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Down syndrome: Cryptic paternal translocation (9;21).

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Abstract

Down syndrome is the most frequent autosome aneuploidy in live newborns, caused by trisomy of all or the critical region of chromosome 21. We described a 20 month old boy with Down syndrome phenotype. Full clinical examination was done. Cytogenetic studies for the proband and his parents using GTG- banding technique and Fluorescence Insitu Hybridization technique using LSI 21 and chromosome 9q subtelomere probe were used according to the manufacturer's instructions. The proband;s karyotype is: 46,XY,der(9)t(9;21)(p24;q22.2) resulted from paternal balanced cryptic translocation. Karyotype of the father is 46,XY, t(9;21)(p24;q22.2) , while mother showed normal Karyotype. FISH analysis using LSI 21 probe and 9q subtelomere probe for the proband's and father demonstrated the presence of three signals, and two signals respectively. This is the first report describing a paternal cryptic balanced translocation involving chromosome 9 with the generation of aneuploidy contributing to abnormal chromosomal segregation.

Keywords: Down syndrome, cryptic translocation, FISH

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Introduction

Down syndrome DS the most frequent form of mental retardation caused by a chromosomal aberration is characterized by well-defined and distinctive phenotypic features and caused by trisomy of all or a critical portion of chromosome 21 with a frequency of 1 in 650 to 1,000 live births in the population.

It has long been recognized that the risk of having a child with trisomy 21 increases with maternal age; the risk of having a liveborn with DS at maternal age 30 is 1 in 1,000 and at maternal age 40 is 9 in 1,000.

Reviewed studies [1,2,3,4,5] reported errors in meiosis that lead to trisomy 21 were overwhelmingly of maternal origin; only 5% occur during spermatogenesis, while most errors occur in meiosis I which account for 76 to 80% and maternal meiosis II errors constitute 20 to 24%. In families in which there is paternal nondisjunction, most of the errors occur in meiosis II, while in 5% of trisomic individuals the supernumerary chromosome 21 appears to result from an error in mitosis. These data were also supported in many studies as approximately 91% of DS cases arise from maternal nondisjunction (ND) and only 9% from paternal aneuploidies.[6] All de novo t (14; 21) trisomies have originated in maternal germ cells, [7] the mean maternal age was 29.2 years. In de novo t (21; 21) DS the situation is different, [8,9,10] in most cases the t (21; 21) is an isochromosome (dup 21q) rather than the result of a Robertsonian translocation caused by a fusion between 2 heterologous chromatids. About half were of paternal and half of maternal origin. Less frequently t (21; 21) true Robertsonian trisomy 21 cases, the extra chromosome 21 was maternal.

Here we report on a family who had a child with DS due to paternal ND in Meiosis II**,** FISH analysis using the LSI 21 probe (Vysis) spectrum orange (21q22.13-q22.2) showed that the father had an extra positive signal on chromosome 9. We consider being a cryptic translocation, which seems to be related to the paternal ND that gave rise to two sibs with DS.

Clinical Examination

A male patient aged one year and 8ms. at time of examination was referred to the clinic of Children with Special Needs, National Research Center having DS. The parents were healthy, nonconsanguineous, under 35 years of age when the DS child was born, and had another DS daughter. History of a previous child diagnosed as DS who died at the age of 5m and a paternal female cousin with DS.

The proband was delivered by C.S. (elective), ± full term after a pregnancy with threatened abortion during the second month of gestation. Physical examination showed a classical DS phenotype with weight 25th centile, length 10th centile, head circumference 3rd centile, mild brachycephaly, flat nasal bridge, upward slanting palpebral fissures, epicanthal folds, asymmetric dysplastic ears with folded right helix, high arched palate, micrognathia, loose skin on neck nape, small genitalia, brachydactyly, single transverse palmar crease and gap between first and second toes. Echo Cardiography demonstrated the presence of VSD.

Cytogenetic studies and Results

Chromosomal analysis for the family was done using G-banding technique according to the described methods, a total of 25 metaphases were analyzed for each case [11,12]. Structural or numerical anomalies were recorded and karyotyped according to the ISCN (2009). [13] Cytogenetic analysis of the proband revealed 46, XY, der(9)t(9;21)(p24;q22.2) , Fig. (1). Cytogenetic analysis for the sister was not available, mother showed normal Karyotype 46, XX and father showed 46, XY, t(9;21)(p24;q22.2) respectively.

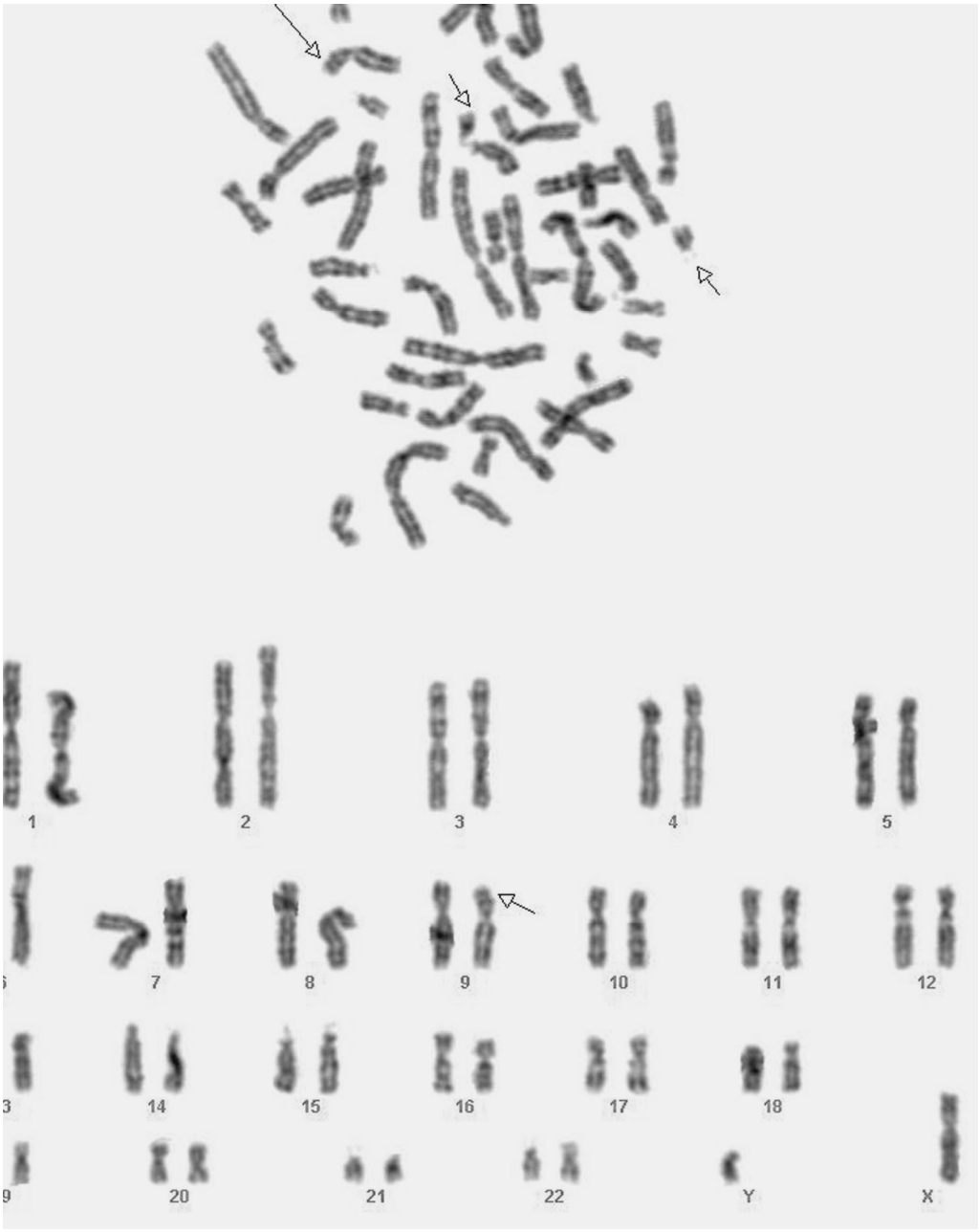

Figure 1. *Karyotype of the proband: 46, XY, t(9:21)(p24:q22.2).*

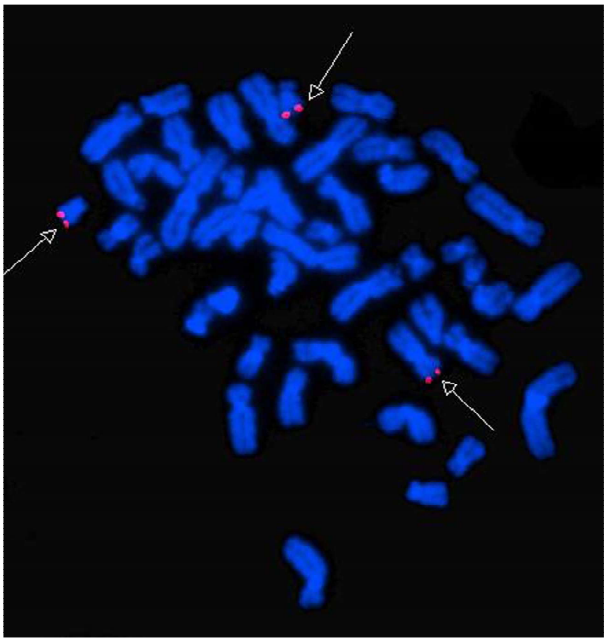

Figure 2 *FISH using LSI 21 probe (Vysis) (spectrum orange) of the proband demonstrating the presence of 3 signals.*

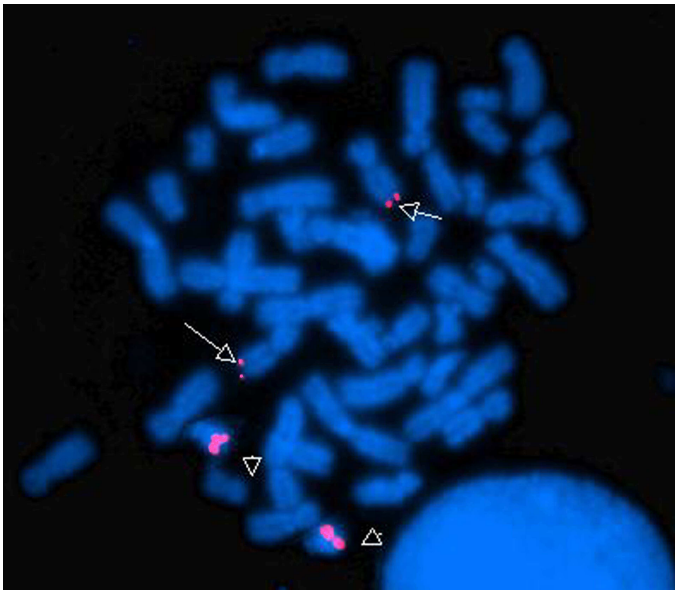

Figure 3. *FISH using LSI 21 probe (Vysis) (spectrum orange)(arrow heads) and Tel. 9q probe (Vysis) (spectrum orange) (long arrows) of the mother .*

Fluorescent in situ hybridization (FISH) using LSI 21 probe (Vysis) spectrum orange (21q22.13-q22.2) to identify the critical region of Down syndrome and Tel. 9q probe (Vysis) spectrum orange to identify the long arm of chromosome 9 was applied. [14] Slides prepared from 3:1 methanol: acetic acid fixed cells, pretreated with 2xSSC at 37° C for 30 min., dehydrated for 2 min. in

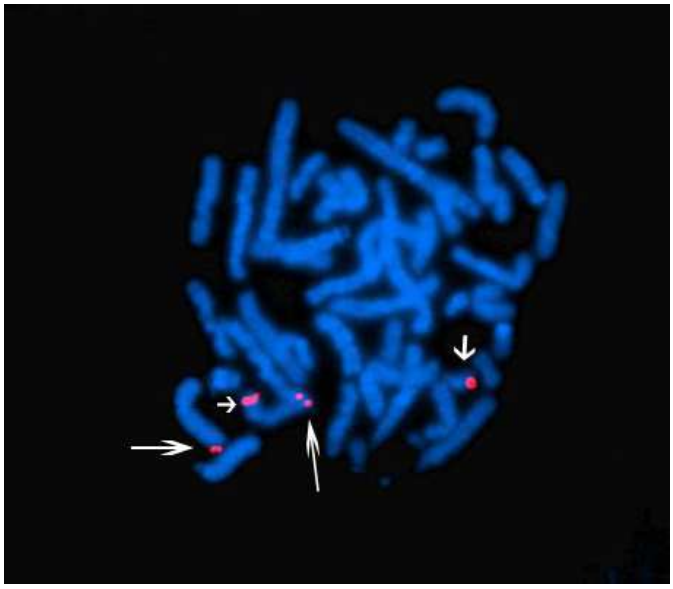

Figure 4. *FISH using LSI 21 probe (Vysis) (spectrum orange)(arrow heads) and Tel. 9q probe (Vysis) (spectrum orange)(long arrows) of the father . washed in 0.4xSSC in 73°C for 2 min. followed by (2xSSC/NB-40) for one min. and lastly in 2xSSC for 2 min at room temperature. Counterstaining of cells was done using 10 µl DAPI.*

70%, 90% and 100% ethanol respectively. Denaturation of the slides in 70% formamide at 73 ± 1° C for 5 min. were done followed by dehydration in cold ethanol 70%, 90% and 100%. Denaturation of 10 µl of the probe in a water bath at 74 ± 1 °C for 5 min. was done, added to slides, covered and sealed with rubber cement and incubated at 37°C overnight. Slides were FISH using LSI 21 probe (Vysis) (spectrum orange) of the proband demonstrating the presence of 3 signals as indicated by the arrows Fig. (2), two signals represent normal signals on chromosome 21q22.2 while the third one represent the translocated signal to the short arm of chromosome 9 (9p24). FISH of the mother using LSI 21 probe (Vysis) (spectrum orange) (arrow heads) and Tel. 9q probe (Vysis) (spectrum orange) (arrows) revealing normal signals. Fig. (3). FISH of the father using LSI 21 probe (Vysis) spectrum orange (short arrow) showed two signals, one signal on 21q22.2 and a translocated signal to the terminal end of the short arm of chromosome 9 (9p24) Fig. (4).

Discussion

To date, advanced maternal age is the only factor confirmed to be associated with chromosome 21 ND, while ND of paternal origin shows no association with advanced age. A relatively small region may play a major

role in DS phenotypes, and proposed the concept of a DSCR [15] which was defined with a proximal boundary between markers D21S17 (35 892 kb) and D21S55 (38 012 kb), and a distal boundary at *MX1* (41 720 kb) [16,17].

We present a case with DS phenotype where cytogenetic examination revealed 46, XY, t(9:21)(p24:q22.2). FISH examination demonstrated the presence of a translocated signal to the short arm of chromosome No.9. Parental examination has revealed normal 46, XX for the mother and 46, XY for the father while FISH analysis revealed a translocated signal to 9p proofing that segregation is an apparently balanced cryptic translocation from the father.

Partial duplication of the DSCR (D21S55, sub band 21q22.2) of maternal origin in the patient was documented; [18] the trisomy was due to segregation of an apparently balanced cryptic translocation from the mother. The patient's karyotype was 46,XX,-12,tder(12)t(12;21)(p13.1;q22.2)maternal. In our study the father showed a balanced translocation between 21q22.2 and 9p24 as evidenced from the normal size of the two chromosomes 21, however the possibility of other chromosome translocations cannot be excluded. The cryptic translocation found on the paternal der (9) chromosome could have led to ND of chromosome 21 in sperm, giving rise to a DS child. Gair et al. [2005] suggested that a cryptic translocation on chromosome 21 may have increased the frequency of ND of this chromosome in a family with recurrent trisomy 21 in three generations. [19]

Two cases were reported with partial trisomy 21 having the clinical features distinct for DS, each patient had a similarly sized extra chromosome 21 , each of their extra chromosomes contained a distal part of chromosome 3p or 14q at the telomeric region of chromosome 21q (from maternal origin) [20]. In another study , they reported many individuals displaying subtle translocations and internal rearrangements causing duplications and deletions of HSA21 region, for some patients additional copies of several non-contiguous regions were detected indicating that complex rearrangements involving multiple events had occurred. [21] The present study represents a rare balanced translocation in a proband in a DS family where the origin is from a cryptic paternal translocation as most reported familial non- acrocentric translocation cases were of maternal origin and also highly evaluate the use of different FISH techniques for proper diagnosis and genetic counseling. It is important to establish the frequency of cryptic translocations, which may be an important etiologic factor in the generation of the most frequent chromosomal syndromes in future generations.

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