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Phenotypic and behavioral variability within Angelman Syndrome group with UPD

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Abstract

The Angelman syndrome (AS) (developmental delay, mental retardation, speech impairment, ataxia, outbursts of laughter, seizures) can result either from a 15q11-q13 deletion, or from paternal uniparental disomy (UPD), imprinting, or UBE3A mutations. We describe here the phenotypic and behavioral variability detected in eight UPD patients out of a group of 58 AS patients studied. All of them presented developmental delay, mental retardation, ataxia, speech impairment, and frequent drooling. Only one had microcephaly, whereas in two of them the OFC (head circumference) was above the 98th percentile. The weight of all patients was above the 50th percentile, and in three of them the height was above the 90th percentile. Three were able to say a few words and to communicate by gestures. Two patients presented hyperphagia, and three presented skin picking, common features in the Prader-Willi syndrome (PWS). Four patients (4/7) had wide-spaced teeth. Five presented seizures, and two others did not manifest frequent laughter. One patient was very different from the others, as he showed a better understanding and abilities to communicate, to play video games and to draw. We suggest here that there seems to be an extreme phenotypic and behavioral variability within the UPD group, and that both typical patients and those with mental retardation, language impairment, happy disposition, and hyperactivity should be tested for AS.

Key words: Angelman syndrome, uniparental disomy, macrosomy, macrocephaly.

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Introduction

Angelman syndrome (AS) (Angelman, 1965) is characterized by hypotonia, severe mental retardation, absent speech, seizures, ataxia, outbursts of laughter, micro and/or brachycephaly, macrostomia, and prognathism. The gait is described as wide-based with arms held flexed and upheld at the elbows (Clayton-Smith and Pembrey, 1992; Fryburg *et al*., 1991; Robb *et al*., 1989).

Approximately 70-75% of individuals with AS have 15q11-q13 deletions which are of maternal origin (Knoll *et al*., 1989; Magenis *et al*., 1990). Furthermore, paternal uniparental disomy (UPD) of chromosome 15 is found in about 2-3% of the patients (Driscoll, 1994; Magenis *et al*., 1990) . About 1-5% of patients have biparental inheritance of chromosome 15, but show abnormal methylation pattern and gene expression. These patients have a mutation in the imprinting center (Buiting *et al*., 1995; Dittrich *et al*., 1996; Ohta *et al*., 1999; Saitoh *et al*., 1996). In addition, there is a class of patients (~8%) with mutations in the UBE3A gene (Kishino *et al*., 1997; Malzac *et al*., 1998; Matsuura *et al*., 1997).

In a previous study (Fridman *et al*., 2000a), we described the clinical and behavioral manifestations of 4 cases of paternal UPD15 among Brazilian AS children, and compared these cases to UPD cases from the literature (Bottani *et al*., 1994; Freeman *et al*., 1993; Gillessen-Kaesbach *et al*.,1995; Malcolm *et al*., 1991; Nicholls *et al*., 1992; Prasad and Wagstaff, 1997; Smeets *et al*., 1992; Smith *et al*., 1997, 1998). We also compared the data with those of our deletion patients (n = 21). We concluded that better speech development, weight above the 75th percentile and OFC (head circumference) in the upper normal range are characteristics that should be added to the spectrum of clinical variability present in the Angelman syndrome.

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In this paper, we compare the phenotypic variability showed by our UPD group consisting of the patients mentioned above (Fridman *et al*., 2000a) plus 4 new cases, and present the unexpected features showed by an AS patient with paternal isodisomy.

Subjects and Methods

Patients

The eight UPD patients (4 boys and 4 girls, ranging in age from 2 ys 7 mo to 21 years) with Angelman syndrome were detected in a group of 58 AS patients diagnosed in our laboratory and referred by neurologists of the Hospital das Clínicas, School of Medicine, University of São Paulo, Brazil.

DNA analysis

The patients were diagnosed by methylation analysis of SNURF-SNRPN exon 1 (data not shown).

Microsatellite analyses were performed with 3 markers within the critical region 15q11-q13, 4-3RCA (*D15S11*), LS6-1CA (*D15S113*), and GABRB3CA (*GABRB3*). Five loci outside the PWS/AS region (D15S131, D15S984 CYP19, D17S117 and D15S115) were studied to distinguish between deletion and UPD (data not shown), and to detect crossover regions. Loci D15S541 and D15S542, located close to the centromere, were analyzed to determine the meiotic origin of the non-disjunction (Robinson *et al*., 1998). Multiplex PCR and polyacrylamide gel electrophoresis of ^{32}P end-labeled amplification products followed the method described by Mutirangura *et al*. (1993).

Results

Patients #2, #3, #6, #7, and #8 were typical AS children with developmental delay, speech impairment, ataxia, happy disposition, and a wide mouth.

Patient #1 was referred to us years ago, and at that time he was diagnosed as having an overgrowth syndrome and a 15;15 translocation (Wajntal *et al*., 1993). Later on, he was tested with the methylation assay, because he showed some AS features, such as: absence of speech, ataxia, outbursts of laughter, late-onset seizures, and also hyperphagia, obesity, behavioral problems and skin picking (features commonly seen in PWS patients). In addition, his weight, length and OFC were above the 90th percentile (Fridman *et al*., 1998).

Patient #4 was clinically diagnosed as having PWS at the age of 3, based on his history of neonatal hypotonia, poor sucking, developmental delay, obesity and absence of speech. His weight and height were above the 97th percentile.

Patient #5, although having developmental delay, was able to play video games and to draw, and to say a few words; his comprehension and non-verbal communication were excellent, and he was the only child in our sample with toilet training; while he did not present frequent laughter, hyperphagia and skin picking were present; his OFC was above the 98th percentile.

Patients 1- 4 were previously described in the report by Fridman *et al*. (2000a). Figure 1 shows patient #5.

Analysis of microsatellites within and outside the PWS/AS region performed in patients 5-8 disclosed isodisomy with the centromeric markers and reduction to homozygosity with the markers localized along the chromosome 15 in patient 5, indicating a post-zygotic event. In patients #7 and #8, the centromeric markers showed heterodisomy; patient #6 was non-informative. Three previously studied patients (#s1, 2, and 4) presented isodisomy, with patients #1 and #4 showing reduction to homozygosity with all markers tested. Patient #2 showed one crossover region, and patient #3 was non-informative regarding the meiotic origin of non-disjunction (Fridman *et al*., 2000 a,b).

A summary of the clinical and behavioral characteristics of our UPD patients is presented in Table I.

Discussion

We reviewed the literature and compared the clinical data from 15 published UPD patients (Bottani *et al.*, 1994; Freeman *et al*., 1993; Gillessen-Kaesbach *et al*.,1995; Malcolm *et al*., 1991; Nicholls *et al*., 1992; Prasad and Wagstaff, 1997; Smeets *et al*., 1992; Smith *et al*., 1997, 1998), including our own 4 patients, with data from our 21 deletion patients. In this comparison, we found other statistically significant clinical differences, besides those already described, i.e., delayed age-at-diagnosis, weight above the

Figure 1 - Patient #5 at 7 years of age.

Table I - Clinical features of AS patients with UPD.

	1(1)	2(1)	3(1)	4(1)	5	6	7	8
Age at diagnosis (yrs)	9	10 6/12	7	3 6/12	7	2 7/12	21	17
Sex	m	f	f	M	M	F	M	F
Maternal age (yrs)	30	40	23	29	37	26	24	23
Paternal age (yrs)	33	47	25	37	39	30	26	31
Genetic mechanism	Isodisomy [t(15;15)]	Isodisomy	Isodisomy	Isodisomy	Isodisomy	0	Heterodisomy	Heterodisomy
Birth weight	3780 g	2400 g	3600 g	3150 g	3900 g	3470 g	3350 g	3100 g
Birth height	50 cm	46 cm	0	48 cm	51 cm	47 cm	50 cm	47.5 cm
Hypotonia (1/4)	0	0	0	+	-	0	-	-
Developmental delay (8/8)	+	+	+	+	+	+	+	+
Weight (centile)	97	75-90	75	>97.5	>97.5	50-75	50-75	75-90
Length (centile)	90-97	90	25-50	97.5	50-75	10-25	75-90	25
OFC (centile)	98	2-50	2-50	50	98	75	50	2
Microcephaly (1/8)	-	-	-	-	-	-	-	+
Brachycephaly (4/7)	+	+	+	-	-	-	+	0
Macrostomia (7/8)	+	+	+	+	-	+	+	+
Protruding tongue (6/8)	-	+	+	-	+	+	+	+
Wide-spaced teeth (4/7)	-	+	+	+	-	0	-	+
Severe mental retardation (8/8)	+	+	+	+	+	+	+	+
Seizures (age of onset - yrs)	6	8	-	-	-	3	13	1 6/12
Speech impairment (8/8)	few words	absence	few words	Absence	few words	absence	absence	absence
Outbursts of laughter (6/8)	+	+	+	-	-	+	+	+
Ataxic gait (7/7)	+	+	+	(2)	+	+	+	+
Independent gait (onset - yrs)	2 8/12	0	3 8/12	-	1 6/12	2 1/12	4	2 6/12
Hyperactivity (3/4)	+	0	+	-	0	+	0	0
Hyperphagia (2/5)	+	0	0	-	+	0	-	-
Skin picking (3/6)	+	-	0	-	+	0	+	-
Frequent drooling (7/7)	+	+	+	+	+	0	+	+

(2) - this patient did not walk at all.
+ = presence - = absence 0 = not known.

75th percentile, capacity to say a few words, and early walking in the UPD cases; prevalence of microcephaly, complete absence of speech, and earlier onset of seizures in the deletion group (Fridman *et al*., 2000a).

In this report, we describe the clinical and behavioral phenotypes observed within our UPD group (Table I). All 8 patients presented the common features of AS, such as developmental delay, mental retardation, ataxia, speech impairment and frequent drooling. However, only one had microcephaly, as opposed to patients #1 and #5, who had an OFC above the 98th percentile. Patients #1, #3, and #5 were able to say a few words and to communicate by gestures. Only patients #1, #2, and #7 had late-onset seizures (at 6, 8, and 13 years of age, respectively). Patients #4 and #5 did not manifest frequent laughter. All but two patients showed weight above the 75th percentile, and patients #1, #2, and #4 presented height above the 90th percentile. Patients #1 and #5 presented hyperphagia, and patients #1, #5, and #7 showed skin picking, features which are common in the Prader-Willi syndrome. Patients #1 and #5 had no wide-spaced teeth. Patient #5 was very different from the others, as he had a better understanding and abilities to communicate, to play video games and to draw.

Previous reports have indicated that in patients with UPD the AS phenotype is milder, as compared to patients with deletions (Bottani *et al*., 1994; Freeman *et al*., 1993; Gillessen-Kaesbach *et al*., 1995; Smith e al., 1997), pointing out that children with UPD have a better physical growth, fewer or no seizures, less ataxia and higher cognitive skills. We suggest here that phenotypic and behavioral variability can also be found within the UPD group, and not only between the deletion and the UPD groups, since we found patients with features ranging from the typical AS phenotype and behavior to some of those also seen in PWS patients, and children with better communication and comprehension skills than usually seen in AS.

In conclusion, we suggest that, in addition to the features previously appointed by us to be included in the clinical variability of AS (Fridman *et al*., 2000a), atypical patients as those with mental retardation, language impairment, happy disposition, and hyperactivity should be tested for AS by methylation analysis of SNRPN exon 1. Thus, the spectrum of phenotypic and behavioral characteristics in the Angelman syndrome seems to be broader than previously described.

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