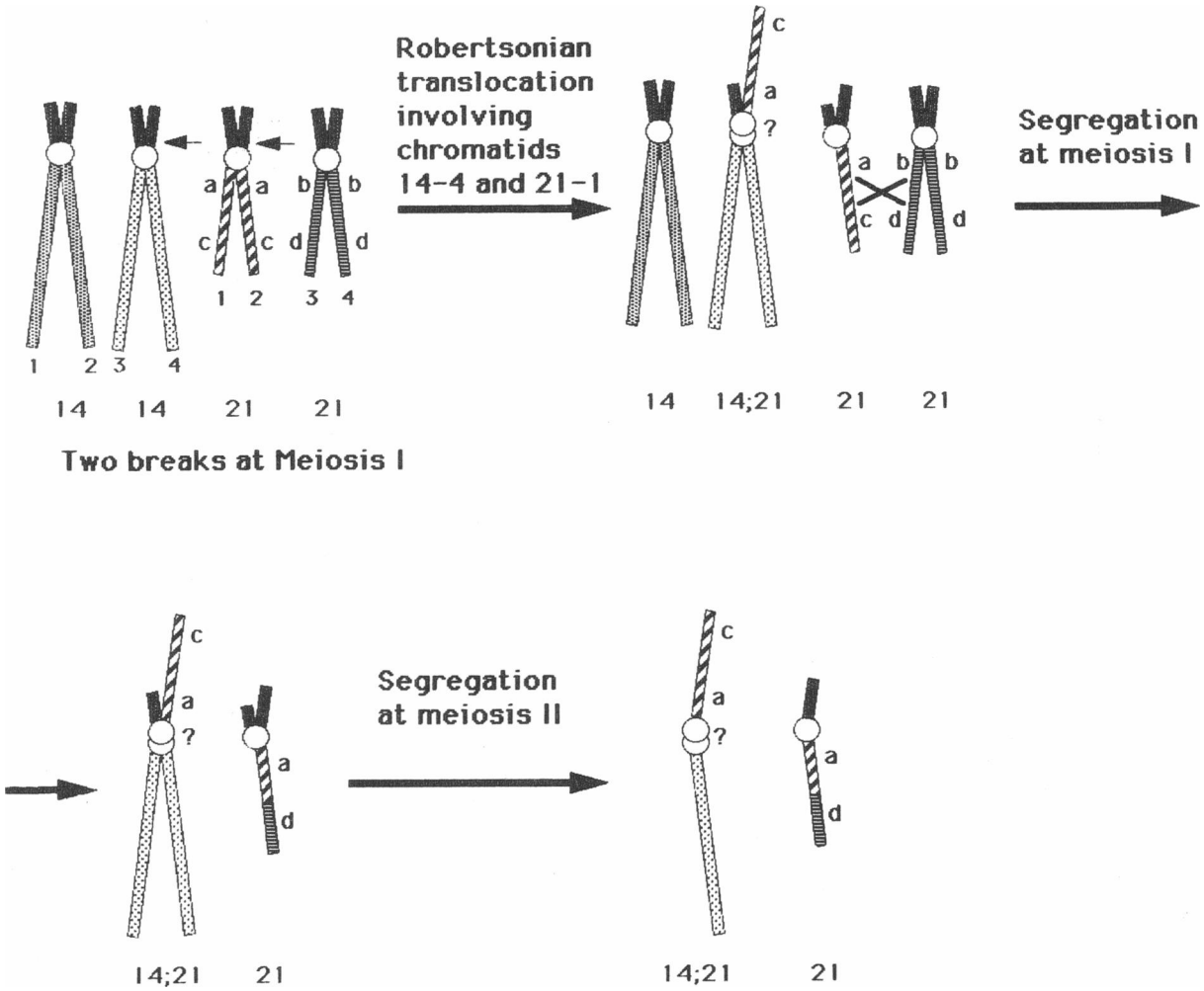

Figure 3 Mechanism for alternative *D* fig. 1, in which both extra chromosome 21 and de novo t14q;21q are maternal in origin. For this mechanism a chromatid translocation is required in meiosis I. Chromatid 1 of chromosome 21 is translocated onto chromatid 4 of chromosome 14. Normal crossing-over occurs in meiosis I, and normal segregation at meiosis I and II is shown (not all the segregation products are shown, only the one pertinent to the final abnormality). Note that DNA markers below the crossover in chromosome 21 maintain their heterozygosity (alleles c and d); however, DNA markers between the centromere of chromosome 21 and the crossover show reduction to homozygosity (allele a and b). ? = Uncertainty of the involvement of one or two centromeres of the acrocentric chromosomes in the Robertsonian translocation.

chromosome is maternal. The definitive answer to alternatives *A* and *D* illustrated in figure 1 will require both cloning of the t(14q;21q) chromosome in somatic cell hybrids and study of its DNA polymorphic markers which will establish its parental origin. The mechanism of formation of the t(14q;21q) chromosomes seems thus different from the mechanism described for dup(21q) chromosomes in patients with Down syndrome, where most of the cases are due to isochromosomes (centromere misdivision) and when only a few cases are Robertsonian translocations (Grasso et al. 1989; Antonarakis et al. 1990; Shaffer et al. 1991).

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References

Antonarakis SE, Adelsberger PA, Petersen MB, Binkert F, Schinzel AA (1990) Analysis of DNA polymorphisms suggests that most de novo dup(21q) chromosome in patients with Down syndrome are isochromosomes and not translocations. Am J Hum Genet 47:968–972

Antonarakis SE, Lewis JG, Adelsberger PA, Petersen MB, Schinzel AA, Binkert F, Schmid W, et al (1991) Parental origin of the extra chromosome in trisomy 21 as indicated by analysis of DNA polymorphisms. N Engl J Med 324: 872–876

Arnheim N, Krystal M, Schmickel R, Wilson G, Ryder O, Zimmer E (1980) Molecular evidence for genetic exchanges among ribosomal genes on nonhomologous chromosomes in man and apes. Proc Natl Acad Sci USA 77: 7323–7327

Burmeister M, Cox DR, Myers RM (1990) Dinucleotide repeat polymorphism located at D21S120. Nucleic Acids Res 18:4969

Cheung SW, Sun L, Featherstone T (1990) Molecular cytogenetic evidence to characterize breakpoint regions in Robertsonian translocations. Cytogenet Cell Genet 54: 97–102

Choo KH, Vissel B, Brown R, Filby RG, Earle E (1988) Homologous alpha satellite sequences on human acrocentric chromosomes with selectivity for chromosomes 13, 14, and 21: implications for recombination between nonhomologues and Robertsonian translocations. Nucleic Acids Res 16:1273–1284

Choo KH, Vissel B, Earle E (1989) Evolution of α-satellite DNA on human acrocentric chromosomes. Genomics 5: 332–344

Devilee P, Cremer T, Slagboom P, Bakker E, School HP, Hager HD, Stevenson AFG, et al (1986) Two subsets of human alphoid repetitive DNA show distinct preferential localization in the pericentric regions of chromosomes 13, 18, and 21. Cytogenet Cell Genet 41:193–201

Economou EP, Bergen AW, Warren AC, Antonarakis SE (1990) The polydeoxyadenylate tract of Alu repetitive elements is polymorphic in the human genome. Proc Natl Acad Sci USA 87:2951–2954

Gardiner K, Horisberger M, Kraus J, Tantravahi U, Korenberg J, Rao V, Reddy S, et al (1990) Analysis of human chromosome 21: correlation of physical and cytogenetic maps: gene and CpG island distributions. EMBO J 9:25–34

Grasso M, Giovannucci-Uzielli ML, Pierluigi M, Tavellini F, Perroni L, Dagna-Bricavelli F (1989) Isochromosome not translocation in trisomy 21q21q. Hum Genet 84:63–65

Hansson A, Mikkelsen M (1978) The origin of the extra chromosome 21 in Down syndrome: studies of fluorescent variants and satellite association in 26 informative families. Cytogenet Cell Genet 20:194–203

Hecht F, Kimberling WJ (1971) Patterns of D chromosome involvement in human (DqDq) and (DqGq) Robertsonian rearrangements. Am J Hum Genet 23:361–367

Jabs EW, Warren AC, Taylor EW, Colyer CR, Meyers DA, Antonarakis SE (1991) Alphoid DNA polymorphisms for chromosome 21 can be distinguished from those of chromosome 13 using probes homologous to both. Genomics 9:141–146

Jorgensen AL, Bostock CJ, Bak AL (1987) Homologous subfamilies of human alphoid repetitive DNA on different nucleolus organizing chromosomes. Proc Natl Acad Sci USA 84:1075–1079

Jorgensen AL, Kolvraa S, Jones C, Bak AL (1988) A subfamily of alphoid repetitive DNA shared by the NOR-bearing human chromosomes 14 and 22. Genomics 3:100–109

Kunkel LM, Smith KD, Boyer SH, Borgaonkar DS, Wachtel SS, Miller OJ, Breg WR, et al (1977) Analysis of human Y chromosome specific reiterated DNA in chromosome variants. Proc Natl Acad Sci USA 74:1245–1249

Lewis JG, Weber JL, Petersen MB, Slaughenhaupt SA, Kwitek A, May PE, Warren AC, et al (1990) Linkage mapping of the highly informative DNA marker D21S156 to human chromosome 21 using a polymorphic GT dinucleotide repeat. Genomics 8:400–402

Litt M, Luty JA (1989) A hypervariable microsatellite revealed by in vitro amplification of a dinucleotide repeat within the cardiac muscle actin gene. Am J Hum Genet 44:397–401

McCormick MK, Schinzel A, Petersen MB, Stetten G, Driscoll DJ, Cantu ES, Tranebjaerg L, et al (1989) Molecular genetic approach to the characterization of the "Down syndrome region" of chromosome 21. Genomics 5:325–331

Magenis RE, Chamberlin J (1981) Parental origin of nondisjunction. In: de la Cruz FF, Gerald PS (eds) Trisomy 21 (Down syndrome) research perspectives. University Park Press, Baltimore, pp 77–93

Mikkelsen M, Fischer G, Stene J, Stene E, Petersen E (1976) Incidence study of Down's syndrome in Copenhagen, 1960–1971: with chromosome investigation. Ann Hum Genet 40:177–182

Mikkelsen M, Poulsen H, Tommerup N (1989) Genetic risk factors in human trisomy 21. In: Hassold TJ and Epstein CJ (eds) Molecular and cytogenetic studies of nondisjunction. Alan R Liss, New York, pp 183–197

Mirre C, Hartung M, Stahl A (1980) Association of ribosomal genes in the fibrillar center of the nucleolus: a factor influencing translocation and nondisjunction in the human meiotic oocyte. Proc Natl Acad Sci USA 77:6017–6021

Nakamura Y, Culver M, Gill J, O'Connell P, Leppert M, Lathrop GM, Lalouel JM, et al (1988) Isolation and mapping of a polymorphic DNA sequence pMLJ14 on chromosome 14 (D14S13). Nucleic Acids Res 16:381

Ohno S, Trujillo JM, Kaplan WD, Kinosita R (1961) Nucleolus-organisers in the causation of chromosomal anomalies in man. Lancet 2:123–125

Petersen MB, Economou EP, Slaugenhaupt SA, Chakravarti A, Antonarakis SE (1990) Linkage analysis of the human HMG14 gene on chromosome 21 using a GT dinucleotide repeat as polymorphic marker. Genomics 7:136–138

Petersen MB, Schinzel AA, Binkert F, Tranebjaerg L, Mikkelsen M, Collins FA, Economou EP, et al (1991*a*) Use of short sequence repeat DNA polymorphisms after PCR amplification to detect the parental origin of the additional chromosome 21 in Down syndrome. Am J Hum Genet 48:65–71

Petersen MB, Slaugenhaupt SA, Lewis JG, Warren AC, Chakravarti A, Antonarakis SE (1991*b*) A genetic linkage map of 27 markers on human chromosome 21. Genomics 9:407–419

Petersen MB, Weber JL, Slaugenhaupt SA, Kwitek AE, McInnis MG, Chakravarti A, Antonarakis SE. Linkage mapping of D21S171 to the distal long arm of human chromosome 21 using a polymorphic $(AC)_n$ dinucleotide repeat. Hum Genet (in press)

Pulliam LH, Huether CA (1986) Translocation Down syndrome in Ohio 1970–1981: epidemiologic and cytogenetic factors and mutation rate estimates. Am J Hum Genet 39:361–370

Robinson JA (1973) Origin of extra chromosome in trisomy 21. Lancet 1:131–133

Saiki RK, Scharf S, Faloona F, Mullis KB, Horn GT, Erlich HA, Arnheim N (1985) Enzymatic amplification of β-globin genomic sequences and restriction site analysis for diagnosis of sickle cell anemia. Science 230:1350–1354

Schmickel RD, Gonzalez IL, Erickson JM (1985) Nucleolus organising genes on chromosome 21: recombination and nondisjunction. Ann NY Acad Sci 450:121–131

Shaffer LG, Jackson-Cook CK, Meyer JM, Brown JA, Spence JE (1991) A molecular genetic approach to the identification of isochromosomes of chromosome 21. Hum Genet 86:375–382

Stahl A, Luciana JM, Hartung M, Devictor M, Bergé-Lefranc JL, Guichaoua M (1983) Structural basis for Robertsonian translocations in man: association of ribosomal genes in the nucleolar fibrillar center in meiotic spermatocytes and oocytes. Proc Natl Acad Sci USA 80:5946–5950

Tautz D (1989) Hypervariability of simple sequences as a general source for polymorphic DNA markers. Nucleic Acids Res 17:6463–6471

Warren AC, Chakravarti A, Wong C, Slaugenhaupt SA, Halloran SL, Watkins PC, Metaxotou C, et al (1987) Evidence for reduced recombination on the nondisjoined chromosomes 21 in Down syndrome. Science 237:652–654

Warren AC, Slaugenhaupt SA, Lewis JG, Chakravarti A, Antonarakis SE (1989) A genetic linkage map of 17 markers on human chromosome 21. Genomics 4:579–591

Weber JL (1990) Informativeness of human $(dC\text{-}dA)_n$ $(dG\text{-}dT)_n$ polymorphisms. Genomics 7:524–530

Weber JL, May PE (1989) Abundant class of human DNA polymorphisms which can be typed using the polymerase chain reaction. Am J Hum Genet 44:388–396

White MJD (1973) Animal cytology and evolution. Cambridge University Press, Cambridge