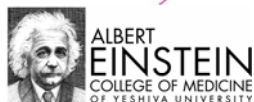


Delineation of the breakpoints of pure duplication 3q due to a de novo duplication event using SOMA



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Introduction

Partial duplication of chromosome 3q is a well-described condition of multiple congenital anomalies that resemble the Cornelia de Lange syndrome (CdLS). Most cases of dup(3q) syndrome result from an unbalanced translocation, inversion or insertion. Pure duplication of 3q is very rare and has been reported only eight times previously. The minimal region of overlap in the duplication 3q syndrome although variable in size includes 3q26.3-q27 (Baldini, 1994). We report a case of a 3 and 6/12 year old male with a de novo duplication of 3q21-q29. The duplicated region includes the critical region. He has many of the salient features of the dup 3q syndrome with an unusually severe phenotype.

Case report

EC was the 3160 gm product of a full term pregnancy delivered to a 26 year old primigravida by NSVD. He remained in the NICU for 10 days because of feeding difficulties. He was admitted to our hospital at 5 weeks because of apnea and bradycardia. His physical examination at 4 and ½ months revealed a stigmatized youngster (Fig. 1). His weight was 5.42 kg, length 58.5 cm and HC 39 cm all beneath the 2%. He had bushy eyebrows, long eyelashes, synophrys and a nevus flammeus of the forehead. The pinna were crumpled, the nasal bridge was depressed, the philtrum long and the jaw micrognathic. He had syndactyly of toes 2 and 3 bilaterally and a remnant of a right-handed post-axial polydactyly and bilateral fifth digit clinobrachydactyly. He had bilateral optic nerve hypoplasia and a right facial palsy with exposure keratopathy. He was receiving phenobarbital for a focal seizure disorder. Primary aspiration was thought to be responsible for recurrent pneumonia. A sleep study revealed obstructive sleep and central apnea. The obstructive events improved after an tonsillectomy and adenoidectomy. He also underwent bilateral orchiopexies and a glossopexy. A CT scan done at 17 months revealed craniosynostosis of the right inferior lambdoidal suture resulting in asymmetric posterior plagiocephaly, absence of the corpus callosum and decreased white matter (Fig. 2). A scoliosis survey showed a shallow left acetabulum and 12 ribs bilaterally. When last examined at 3 and 6/12 years of age, his weight was 13.6 kg(10-25%), length 89.5 cm (<3%), and HC was 45.25 (-4SD's) (Fig. 3). He was nonverbal but able to sit and crawl and cruise. He was visually impaired and was being fitted for bilateral hearing aides because of a conductive hearing loss. Cytogenetic analysis revealed a de novo duplication of 3q: 46, XY .ish dup(3)(q21q29)(wcp3+,D3S4560+) (Fig. 4).

SNP Oligonucleotide Microarray Analysis (SOMA)

SOMA was performed using the Affymetrix Genome Wide Human SNP Array 6.0, which includes over 906,600 single nucleotide polymorphisms (SNPs) and more than 946,000 probes for the detection of copy number variation. Sample preparation, hybridization and scanning were performed using GeneChip® Instrument System hardware according to manufacturer's specifications (Affymetrix, Santa Clara, CA). Analysis was performed using the Affymetrix Genotyping Console software. The samples met Affymetrix recommended values for Contrast QC (SNP) and MAPD QC (CNV). The intensities of both SNP and CNV probes were used to determine segments that varied in copy number. The Segment report was restricted to regions of 100kb or greater with 10 or more consecutive probes that differed significantly from the expected normalized diploid values. SNP intensities were normalized against the values for the HapMap 270 samples. Copy number intensities were normalized against a group of 530 samples from the NIGMS Chromosomally Abnormal Collection.

Results

SOMA studies using the Affymetrix® GenomeWide Human SNP Array 6.0 indicated the duplication to be 61.07 Mb with proximal and distal breakpoints at 3q22.2 and 3q29 respectively. The duplicated segment extends from 136041097 to 197291124 bps (Fig. 5).

Discussion

The dup 3q syndrome was first reported by Falek in 1966. More than 40 cases have been reported but 75% have arisen from segregation of a parental rearrangement. Only a few cases with pure dup 3q have been described (Table 1). A CdLS gene within the critical region has been thought to result in the overlap of mild CdLS and the dup 3q syndrome. A CdLS gene location (CdL1) was assigned to 3q26.3 based upon phenotypic overlap with the duplication 3q syndrome but has been excluded by family linkage studies. The CdL1 locus did not segregate in more than 50% of cases (Krantz, 2001). Mutations in a number of candidate genes in the critical area including *CHRD*, *SOX2*, *SHOT*, *GSC*, *NAALADL2* and *NLGN1* were not identified in patients with CdLS.

Our patient has many of the dysmorphic features typical of the dup 3q syndrome and appears to be among the more severely clinically affected. In addition, he demonstrates a number of previously unreported findings including polydactyly, 12 ribs, optic nerve hypoplasia, absence of the corpus callosum and decreased white matter, and central apnea. The duplicated region contains more than 60 Mb of genomic information and hundreds of genes. The extent of the duplication and the variability of the clinical severity in patients with dup 3q suggests that dup 3q may be a contiguous gene syndrome with more than one gene in the critical region responsible for the phenotype. The clinical severity, unusual clinical features and the extent of the duplicated material in our case supports the observation that dup 3q is a contiguous gene syndrome. This case represents the first application of SOMA to the dup 3q syndrome. The delineation of the duplicated region by SOMA in this case and future cases will enable us to identify disease genes associated with this phenotype.



Figure 1. Patient at 4 1/2 months of age.

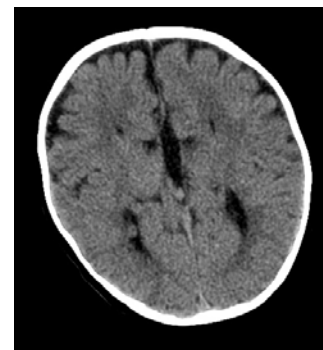


Figure 2. Axial view of noncontrast CT of brain demonstrates parallel appearance to the lateral ventricles.



Figure 3. Patient at 3 6/12 years of age.



Figure 4. Standard metaphase karyotype.

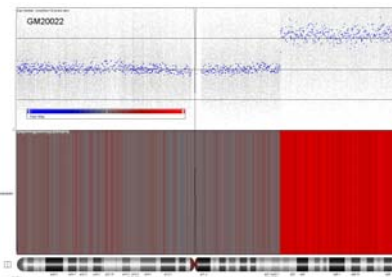


Figure 5. SOMA identifies a duplication in the long arm of chromosome 3 in GM20022. The duplicated segment on chromosome 3 lies between marker SNP_A-8544030 at chr3:136044785 and marker SNP_A-8452318 at chr3:197194059.

Table 1. Clinical features of pure dup(3q) patients.

		Wilson et al., 1978, case 1	Wilson et al., 1978, case 2	Stengel-Rutkowski et al., 1979	Sciorra et al., 1979	Van Essen, et al., 1991	Faas et al., 2002	Lim et al., 2004	Meins et al., 2005	Present case
Mental retardation		+	+	NA	+	+	+	ND	+	+
Growth retardation	prenatal	-	-	+	+	-	-	ND	-	-
	postnatal	+	+	+	+	ND	+	ND	ND	+
Facial features	microcephaly	+	+	-	+	-	-	ND	+	+
	synophrys	+	+	ND	+	+	-		-	+
	long eyelashes	+	+	ND	+	+			-	+
Extremities	small hands and feet	+	+	ND	ND	ND	+	ND	+	-
Other	hirsutism	+	+	ND	-	+	-	ND	-	-
	congenital heart defects	ND	ND	+	+	+	+	+	-	-

Adopted from Faas et al., Clinical Genetics 2002 62:315-320; NA, not available; ND, no data available; +, present; -, absent.