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A Cytogenetic Study in 120 Turkish Children with Intellectual Disability and Characteristics of Fragile X Syndrome

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We review the evidence for the frequency of the fragile X syndrome (FXS), other X-linked abnormalities, and other chromosomal disabilities of Turkish pediatric psychiatry outpatients with intellectual disability. Reported clinical features and genetic findings were used in cytogenetic screenings to estimate the prevalence of the fragile X (fra X) and other chromosomal aberrations in 120 patients with mental retardation, language disorders, attention deficit hyperactivity, or developmental delay, in comparison with 30 healthy children. Data on the clinical, intellectual and behavioral findings in 14 fra X positive children (11.7%) is presented. Ten of the 120 patients (8.3%) had enlargement of the heterochromatin region of chromosome 9. Other chromosomal aberrations and autosomal fragile sites (FS) were also observed. There was a statistically significant difference in the autosomal and X-linked FS between the study and control groups ($p<0.05$). The tests for the fra X chromosome are likely to be of diagnostic benefit in young children with autism or developmental delay, particularly in speech, and who have large and prominent ears.

Key Words: Chromosomal abnormality, fragile sites, fragile X syndrome, intellectual disability.

INTRODUCTION

Mental retardation, language disorders, attention deficit hyperactivity disorder, and pervasive developmental disorders have been diagnosed with increasing frequency in pediatric psychiatry clinics. There are no specific, biologic tests to uncover the etiology of these disorders. There is convincing evidence that at least some of these cases have an organic and genetic etiology. Fragile X syndrome (FXS) is the leading inherited cause of mental retardation. However, it also causes a spectrum of learning and attention problems without mental retardation. Other clinical features of FXS include macroorchidism, long face, prominent ears, highly arched palate, flat feet, and autistic behaviors.[1] Studies of the fra X chromosome in autistic populations have also been carried out by several authors.[2,3] Most prepubertal males with FXS are mildly retarded, although their reading and spelling skills are relatively advanced in contrast with poor arithmetic performance.[4] Speech disorders, visuospatial disturbances, attention deficit, poor concentration and restlessness and fidgetiness have also been reported in FXS.[5] Many of the genes located on the X chromosome are expressed in the brain. Mutations in any of these genes could lead to X-linked mental retardation (XLMR). XLMR is thought to account for 20% to 25% of all mental retardation. Frequencies of fra X in previous studies have reported as 2.6 to 8.7% among moderate to severely retarded males and 2.9 to 5.4% in mildly retarded females.[6] A high frequency of FXS among Turkish patients with mental retardation of previously unknown etiology has been reported in a well documented study[7] which suggests that further genetic studies are indicated in children with intellectual disability. Cytogenetics must be the first step toward identification of index cases because a range of other chromosomal anomalies in addition to fra X

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can cause mental impairment or developmental delay.

In this article we report the clinical features and genetic findings (using cytogenetic screening to estimate the frequency of FXS, other X-linked abnormalities, and other chromosomal disabilities) of Turkish pediatric psychiatry clinic outpatients with intellectual disability (mental retardation, language disorders, attention deficit hyperactivity, or developmental delay) in comparison with healthy controls.

MATERIALS AND METHODS

Included in the study were 120 children (92 boys and 28 girls) diagnosed from June 2000 to June 2001 at the Child Psychiatry Department of Cukurova University, Faculty of Medicine, with mental retardation, language disorders, attention deficit hyperactivity, or developmental delay. Subjects were analyzed from two regions of mainland Turkey: the Mediterranean (Adana, İçel, Hatay and Osmaniye cities) and Southeast regions (Urfa, Maraş and Diyarbakır cities). Thirty healthy children with no family history of mental retardation, developmental disorders, or attention deficit hyperactivity disorder were also included in this study as a control group. Children in the control group were evaluated at the same pediatric psychiatry clinics. Patients were evaluated and diagnosed separately by two certified pediatric psychiatrists using DSM-IV diagnostic criteria for mental retardation, autistic disorder, language disorders, and attention deficit hyperactivity disorder.[8] Both psychiatrists agreed on all diagnoses. Psychological tests were administered by a certified clinical psychologist. During the evaluation process Ankara Developmental Screening Inventory (ADSI), Porteus Mazes Test (PMT) and Weschler Intelligence Scale for Children-Revised (WISC-R) were used to assess the intellectual disability. ADSI, used to identify the developmental delay, consists of 154 items and represents the general development of the child with the sum of its 4 subscales. The subscales are language/cognitive, fine-motor, gross-motor, and social capability/self attention. The inventory is administered by obtaining specific knowledge for each item from the mother or primary caregiver. ADSI has been developed with the help of various similar inventories used in other countries and the validity and reliability of the inventory has been demonstrated in Turkish children.[9] PMT and WISC-R are widely used to assess IQ in children and the validity and reliability of PMT[10] and WISC-R[11] have been demonstrated in Turkish children. All parents signed informed consent to participate in the study and the ethical committee of the faculty approved the study protocol.

All subjects were evaluated with regard to any genetic causes that might underlie these types of disorders. All had one of these disorders of unknown etiology. Each child was also examined for the fra X chromosome and other chromosome aberrations. It is widely recognized that molecular methods are reliable in the diagnosis of FXS. However these methods are, at present, not available in our hospital. Venous blood was studied in the majority of cases, but in small children, capillary blood (collected by heel or finger prick) was also used. Capillary blood was also obtained from 30 healthy children. The patients referred for diagnostic chromosome study had their lymphocytes cultured in two mediums initially: normal medium (RPMI 1640), and special medium for the expression of fragile sites (FS) (RPMI 1640, Sigma, R6767, without folic acid). Chromosome slides were made according to routine procedures.[12,13] A hundred metaphases of each individual were examined for the presence of fra X chromosomes after solid Giemsa staining. All numerical or structural anomalies, isochromatid groups and/or breaks were recorded according to the International System for Human Cytogenetic Nomenclature.[14] All other chromosome abnormalities were photographed, destained, and subsequently GTG-banded for evaluation. Statistical analyses were done using SPSS 9.0 program for windows. The χ^2 test was used to compare the groups and $p < 0.05$ was accepted for significance.

RESULTS AND DISCUSSION

Table 1 lists the numerical and structural chromosomal rearrangements seen in our patients. The mean ages of the study population and the control

Table 1. Summary of Genetic Findings of 120 Patients

Case no	Sex	Age (years, month)	Normal culture	Special culture (folic acid-sensitive-fragile sites)			
			Karyotype (%)	% Fra X positive cell	Xq(%)	Xp(%)	Auto. Fra(+)
1. S.B.	m	4^{4}	46,XY	47	q22(1)	-	+
2. M.C.	m	7^{3}	46,XY	25	q22,24(2),26(1)	-	+
3. U.Ç.	m	7^{4}	46,XY	12	q26(3)	p11(1), p22(1)	+
4. S.K.	m	7^{8}	46,XY	8	-	-	+
5. D.I.	m	6^{8}	46,XY	4	q21(1),24(2)	p21(2)	+
6. G.B.	m	12^{1}	46,XY	3	q22(1),26(1)	-	+
7. S.A.	m	3	46,XY	3	q26(2)	-	+
8. D.D.	f	11^{5}	46,XX	9	q22(1)	-	+
9. M.K.	m	3^{7}	47,XXY	4	-	p22(1)	+
10. H.B.	f		46,XX	7	q22(1)	p21(1)	+
11. Ţ.C.	m	11	46,XY,9qh+ (100)	10	q22(3),24(1),26(3)	p22(1)	+
12. S.U.	f	2^{7}	46,XX,9qh+ (80)	3	q21(1),22(1),26(2)	-	+
13. B.T.	f	7^{4}	46,XX,9qh+ (65)	5	chtb(X)(q22)(2),q22(2),24(1)	-	+
14. B.D.	f	6^{2}	46,XX,auto fra (60)	4	chtb(X)(q24)(2),inv(X)(q24)(2), q22(5),24(1),26(3),28(3)	p21(2),22(3)	+
15. O.D.	m	5^{7}	47,XY,+21(6)/46,XY	-	-	-	+
16. E.Ö.	f	3	45,X(3)/ 46,XX(97)	-	Q22(1),24(2),26(2)	p22(2)	+
17. Z.B.	f	9^{1}	46,XX,r(21)(p13q22)(84)/ 45,XX,-21(10) /46,XX(6)	-	-	-	+
18. B.Y.	m	3^{7}	46,XY,aneuploidy (6)	-	-	-	+
19. O.A.	M	4^{3}	46,XY,auto fra (27)	-	Q22(1)	chtb(X)(p11)	+
20. C.T.	M	6^{3}	46,XY,auto fra (40)	-	-	-	+
21. D.A.	M	7^{7}	46,XY,auto fra (35)	-	q22(1)	-	+
22. R.Y.	M	8^{9}	46,XY,auto fra (40)	-	chtb(X)(q22)(1)	-	+
23. E.G.	M	2^{6}	46,XY,auto fra (35)	-	q22(2),24(1),26(7)	-	+
24. O.H.	M	6^{10}	46,XY,auto fra (70)	-	-	-	+
25. E.D.	M	2^{3}	46,XY,auto fra (80)	-	q22(2),26(2),28(1)	-	+
26. M.K.	M	6^{7}	46,XY,auto fra (20)	-	q24(2),26(2)	chtb(X)(p22)(1)	+
27. M.S.	M	0^{6}	46,XY,auto fra (50),9qh+(10)	-	-	-	+
28-36	m(8) f(1)		46,XX,9qh+ and 46,XY,9qh+(7-100)	-	chtb(X)(q24)(1),q21(2),22(2),24(2), q26(13),27(1)	p21(2)	+
37-120	m(65) f(17)		46,XX and 46,XY	-	chtb(X)(q22)(10),chtb(X)(q24)(3), chtb(X)(q26)(3) q11(1),13(1),21(6), q22(41),23(1),24(21),25(3),26(107),27(8),28(7)	chtb(X)(p22)(3)	+

auto fra, Autosomal fragility; chtb, Chromatid breackage.

group were $6^{5}(0^{6}\text{-}12^{1})$ and $6^{7}(2^{6}\text{-}12)$ years, respectively. The male/female ratio was 92/28 (76.6/23.3%).

Fragile X syndrome (FXS)

Fourteen patients (11.7%) in the study group had FXS (range 3 - 47%), but FXS was not seen in the normal control group lymphocytes exposed to folic acid-free medium. Six of these patients were less than 7 years of age; 4 of them were 7 years of age. The oldest boy was 12 years of age. Nine of the fra X positive patients (64%) were male and five (36%) were female, and their mean age was $6^{10}(3\text{-}12^{1})$. These patients had other X-linked FS and chromatid breaks and autosomal FS in special culture exposed to folic acid-free medium. Three of the 14 fra X positive patients also had chromosomal heteromorphism (9qh+) and one had autosomal FS in normal medium.

The clinical and psychological data in the 14 fra X positive children are listed in Table 2. The percentage of children with parental mental retardation and/or intellectual disability was 50% and the first- and second-degree parental consanguinity of the FXS children in this study was 25%, compared to 6.7% in the controls. Many studies have been published on the frequency of FXS in retarded male boys from different populations. The prevalence of FXS in Asian population was reported to range from 0% to 11%[6]. Molecular screening studies of Turkish male patients with MR of unknown etiology gave a prevalence of 3.0%.[7] Among Japanese MR subjects, FXS varied between 0.8% and 2.7%.[15,16] An FXS frequency of 2.8% has been demonstrated in a population of Chinese MR individuals.[17] FXS frequencies in previous studies have ranged from 2.6 to 8.7% among moderately to severely retarded males and from 2.9 to 5.4% in mildly retarded females.[6] In our study population, 11.66% (14/120) of male and female patients with mental retardation of unknown etiology, hyperactivity, autistic-like illness, or language disorders had FXS. This relatively higher frequency may be due to some other common FS in the Turkish population, or the high frequency of cytogenetically determined fra X which was misinterpreted. Inconsistency in the results in the study also may have resulted from the difficulty of distinguishing the FRAXA locus from two other FS, FRAXE and FRAXF. Both FRAXE and FRAXF are located in a similar region at Xq27.3-28.[18] However expression of the FRAXD (Xq27.2) region could be confused with the rare FRAXA (Xq27.3), and so give rise to errors in the diagnosis of fragile XLMR. Furthermore, the wide range of frequencies may be due to different sizes and ages of the study population, different diagnoses of the study population, the use of different selection criteria, or the methods used to diagnose FXS.

Clinical symptoms

In 10 of the 14 patients stigmata (considered to be characteristic findings in fra X positive males) were found; i.e. pre-and postnatal overgrowth macrocephaly, highly arched palate (3 patients), large ears (6 patients), and long face (4 patients). Hyperextensible joints were also present in eight patients. Developmental delay was noted in 35.7% of the patients. Only one patient was considered clinically to have a relative increase in testicular volume. Of the 14 patients with FXS, 4 of these patients had a long face before 11 years of age, a symptom which may have been seen in some of the children as young as 4 to 6 years of age. Lubs et al. (1983) reported that the adult features were not clearly seen in seven boys less than 7 years of age.[19] Their primary finding showed that five boys, or 70%, had large, low-set, posteriorly rotated ears. Our FXS patients had a lower incidence (42.9%) of long and/or wide and/or protruding ears, which was the most prominent finding in the young children. In our study, the four affected children (case nos. 5, 7, 10, and 12 in table 2) did not exhibit the physical features but two of them were developmentally delayed. It is reported that the unaffected carrier with normal intelligence has no distinguishing features. A number of different, minor, nonspecific, dysmorphic characteristics have been noted in affected females.[20]

Developmental characteristics

A widespread variation of IQs was noted in these 14 patients. Five patients had moderate mental retardation (IQ 35-50), seven patients had mild mental retardation (IQ 51-70), and two patients were functioning at a low normal level (IQ

Table 2. Summary of Clinical Findings of the Fra-X Positive Patients

Patient No	Sex	Age	IQ	Physical Features	Behavioural Characteristics
1. S.B.	m	4^{4}	64 (ADSI)	Developmental delay, large and prominent ears, high arched palate, long face	- Autistic-like bahaviours, hand-flapping speech delay, short attention span, poor eye contact, hyperactivity, stereotype
2. M.C.	m	7^{3}	55 (PORTEUS)	Relative macrocephaly, large and prominents ears	- Mental retardation, speech delay, speech disorder, aggressivity, impulsivity
3. U.C.	m	7^{4}	46 (WISCR)	Prominent ears relative, macrocephaly	- Mental retardation, speech disorder, hyperactivity, short attention span, impulsivity
4. S.K.	m	7^{8}	40 (PORTEUS)	Large and prominents ears	- Mental retardation, hyperactivity, short attention span, compulsifve speach, speech delay, hand-flapping and biting, stereotype, Impulsivity
5. D.I.	m	6^{8}	50 (ADSI)	-	- Mental retardation, hyperactivity, autistic-like behaviours, stereotipi, impulsivity, aggressivity, speech delay, short attention span
6. G.B.	m	12^{1}	92 (WISCR)	Developmental delay	- Autistic-like behaviours, poor eye contact, hyperactivity, hand-flapping, stereotype, compulsive speach, irrtability
7. S.A.	m	3^{1}	70 (ADSI)		- Hyperactivity, speech delay, short attention span.
8. B.D.	f	11^{5}	45 (WISCR)	Developmental delay, long face, high arched palate, hyperextensibility of joints	- Mental retardation, hyperactivity, impulsivity, learning problems, speech disorder, aggressivity, grimas
9. M.K.	m	3^{7}	58 (ADSI)	Developmental delay, macroorchidism	- Mental retardation, hyperactivity, short attention span, head-banging, speech disorder, speech delay, stereotype
10. H.B.	f	8^{3}	55 (WISCR)		- Short attention span, hyperactivity
11. Ţ.C.	m	11	60 (WISCR)	Large and prominent ears, relative macrocephaly	- Hyperactivity,short attention span, speech disorder, aggressivity
12. S.Ü.	f	2^{7}	50 ADSI)	Developmental delay	- Mental retardation, autistic-like behaviours, hyperactivity, short attention span, hand-flapping, stereotipi, poor eye contact, speech disorders, speech delay
13. B.T.	f	7^{4}	75 (WISCR)	Long face	- Hyperactivity, short attention span, nail biting, impulsivity, learning problems, Hyperminezy
14. B.D.	f	6^{2}	60 (ADSI)	High arched plate, prominent ears, long face	- Hyperactivity, speech delay, irritability, impairment in reciprocal interaction

75 and 92). The mean IQ for this group was 53.6.

The fra X expression in the 14 fra X children in our study varied between 3% and 47%, and no correlation was seen between the percentage of fra X positive cells, patient intelligence or phenotype. A positive fra X screening was found in only 50% of the patients.[20] In obligate female fra X carriers, the screening rate was reported as only 30%.[21] The authors (Ed- subject required as the author name was parenthesized in the preceding sentence) explained this lower incidence of positive screening as a result of the higher age of their study group. In addition to age, Chudley et al. (1983) found a correlation between fra X expression and the intellectual level of female carriers, although they also reported a negative correlation between intelligence and the expression of fra X in female carriers.[22] In our study 36% of the children were developmentally delayed and the parents were often mostly concerned about the relative lack of speech (72%). There were striking deficits in

expressive language ability with poor grammatical structure and a tendency towards stuttering.[23] Distinctive speech characteristics included rapid speech rhythm, preservative speech, and impulsiveness.[24] In 14% (two patients) of our FXS children, there was a history of mental retardation in the children's family and/or maternal relatives. There was a family history of learning disabilities in 14% (two) of the children and hyperactivity in 86% (12 patients). Previous studies of obligate female fra X carriers have shown that 30% or more of the female heterozygotes are cognitively impaired, ranging from mild learning disorders to moderate mental retardation.[20,25] Wolff et al. (1988) reported that more than 50% of their female subjects were either retarded or learning disabled.[21] All children with FXS were found to be slow learners.

Behavioral characteristics

Hyperactivity was noted in 108 of 120 children. They could not sit still for a long time and exhibited overacting and excited behavior. Other common problems were short attention span, poor eye contact and autistic-like behaviors. Mild to moderate Attention Deficit Disorder was observed in 9 patients. They experienced great difficulties in concentrating on a specific object. We observed poor topic maintenance in their speech. Less frequent complaints included impulsive irritability, stereotypic aggressivity, grimacing, hand flapping and biting. Hand flapping, a typical behavioral feature in young fra X boys, was seen in four patients. One exhibited head-banging. A positive family history for mental retardation was found in 50% of fra X children.

In our FXS population, behavior was marked by hyperactivity, discipline problems, temper tantrums, and self-abusive behavior with hand flapping, biting and head banging in 36% (five) of the patients. Autistic-like behaviors, including rocking, spining, poor eye contact and talking to one's self, were seen in 14% (two) of the FXS children. As many as 20% of autistic boys may have FXS[26] and the pediatrician should include it in the differential diagnosis of a child with autistic behaviors. Similarly, some children with FXS in this study exhibited a pattern of autistic like behaviors very similar to the pattern described for boys with FXS.[27] This pattern included abnormalities in social and imaginative play, poor eye contact, language form and stereotypic/restricted behaviors. In FXS or autistic subjects, the effects of the FXS mutation on other genes might account for autistic syndrome. These genes might be involved in other autistic cases.

Enlargement of the heterochromatin region of chromosome 9

Ten (8.3%) of the 120 patients with intellectual disability had enlargement of the heterochromatin region of chromosome 9, nine (90%) of whom were male. Their IQ ranged from 24 to 71. Developmental delay was noted in 40% of patients. Other common physical findings were long face, large and prominent ears, highly arched palate and hyperextensible joints. Hyperactivity and language delay were present in 70% of the study group. Other common problems were short attention span, poor eye contact, impulsivity, aggressivity, irritability, stereotypic, autistic-like behavior, hand-flapping, and biting. Less frequent complaints included atrial septal defect and motor coordination disorder. First and second degree parental consanguinity was noted in 24 patients (20%) and in 2 (6.7%) controls.

The noted enlargement of the heterochromatin region of chromosome 9 (9qh+) in 8% of the study population is of interest. The possible clinical effects of 9qh+ certainly remain unknown but it has been suggested that inv(9) and 9qh+ were associated with various diseases and appear to be unfavorable for human reproduction.[28] Recent studies indicate that the pericentric region of chromosome 9 may be etiologically linked to schizophrenia.[29,30] Further studies are necessary to elucidate the role of 9qh+ in fra X individuals and psychiatric disabilities.

Other cytogenetic anomalies

We identified one patient with Trisomy 21 mosaicism (6%), one with Turner syndrome mosaicism (6%), one with normal ring chromosome 21/partial monosomy 21, and one aneuploidy (6%). Nine patients had autosomal FS in normal culture although some cells in three patients had

other X-linked FS, chromatid breaks and autosomal FS in special culture exposed to folic acid. These patients had common clinical and psychological findings. One of these patients had cerebellar vermis hypogenesis. They had long and/or wide and/or prominent ears (55%) long face (22%), and a highly arched palate (22%). Developmental delay was present in two patients, and one of nine patients with autosomal FS had hyperextensible joints. Head tilting, nystagmus, trembling, and drowsiness were noted in one patient. One had a prominent jaw, small testicles and flat-feet. Their IQ ranged from 20 to 98. Hyperactivity was observed in 70% of the children with speech disorders. Other common findings were short attention span, poor eye contact, hand-flapping and biting, autistic-like behavior, impulsivity, and aggressivity. One patient had Williams Syndrome and deafness.

Clinical and biological examinations in our study showed that the specific genetic diseases described below were associated in 6 patients (case 9, 14, 15, 16, 17, and 18). One of the children with FXS was a 47, XXY male (case 9). In individuals with FS, 5 - 10% of metaphases expressing the FS are aneuploidy as a result of breakage at those sites.[31] There is some evidence that meiotic and mitotic nondisjunction may occur at a higher frequency in fra X carriers.[32] This sex chromosome abnormality has been observed several times in autistic subjects.[26] In Klinefelter syndrome males, mental retardation (IQ < 70) is unlikely but intelligence is often less than that of siblings. Learning disabilities, especially at reading, are common. There is an increased likelihood of a variety of developmental problems that vary in severity, including speech delays and neuromotor deficits. These behavioral characteristics were also noted in our patients.

Case 15 had trisomy 21 mosaicism. The percentage of children with mosaic Down syndrome is considerably higher than the 2% incidence reported in several studies of population surveys.[33] However, it is known that, due to their milder symptoms, mosaic children are usually more frequent among cases of Down's syndrome referred for genetics counseling.

One patient with hyperactivity and speech delay had ring chromosome 21/monosomy 21/normal (case 17). Ring chromosome 21 is due to partial monosomy for the distal long arm of chromosome 21. If the latter predominates, the phenotype may resemble that of trisomy 21. Some frequent features are growth and developmental retardation, mental retardation, microcephaly, and large or low-set ears.

Case 16 was 45, X/46, XX mosaicism. Cognitive abnormalities in Turner syndrome patients have been described as being linked to bilateral hemispheric dysfunction.[34] Preadolescent and adolescent girls with Turner syndrome have been described as having significant difficulty in understanding social cues, having poor peer relationship, needing more structure to socialize, and having a poor self-image.[35] The intellectual and psychosocial characteristic of this syndrome can be quite variable. It is apparent that the X chromosome is an integral component of neurodevelopmental processes crucial to cognition, language, affect, and social behaviors.

One of our patients with developmental delay, hyperactivity, speech delay and deafness had Williams syndrome, i.e. well-recognized multiple malformation syndrome in which virtually all cases have been sporadic, and until recently the etiology was unknown. The characteristic psychological profile indicates delay in motor and perceptual development with relatively good verbal performance and sociability.

One of the children with FXS had autosomal FS positive (60%) (case 14) and also had cerebellar vermis hypogenesis, hyperactivity, speech disorder, aggressivity and short attention span. Neuroanatomical measures of the cerebellar vermis were reported to be strongly associated with the degree of stereotypic and restricted behaviors. The cerebellar vermis plays a role in mediating sensory stimulation and arousal through its connection to the somatosensory, auditory and visual cortices.[36] Maldevelopment of the cerebellar vermis may account for the pathogenesis of stereotyped, ritualistic, preservative, behaviors; impaired social communication; and oversensitivity to sensory stimulation through the cerebellum's role in integrating auditory, visual and tactile stimulation, modulating arousal, and facilitating voluntary shifts in attention.

Fragile sites on chromosome X

We identified 320 folate-sensitive FS along the X chromosome, excluding the Xq27 fragile region, and 27 isochromatid gaps or breaks in the study population. Twenty-one (6.5%) of the FS were at Xp and 299 (93.5%) of them were at Xq. The distribution of these regions on the X chromosome was; 148 at Xq26 (46%), 67 at Xq22 (21%), 35 at Xq24 (11%), 11 at Xq28 (3%), 10 at Xq21 (3%), 8 at Xp22 (2.5%), 7 at Xp21 (2%) and 3 at Xq25 (1%). Isochromatid gaps or breaks were 13 at Xq22 (48%), 6 at Xq24 (22%), 4 at Xp22 (15%), 3 at Xq26 (11%) and 1 at Xp11 (4%).

There was a statistically significant difference in autosomal and X-linked FS between the study population and control group (χ^2 test, $p<0.05$). The overall frequency of FS was higher in the study population but there was no increased occurrence of FS between the study population and the fra-X patients. In the present study, there were also many folate-sensitive FS present on Xpter-Yqter in the FXS children and study population. The higher occurrences of FS were at Xq26, Xq22, Xq24, Xq28, Xq21, Xp22, Xp21 and Xq25. It is apparent that the X chromosome is an integral component of neurodevelopmental processes crucial to cognition, language, affect, and social behaviors. This may be particularly true with respect to the distal and long arm of the X chromosome. This region also includes the locus for another XLMR syndrome. However, four FS are recognized cytogenetically in this region. FRAXD in band Xq27.2 is a common FS and has no pathologic significance. Two rare FS are in band Xq28, FRAXE, associated with a very mild nondysmorphic form of mental retardation.[37] FRAXF suggests no pathologic significance. Many of the genes along the X chromosome are expressed in the brain. Mutations in any of these genes are essential for normal brain development and function, and are potential causes of these XLMRs. The higher expression of FS along with the X chromosome in our patients can lead to a local block of several genes in that region around the FS, leading to a variety of neurodevelopmental abnormalities and mental retardation in individuals.

The higher expression of autosomal FS in our study population was unexpected, although it confirmed that common FS occur frequently and that the occurrence of rare FS is low. This finding was significantly different between the control group and study population ($p<0.05$). Expressed FS can lead to interchromosomal recombination. Demonstration of the autosomal FS can be a problem in some families, and presumably in some individuals, due to the rise of new mutations. In addition several reports have documented a variety of neurodevelopmental abnormalities and mental retardation in individuals with rare FS.[38,39] As the difference in incidence of FS between the study population and normal individuals has been confirmed, the mechanism of the effect is worth considering. Williams and Howell (1976) suggested that break age of the FS at critical stages of development could lead to monosomic cell lines that might persist and have a deleterious affect, either genetically or through poor viability.[40] An abnormal phenotype has been identified in autosomal FS individuals in our study, with nonspecific MR and autistic-like illness in some patients. Abnormal phenotypes may occur in offspring with rare FS as a result of a nondisjunction event included by the FS during early organogenesis leading to mosaic monosomy or partial monosomy.[40] Although the nature of the fundamental genetic defect is unknown, it is likely to be situated at the locus of the microscopically observable FS.[41] Our work adds to this ongoing investigation by concentrating on the recognition of the syndrome in young children less than 12 years of age, which allows earlier intervention.

Conclusion: In this study in a pediatric psychiatry clinic, we found a significant increase in the frequency of genetic etiology in children with mental retardation, language disorders, attention deficit hyperactivity or developmental disorders. However, we noted very few neurological signs. Our experience with 14 FXS pediatric patients diagnosed in the study population has led us to believe that the pediatrician should maintain a suspicion for this disorder in the differential diagnosis of children at a young age with intellectual disability. Common behavioral problems such as severe attentional problems, hyperactivity, and language problems seem to be the most important clinical selection criteria for fra X screening in

prepubertal children. Although some of these features may be seen in children without fra X, we believe that the tests for fra X chromosome are likely to be of diagnostic benefit in young children with developmental delay, particularly in speech, and with large and prominent ears. We have also found the presence of a long face, highly arched palate and a history of unusual or autistic behaviors to be significant. The findings from this study support the contention of an association between autism and fra X. Further studies in large samples are needed. Inter-population studies in targeted disorders will also be helpful.

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