Am. J. Hum. Genet. 53:1074-1078, 1993

Normal Phenotype with Paternal Uniparental Isodisomy for Chromosome 21

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Summary

Uniparental disomy (UPD) involving several different chromosomes has been described in several cases of human pathologies. In order to investigate whether UPD for chromosome 21 is associated with abnormal phenotypes, we analyzed DNA polymorphisms in DNA from a family with de novo Robertsonian translocation t(21q;21q). The proband was a healthy male with 45 dup(21q) who was ascertained through his trisomy 21 offspring. No phenotypic abnormalities were noted in the physical exam, and his past medical history was unremarkable. We obtained genotypes for the proband and his parents' leukocyte DNAs from 17 highly informative short sequence repeat polymorphisms that map in the pericentromeric region and along the entire length of 21q. The order of the markers has been previously determined through the linkage and physical maps of this chromosome. For the nine informative markers there was no maternal allele contribution to the genotype of the proband; in addition, there was always reduction to homozygosity of a paternal allele. These data indicated that there was paternal uniparental isodisomy for chromosome 21 (pUPiD21). We conclude that pUPiD21 is not associated with abnormal phenotypes and that there are probably no imprinted genes on chromosome 21.

Introduction

Uniparental disomy (UPD) is defined by the presence of two homologous chromosomes (in a diploid offspring) inherited from one parent in all or part of their length (Engel 1980). In uniparental isodisomy (UPiD), both chromosomes are in addition genetically identical to each other because they are derived from a single parental chromosome. The most likely mechanisms for UPD include fertilization of a nullisomic gamete by a disomic gamete, postzygotic loss of a chromosome in a trisomic conceptus, or compensatory duplication in a monosomic cell; in addition, mitotic recombination in somatic cells may result in a partial UPD (Engel 1980; Spence et al. 1988; Engel and DeLozier-Blanchet 1991; Petersen et al. 1992; Henry et al. 1993). Several human genetic diseases have been associated with UPD for different chromosomes because of homozygosity for recessive alleles, parentally imprinted genes, or a combination of both mechanisms.

Homozygosity for recessive alleles is considered to be the cause of complement deficiency in UPD for chromosome 6 (UPD6) (Welch et al. 1990), cystic fibrosis or a mild form of osteogenesis imperfecta in UPD7 (Spence et al. 1988; Voss et al. 1989; Spotila et al. 1992), rod monochromacy in UPD14 (Pentao et al. 1992), and one case of thalassemia major in UPD11 (Beldjord et al. 1992). On the other hand, parental imprinting seems to be responsible for abnormal phenotypes in maternal or paternal UPD15 in Prader-Willi syndrome and Angelman syndrome, respectively (Nicholls et al. 1989; Malcolm et al. 1991; Robinson et al. 1991; Driscoll et al. 1992), as well as in paternal UPD11 in some cases of Beckwith-Wiedemann syndrome (Henry et al. 1991). Furthermore, paternal or maternal UPD14 has been recently associated with some specific phenotypes (Temple et al. 1991; Wang et al. 1991; Pentao et al. 1992; Antonarakis et al. 1993). We report here the cytoge-

Received May 19, 1993; final revision received July 7, 1993.

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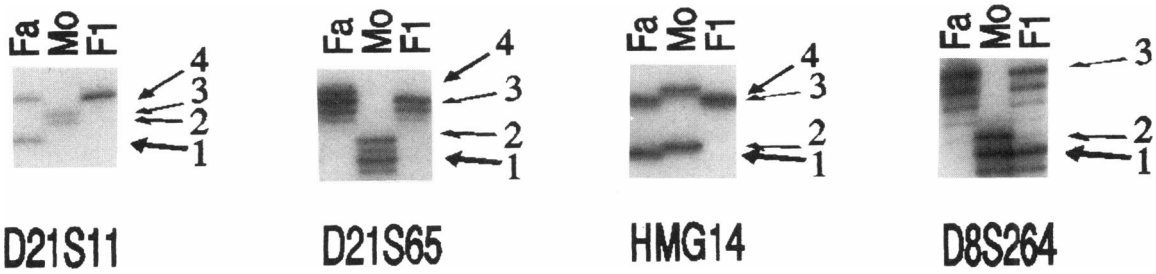

Figure 1 Autoradiograms of representative DNA polymorphism analyses in the members of the family of individual JH-4025. The chromosome 21 markers (D21S11, D21S65, and HMG14) show that there was only paternal contribution in the genotype of the proband. Chromosome 8 marker (D8S264) excludes nonmaternity due to sample error.

netic and genotype analyses of a family in which individual JH-4025 (who was ascertained through a child with t[21q;21q] trisomy 21) was found to have a duplication of chromosome 21q associated with a normal phenotype. The DNA polymorphism analysis revealed a paternal uniparental isodisomy for chromosome 21q (pUPiD21). The present study, as well as a previously reported case of maternal UPD21 (Creau-Goldberg et al. 1987) in a woman with normal phenotype, suggests that genes on chromosome 21q may not be parentally imprinted.

Subject and Methods

Proband

The proband is a 40-year-old Caucasian male (JH-4025) ascertained through his offspring who had trisomy 21. The proband was the product of a normal pregnancy; his birth weight was 3,400 g, and no abnormalities were reported during the neonatal period. He had normal development and onset of puberty at age 14 years. Physical examination at age 40 years revealed no abnormal phenotype. Height was 172 cm, weight was 60 kg, and head circumference was 56.5 cm. The mother of the proband was 29 years old at the time of his birth and had a total of three pregnancies, one of which resulted in spontaneous abortion. The father of the proband was 35 years old at the time of the birth.

Karyotype Analysis

The proband's male offspring was clinically diagnosed as having Down syndrome, and his RHG-banded karyotype revealed a trisomy 21 due to dup(21q). Subsequently, karyotypic analyses of blood lymphocytes were performed in the proband, his wife, and his parents.

DNA Polymorphism Analysis

Genomic DNA from the proband and his parents was purified from blood lymphocytes. DNA polymorphisms were analyzed by PCR amplification of polymorphic short sequence repeats. The markers analyzed were as follows: D21S215, D21S258, D21S120, D21S13, D21S11, D21S210, D21S230, D21S217, D21S213, GART, D21S65, D21S167, HMG14, D21S228, D21S231, D21S212, PFKL (Genome Database [GDB]; Antonarakis 1992; NIH-CEPH Collaborative Mapping Group 1992), D8S264, and D8S262 (Weissenbach et al. 1992). For information on the mapping position of the markers and the oligonucleotides used for amplification, see the appropriate entries in the GDB and McInnis et al. (1993). The PCR using radiolabeled primers, PAGE, and the scoring of the alleles for the polymorphic markers were performed as described elsewhere (Economou et al. 1990; Petersen et al. 1991).

Results

The cytogenetic analysis of the proband revealed a dup(21q) or Robertsonian translocation t(21q;21q); the karyotype performed on more than 30 lymphocytes did not show any mosaicism. Karyotypes of the parents were normal; therefore, the proband had de novo dup(21q). In order to determine the origin of the translocation and the presence of potential UPD, we determined the genotypes of 17 polymorphic loci on chromosome 21q in blood DNA of the proband and his parents. The proband was found to be homozygous for one paternal allele in all the markers typed. In addition, data for nine markers show inheritance of only one paternal allele and no presence of maternal alleles in the proband (fig. 1 shows representative autoradiograms from selected markers). These results suggest pUPiD21. To eliminate the possibility of nonmaternity in the family due to sample error, we also tested the DNA from the proband and his parents for two markers on chromosome 8; the data show contribution of polymorphic alleles from both parents in the genotype of the pro-

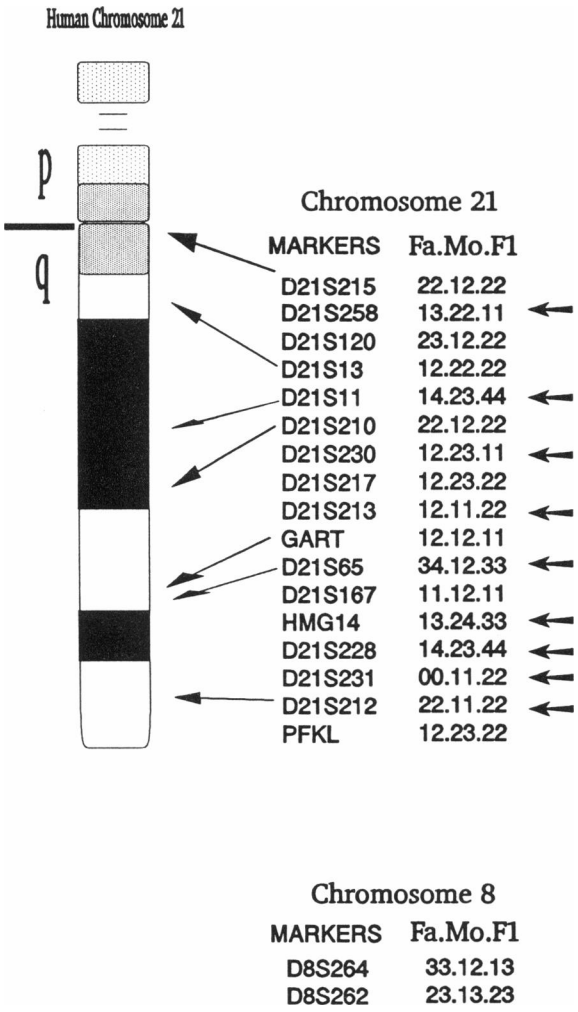

Figure 2 Genotypes of DNA polymorphisms in the family of the proband. The chromosome 21 markers are listed according to their map location. The numbers refer to different alleles. For example, for marker D21S11, the father (Fa) showed alleles 1 and 4, the mother (Mo) showed alleles 2 and 3, and the proband (F1) showed the paternal allele 4 and no contribution from the mother. Arrowheads indicate the informative data for the pUPiD21.

band. All the genotyping results for the chromosome 21q and chromosome 8 polymorphic markers are shown in figure 2.

Discussion

UPD without an Abnormal Phenotype

In this study we describe a healthy individual with pUPiD21. Despite the pUPiD21 observed in blood cells of the patient, no abnormal phenotype and no dysmorphic features were noticed. He came to medical attention because he fathered a child in whom trisomy 21 due to a Robertsonian translocation t(21q;21q) was present, resulting in Down syndrome phenotype.

A case of maternal UPD (mUPD) for chromosome 21 with no abnormal features in the proband has also been reported elsewhere (Creau-Goldberg et al. 1987). The female proband in that study, with a de novo translocation t(21q;21q), also has a child affected with Down syndrome with 46,XX,−21,+t(21q;21q). In the latter study few DNA polymorphisms were examined, and it is not reported whether there was iso- or heterodisomy for 21q or whether the UPD extended through the entire long arm of the chromosome. The present study and that of Creau-Goldberg et al. (1987) suggest that parental imprinting (paternal or maternal) probably does not occur in genes on human chromosome 21.

There are other documented cases of UPD in which there is no associated phenotype due to parental imprinting of genes. These cases include, e.g., (1) individuals with mUPD22 (Kirkels et al. 1980; Palmer et al. 1980) in which there was maternal transmission of a t(22q;22q) chromosome from mothers to normal daughters and (2) some infants with mUPD16 in whom there had been mosaicism for trisomy 16 in the fetus with subsequent somatic loss of one of the three chro-

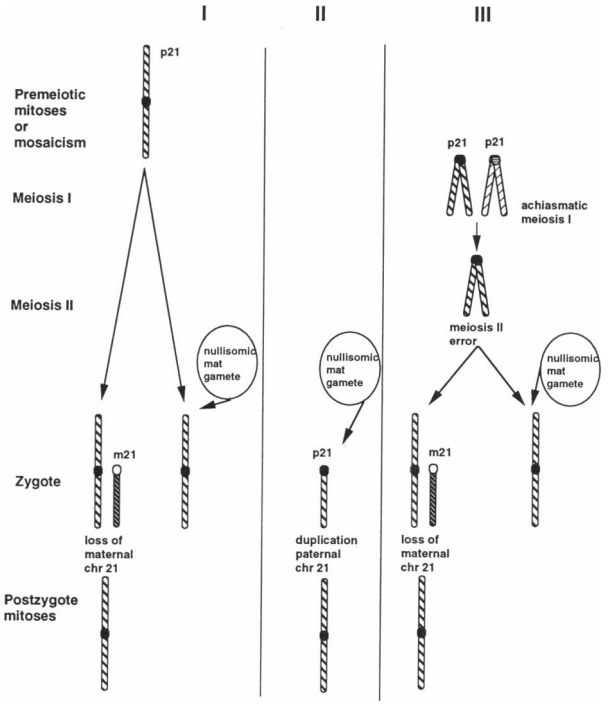

Figure 3 Possible alternative mechanisms for the formation of the pUPiD21. For explanation, see Discussion. p21 = paternal chromosome 21; m21 = maternal chromosome 21.

mosomes 16 (Bennett et al. 1992; Dworniczak et al. 1992; Kalousek et al. 1992).

In the mouse, not all chromosomes are involved in the parental imprinting phenomenon. The mouse chromosomes that correspond to human chromosome 21 are parts of mouse chromosomes 16, 17, and 10 (Reeves and Miller 1992; Silver et al. 1992; Taylor et al. 1992; O'Brien et al. 1993). The absence of abnormal phenotype in human UPD21 provides indirect evidence that the homologous mouse chromosomal regions may not contain parentally imprinted genes; in fact, no genomic imprinting has been observed on mouse chromosome 16 (Berger and Epstein 1989). However, it is also possible that imprinting is not always conserved in evolution. It is of interest that mouse chromosome 17 is involved in parental imprinting (Searle et al. 1989); it is therefore possible that the region of mouse chromosome 17 that contains the parentally imprinted genes lies outside the small area that is homologous to human chromosome 21 and that contains the murine loci d21S56, Crya-1, and Cbs.

Mechanism of the pUPiD21

Several mechanisms of chromosome segregation can explain the occurrence of the pUPiD21 in the family described here (see fig. 3).

I. Premeiotic formation or presence of dup(21q).
   A. Premeiotic duplication of chromosome 21q in the paternal germ cells, followed by loss of maternal chromosome 21 after formation of the zygote or fertilization of a nullisomic maternal gamete.
   B. Mosaicism in the paternal germ cells for a dup(21q), followed by loss of maternal chromosome 21 after the formation of the zygote or fertilization of a nullisomic maternal gamete. No mosaicism has been observed in the proband or his father (65 metaphases examined in the proband, 78 in the father, and 60 in the mother). Furthermore, no evidence for mosaicism was observed by analysis of DNA polymorphisms. In a recent study, mosaicism of 5% was easily detected by analysis of short sequence repeat DNA polymorphisms after PCR amplification (Antonarakis et al. 1993).

II. Formation of a zygote from normal sperm and nullisomic oocyte followed by duplication of the parental chromatid. This mechanism has been previously suggested as a compensatory effect by Petersen et al. (1992).

III. Paternal nonseparation of chromatids in meiosis II, followed by loss of maternal chromosome 21 from the zygote or fertilization of a nullisomic maternal gamete. This is unlikely, since the abnormal paternal meiosis II would have to follow an achiasmatic meiosis I in order to produce the observed reduction to homozygosity for all the DNA polymorphic markers examined in the proband.

Study of the clinical phenotypes associated with UPDs for all human chromosomes and the understanding of the contribution of homozygosity for recessive alleles to the phenotype versus that of parental imprinting might soon uncover the importance of UPD to human pathology and nosology and change the genetic counseling of many conditions. The absence of observable phenotype due to parentally imprinted genes on human chromosome 21 suggests that UPD for other human chromosomes might also not be associated with human disease.

Acknowledgments

This study was supported by NIH grant HD24605 and a Swiss Fonds Nationale de la Recherche Scientifique grant (to S.E.A). J.-L.B. was partially supported by training grant from Philippe Foundation and French Fondation pour la Recherche Medicale. We thank M. Ghika and D. Abatzis for assistance in the cytogenetic analysis, and we thank C. C. Talbot, Jr., for comments on the manuscript.

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