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Further studies on a male monozygotic triplet with schizophrenia: cytogenetical and neurobiological assessments in the patients and their parents

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Abstract We previously described a Swedish set of male schizophrenic monozygotic triplets. In this study the patients as well as their parents were further characterized. By high-resolution chromosomal analysis an extra band at chromosome 15p was found in all the triplets and the father. Microdissection, degenerate oligonucleotide-primed PCR (DOP-PCR) amplification and reverse painting indicates that the extra band probably contains only repetitive DNA sequences with no known effect on the phenotype. Magnetic resonance imaging (MRI) showed similar borderline ventricular enlargement and widened subarachnoid spaces over frontoparietal and basal regions as well as around the pituitary gland (empty sella) in all the triplets. The father also had widened subarachnoid spaces over the frontal and basal regions. The mother had an empty sella indicating widened subarachnoid spaces. All the boys also had a right-sided conductive hearing defect, probably due to malformation and fixation of the ossicular chain. The parents did not present any otological abnormalities. Neuropsychological assessment demonstrated similar marked reductions of attentional, mnestic, and executive functions in all the triplets, but the mother showed a normal pattern. Possible joint etiological mechanisms for the psychological and somatic abnormalities recorded in the triplets are discussed.

Key words Schizophrenia · Monozygotic triplet · Chromosome 15 · Magnetic resonance imaging

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Introduction

Family studies have shown that schizophrenia is more common in the relatives of schizophrenic patients than in relatives of non-schizophrenic controls (Kendler et al. 1993). Twin and adoption studies suggest that genetic factors play a significant role in the familial transmission of the disorder (Kety et al. 1975; Onstad et al. 1991). However, also environmental factors are involved and there is some evidence of gene-environment interaction (Tienari 1991). The relative importance of genetic vs environmental factors and the likelihood of heterogeneity in the pathogenesis of schizophrenia remains unclear.

We recently described a male schizophrenic monozygotic triplet characterized by great similarities regarding symptomatology, course of illness, response to neuroleptic drug treatment, and magnetic resonance imaging (MRI) findings including borderline ventricular enlargement, widened subarachnoid spaces, and empty sella, as well as neuropsychological assessment profile (Härnryd et al. 1995). All the boys also had a right-sided hearing defect with a marked reduction of the ossicular bones on the right side. We now report results from further clinical, cytogenetical, otological, MRI, and neuropsychological investigations of the triplets and their parents aiming at finding new clues to etiological mechanisms producing the concordant abnormalities in the schizophrenic triplet.

Material and methods

All three boys (S, T and M), the mother, and the father had been previously interviewed and examined by two of us (C. H. and E. J.)

with regard to their mental and physical health on several occasions since 1992 (Härnryd et al. 1995). When the psychiatric and psychological investigations (and otological assessments) were made all the triplets were given neuroleptic treatment. S received daily doses of sulpiride 600 mg and fluphenazine 5 mg (sulpiride 600 mg, fluphenazine 5 mg, and clomipramine 25 mg). T and M were treated with zuclopenthixol 20 (20) and 30 (80) mg, respectively.

Psychiatric diagnosis

The Structured Clinical Interview for DSM-III-R (SCID; Spitzer et al. 1986) was performed on the triplet by two psychiatrists (C. H. and E. J.) independently. A similar interview including assessment of personality disorders (Spitzer and Williams 1985) with the mother was performed by C. H. As the father did not want to continue the interview, only a partial SCID interview was performed with the father by E. J.

Chromosome preparation

Metaphase chromosomes were obtained from phytohemagglutinin-stimulated cultures of blood lymphocytes according to standard methods (Johannesson et al. 1991). The chromosomes were analyzed by use of G-banding techniques with a resolution of approximately 550 bands (ISCN 1995). Chromosome 15p was also examined with C-banding techniques (Sumner 1972).

Fluorescence in situ hybridization (FISH) and immunofluorescence

Biotin-labeled probes (Oncor) used in this study were α-satellites for loci D15Z, D13Z1/D21Z1, and D14Z1/D22Z1, respectively, and unique sequences for loci D15S10 (PWCR/ANCR), D15S11 (PWCR/ANCR), and SNRP (PWCR/ANCR) for the region 15q11–q13.

The in situ hybridization was performed according to Pinkel et al. (1986) with slight modifications. The slides were washed in PBS prior to RNAse treatment (100 g/ml in 2 × SSC) for 1 h at 37°C followed by dehydration in ethanol series and air drying. Metaphase spreads were denaturated in a solution of 30 ml of 1.9% NaOH and 70 ml of 99% ethanol for 5 min under absolutely stable conditions, dehydrated with three rinses of cold ethanol (70, 90, and 99%), and air dried.

The probes were denaturated and pre-annealed according to respective protocol (Oncor, Gaithersburg, Maryland, USA). Ten microliters of probe and hybridization mixture were applied under a 25- × 25-mm coverslip to each slide. Hybridization was carried out in a humidified chamber at 37° C for 16 h. After hybridization, the slides were washed at 72° C for 5 min in 0.5 × SSC for α-satellite D15Z1, 0.25 × SSC for the α-satellites D13Z1/D21Z1 and D14Z1/D22Z1, and 2 × SCC for the probes D15S10, D15S11, and SNRP. Signal detection was achieved by incubation in fluorescin-avidin followed by one amplification cycle with biotinylated goat anti-avidin antibody and fluorescein-avidin (both from Vector Laboratories Burlingame, California, USA). Following wash and dehydration, counterstaining was performed in propidium-iodide (0.2 mg/ml) in glycerol containing 2% 1,4 diazabicyclo-(2,2,2) octane (DABCO) as antifade. Hybridization sites were detected by use of a Zeiss (Jena, Germany) Axiophot microscope with appropriate filter and photographed with Kodak (Rochester, New York, USA) Ektachrome Panther 400 ASA slide film.

Uniparental disomy of 15 p and proximal 15q

DNA was isolated from venous blood according to Luthman and Datta (Geijer et al. 1994). The polymerase chain reaction (PCR) used was in accordance with standard procedures at the laboratory. Loci GABRA5 and D15S817 were studied. These loci were selected because both the mother and the father carried two different alleles for each locus.

Chromosome microdissection, degenerate oligonucleotide-primed PCR (DOP-PCR), and FISH

Chromosome preparations were made on coverslips and directly GTG banded. Microdissection was performed according to Senger et al. (1990), using an inverted phase-contrast microscope (Axiovert 135; Zeiss, Jena, Germany) and a micromanipulator (Narishige MMO-2YD, Tokyo, Japan). Eight fragments containing the aberrant band on chromosome 15 were excised and transferred to a 10 nl collection drop containing 10 mM TRIS-HCl pH 7.5, 10 mM NaCl, 0.1% SDS, and 0.5 mg/ml proteinase K. The collection drop was then fused with another drop containing 10 nl of fresh collection solution and incubated at 60°C for 2 h in a waterbath.

After digestion with proteinase K, the collection drop was transferred to a 250-µl reaction tube containing 5 µl of PCR mixture: 5 µM 6-MW-primer - 5'CCG ACT CGA GNN NNN NAT GTG G 3'-, 200 µM of each dNTP, 1 µl of Thermo sequenase buffer, 4 U Thermo sequenase (Amersham Life Science, Little Chalfont, UK). DOP-PCR was performed in a Perkin Elmer Thermal Cycler 2400 according to Telenius et al. (1992) with minor modifications. After initial denaturation at 96°C for 5 min, eight low-temperature cycles were run including annealing at 30°C for 1 min and 10 s, 37°C for 1 min, and 95°C for 30 s. Then came the 45 µl PCR-mixture: 1.1 µM 6-MW, 220 µM of each dNTP, 2.5 mM $MgCl_2$, 4.5 µl Stoffel buffer, and 5 U Ampli Taq DNA polymerase Stoffel fragment (Perkin Elmer, Cetus, Stockholm, Sweden) was added, and 32 cycles of 94°C for 30 s, 56°C for 30 s and 72°C for 1 min were run. The amplified material was labeled with SpectrumOrange-dUTP (Vysis, Tuscon, Arizona, USA) in 35 additional DOP-PCR cycles.

FISH was performed as previously described (Blennow and Tillberg 1996). The DOP-PCR library was labeled with SpectrumOrange-dUTP (Vysis, Tuscon, Arizona, USA) by PCR amplification. After dehydration, the slides were mounted in glycerol containing 2.3% DABCO as antifade, and DAPI (4,6-diamino-2-phenyl-indole) at 0.5 µg/ml as counterstain. The signal was visualized using a Zeiss Axioskop fluorescence microscope equipped with cooled CCD camera (Photometrics Sensys, Stuttgart-Fasanenhof, Germany) controlled by a Power Macintosh Quadra 950 computer. Gray-scale images were captured, pseudocolored, and merged using the SmartCapture software (Vysis, Tuscon, Arizona, USA).

MRI of the brain

The MRI technique was performed using a 1.5-T system (Sigma, GE medical systems). All subjects had the head fixed to the table using a plastic helmet system (Greitz et al. 1980). One helmet was shaped to fit the head of one of the triplets. As all three triplets could use the same helmet, almost identical transaxial slices could be obtained. The father and the mother each had a specific helmet. Tl-weighted (TR/TE = 500/17 ms) 4-mm midsagittal images were obtained; T2-weighted (4000/30/120 ms) transaxial images were obtained, using a 4-mm slice thickness and a 2.5-mm gap. The head was also examined with a 3D gradient-echo sequence using 128 transaxial partitions of 1.3-mm thickness. Herniation of the suprasellar cistern ("empty sella") was considered to be present when an obvious bulge of the suprasellar cisterns into the sella turcica was seen in sagittal MR images (Brismar et al. 1978). This was estimated to correspond to a herniation of 3–5 mm. The ventricular system was estimated by using the Evans Index (EI) which was obtained by dividing the maximum width of the frontal horns with the maximum width of the inner skull (Hanson et al. 1975).

Otological and audiological examination

All five subjects were examined by a specialist in audiology (J. B.). The tympanic membrane and the middle ear structures were examined by microscopic inspection. The hearing was assessed with Bekesy or pure tone audiometry. The condition of the middle ears and tympanic membranes were recorded by tympanometry and stapedius reflex tests. Horizontal and positional nystagmus was recorded behind Frenzel's glasses.

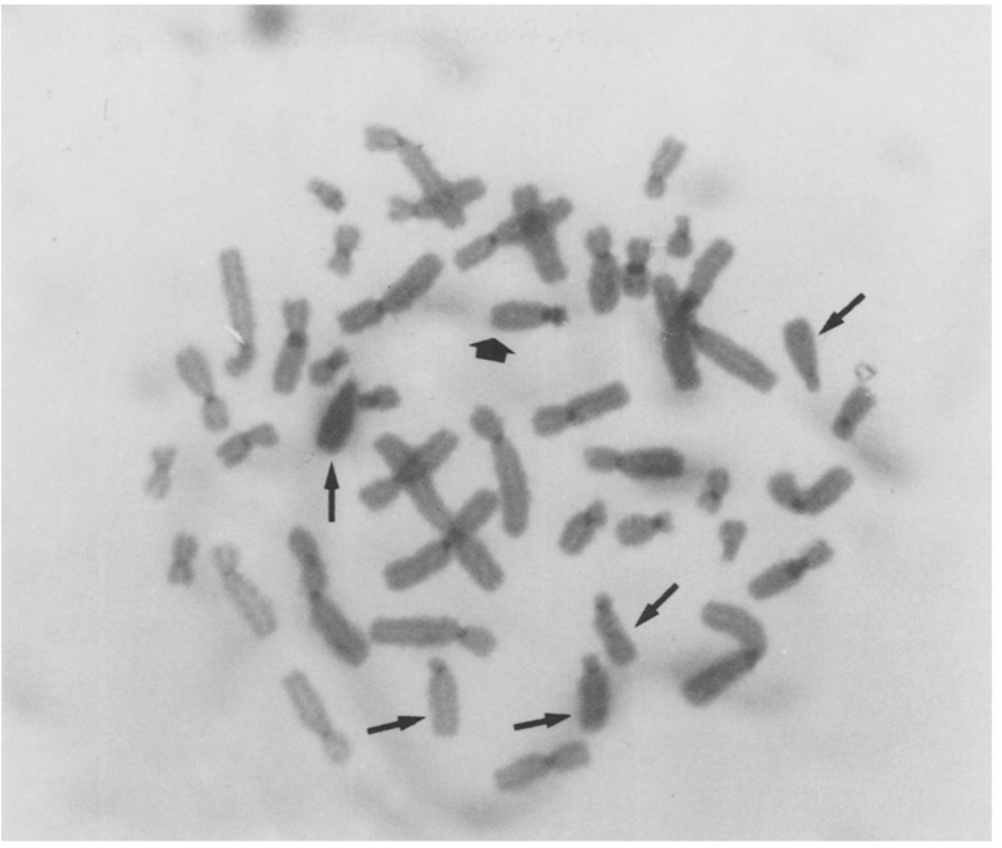


Fig. 1 Karyotype (C-banding) of triplet M. The D-chromosomes 13, 14, and 15 are indicated with *arrows*. Chromosome 15 with the extra band is indicated by the *bold arrow*

Computerized tomography of the temporal bones

Computed tomography of the temporal bones was performed using a high-resolution CT scanner (Toshiba 600S). The triplets were examined in axial and coronal projections in consecutive 1-mm slices.

Neuropsychological assessment

A comprehensive neuropsychological test battery was used to examine the triplets and the mother. The father took part only in a few subtests. Claiming pain after a recent inguinal operation he cancelled the session. A new session was planned but never performed, as the father then was not possible to contact. The battery included all 11 subtests of the WAIS-R (Wechsler 1981), Claeson-Dahl's verbal learning and retention test (Claeson and Dahl 1971), the Rey-Osterrieth Complex Figure (copy and delayed retention scores), verbal fluency (FAS), the Trail Making Test, and the Wisconsin Card Sorting Test (Lezak 1995). Standardized administration and scoring was used, and the results were compared with normative data presented in the references cited.

Results

Psychiatric diagnosis

According to the SCID interviews DSM-III-R criteria for schizophrenia, undifferentiated subtypes, were fulfilled in all the triplets. The SCID interview of the mother did not reveal any psychiatric or personality disorder. The same was true for the partial interview with the father. Initially, it was difficult to contact the father as he had a secret telephone number. During the different investigations the father has been very skeptical concerning participation. After the MRI investigation and initiation of neuropsychological assessment, it was no longer possible to contact him as he changed to a new secret telephone number and did not answer repeated letters. It was also difficult to complete interviews, during which he seemed to be curious but uncomfortable. He denied us contact with his relatives. It has not been possible to trace the causes of the father's withdrawn behavior. On the surface no evidence for major psychiatric illness was found. He lived in a well-cleaned and furnished apartment. He exercised regularly by swimming and walking. He claimed to have friends with whom he met regularly. Medical records showed that he had to retire at age 62 years due to back pains. There were no case notes giving hints of major psychiatric disorder.

Genetics

With C-banding an extra band was found on the short arm on chromosome 15 in all the triplets and the father (Fig. 1). The mother had no detectable abberances in her karyotype. FISH and immunofluroescence showed an expected pattern, but did not reveal any signals at chromosome 15p with any of the probes investigated. For the study of uniparental heterodisomy GABRA5 was informative with the father having two different alleles and the mother hav-

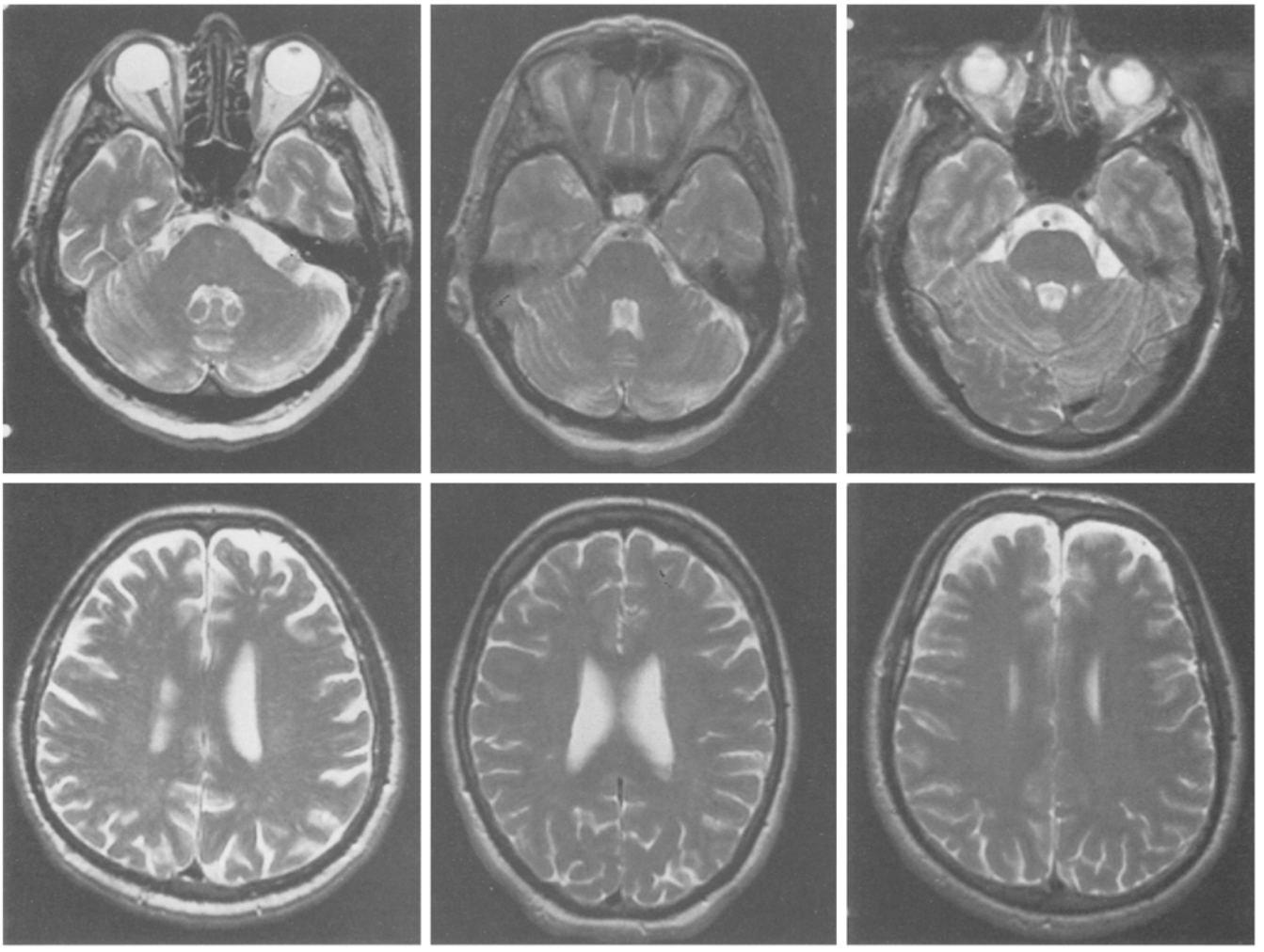


Fig. 2 Transaxial T2-weighted MR images of the father *(left)*, the mother *(middle)*, and triplet M *(right)*. The cerebello-pontine cisterns are markedly enlarged in the father *(upper left)* and the triplet *(upper right)*, but are normal in the mother *(upper middle)*. The frontal subarachnoid spaces are enlarged in the father *(lower left)* and the triplet *(lower right)*, but are normal in the mother *(lower middle)*

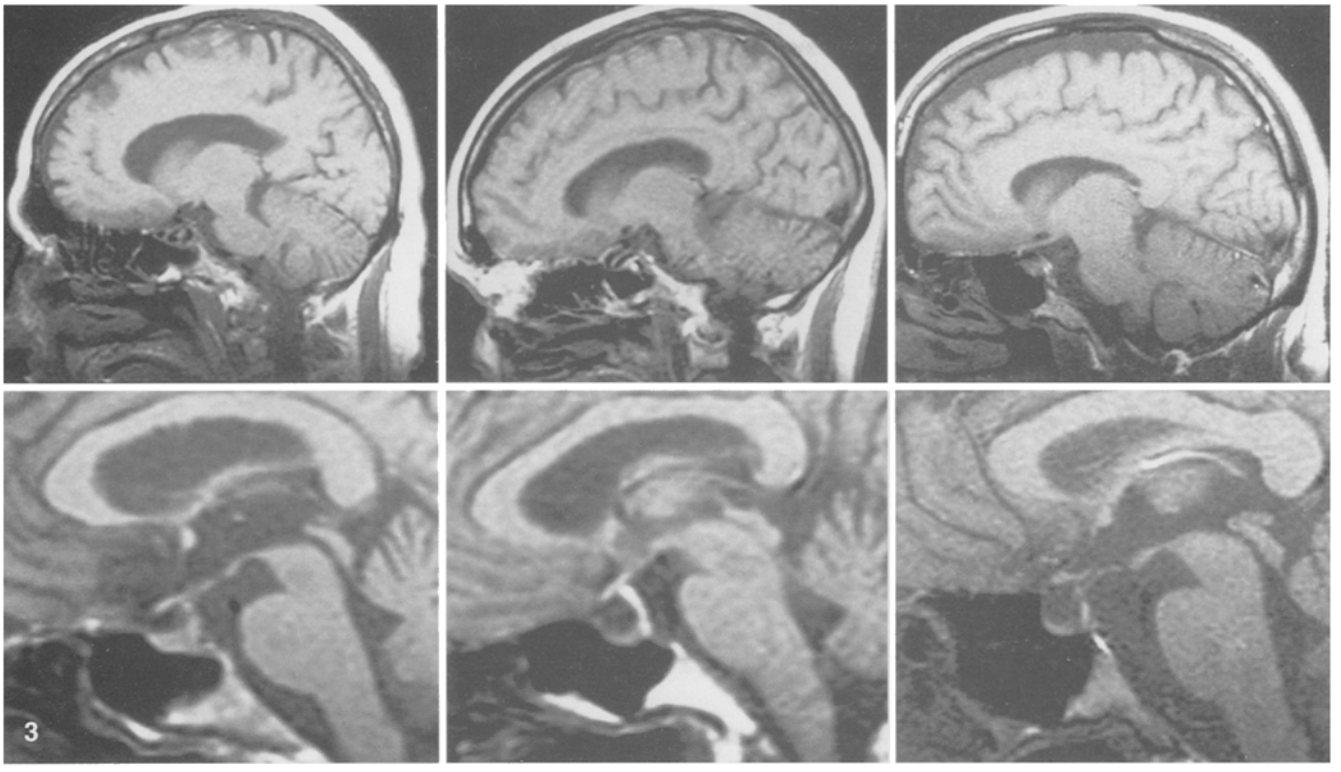
ing another two different alleles. The three triplets had all received one allele from the father and the other from the mother. Both GABRA5 and D15S817 were informative for parental isodisomy. No support for isodisomy was found. Microdissection of the aberrant 15p region, in combination with DOP-PCR amplification and reverse painting, showed labeling of the repetitive regions on the short arms of the acrocentric chromosomes (13, 14, 15, 21, 22) only. No other specific labeling was found, indicating that the extra C-band solely contains repetitive material with no known effect on the phenotype.

MRI of the brain

The MRI results of the triplets have previously been reported (Härnryd et al. 1995). The MRI examinations demonstrated wider subarachnoid spaces in frontoparietal and basal regions in all the triplets as compared with normal subjects (Figs. 2, 3). The father also had wider frontal and basal subarachnoid spaces (Figs. 2, 3). The cortical sulci were generally not widened (less than 5 mm; Gyldensted 1977) in any of the subjects. The ventricular

Fig. 3 Sagittal T1-weighted MR images taken 1 cm lateral to the ▶ midline *(upper row)* and in the midline *(lower row)* of the father *(left)* the mother *(middle)* and triplet M *(right)*. The frontal subarachnoid spaces are enlarged in the father *(upper left)* and the frontoparietal spaces are enlarged in the triplet *(upper right)*, but are normal in the mother *(upper middle)*. The suprasellar cisterns are enlarged and herniate into the sella turcica (empty sella) in the triplet *(lower right)*. Also the mother displays an empty sella *(lower middle)*

Fig. 4 Computed tomography images of the temporal bones of the ▶ triplets. The *arrows* indicate the tympanic cavity with the ossicular chain in center. *Upper left, upper right:* axial sections of the malfunctional right middle ear of triplets T and M. The head of malleus is normal, but the head and short process of incus are underdeveloped. *Lower left:* axial section of the normal left middle ear of triplet T showing normal head and normal short process of incus. The other two triplets demonstrated identical normal left middle ear morphology. *Lower right:* coronal section of the malfunctional right middle ear of triplet S showing coarse ending of the long process of incus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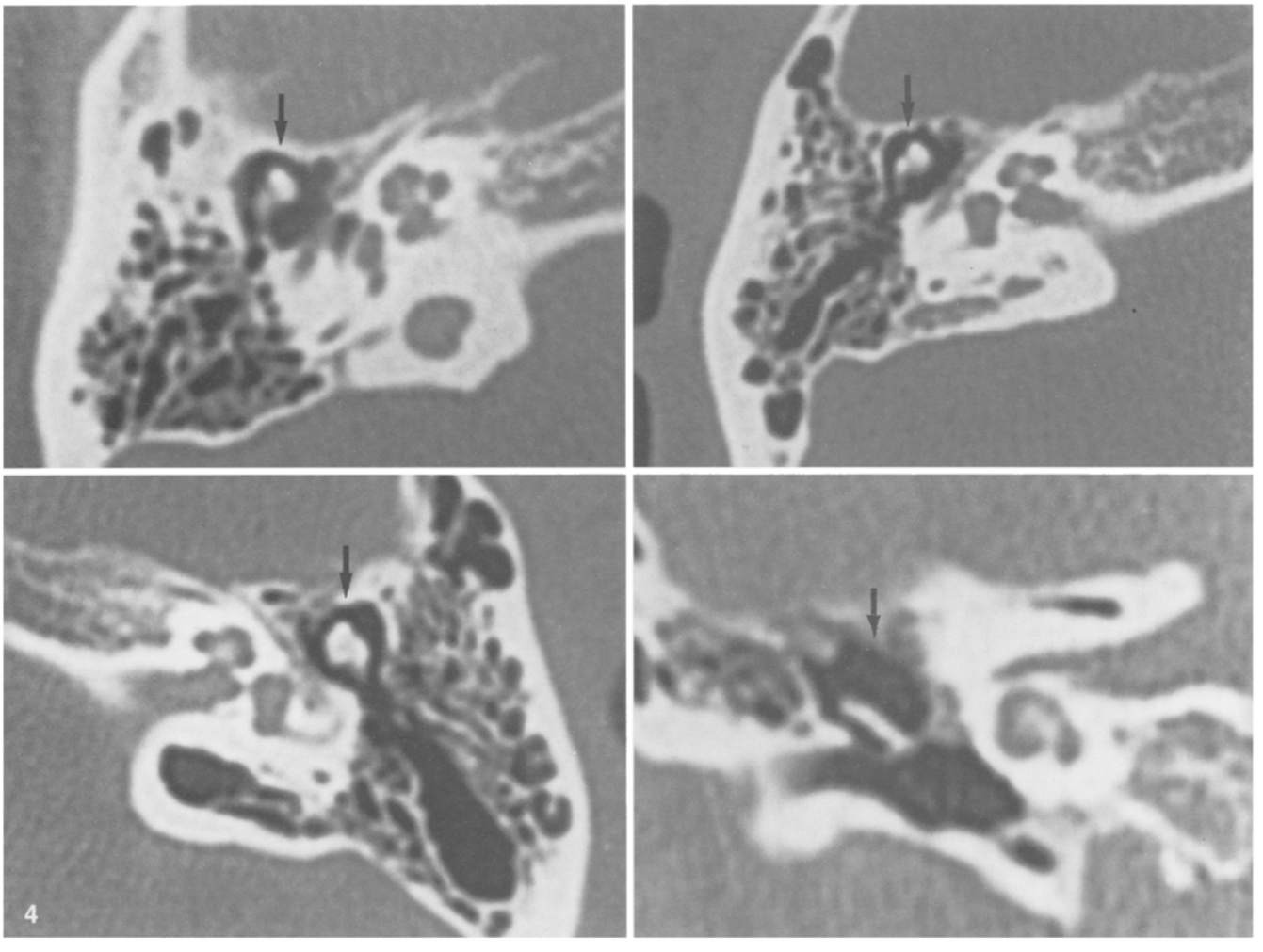
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Table 1 Anatomical features of the right temporal bones in the triplets as assessed with CT. The different parts of the ossicles were assessed as underdeveloped if the parts could be demonstrated but not having normal size or extent. The coarse ending of the long process of stapes could not be explained

	Triplet M	Triplet S	Triplet T
Malleus			
Head	Normal	Normal	Normal
Long process	Underdeveloped	Missing	Underdeveloped
Incus			
Head	Underdeveloped	Underdeveloped	Underdeveloped
Short process	Underdeveloped	Underdeveloped	Underdeveloped
Long process	Coarse end	Coarse end	Missing
Stapes			
Ultrastructures	Missing	Missing	Missing
Oval window	Open	Closed	Open

system in T and M was at the upper border of the normal limit (EI = 0.30). There was also an intrasellar cisternal hernation, "empty sella" (Robertson 1957; Brismar et al. 1978), which was moderate in S and marked in T and M (Fig. 3). Also the mother carried an "empty sella" (Fig. 3).

Otological and audiological assessments

The triplets and mother were previously examined at age 12 and 45 years, respectively. At the present examination their ages were 35 and 71 years, respectively. The father was examined at age 67 years.

The otoscopical examination and the audiological tests were without any remarks in both parents. In neither the triplets nor the parents was there any medical history of recurrent otitis media or of vestibular affections.

In all triplets the otoscopic findings were identical; the tympanic membranes were normal in both ears. However, in the right ear the long arm of malleus was abnormally short. In the same ear the audiometry demonstrated a maximal conductive loss possibly due to malformation and fixation of the ossicular chain. The middle ear function was normal in the left ear. The inner ear function was normal in both ears for all three boys. Compared with the first investigation, no change in subjective hearing or in the hearing tests was recorded at follow up 23 years later. Subjective vestibular symptoms were not met in any of the triplets at follow-up. However, nystagmus was recorded in two of the brothers. M had a spontaneous nystagmus and T had a position-induced nystagmus.

CT of the temporal bones

The CT examinations in the triplets gave a next-to-uniform pattern of ossicular malformation of the right ear and normal findings in the left. Thus, in all the triplets malleus and incus malformations could be demonstrated in the right ear, but stapes ultrastructures were not demonstrable (Fig. 4; Table 1). The ossicular chains of the left ears were normal. There was normal inner ear morphology in all six ears examined.

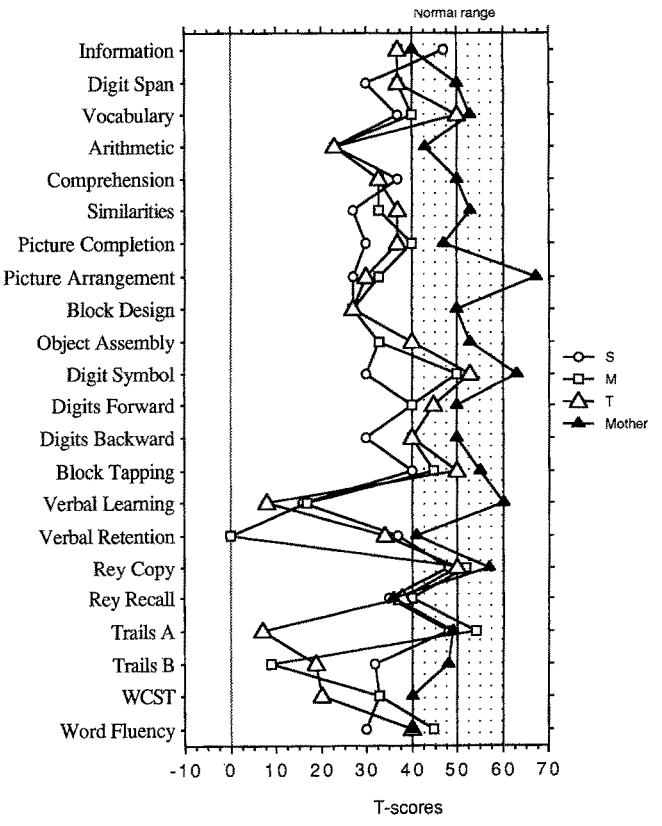

Fig. 5 Neuropsychological performance profiles of the triplets (S, M, and T) and the mother. All raw scores were transformed into standard T-scores according to published results from normal subjects. WCST Wisconsin Card Sorting Test

Neuropsychological functions

Neuropsychological test results of the triplets have previously been reported (Härnryd et al. 1995). By assessment of school records and the results in some of the WAIS-R subtests the premorbid intellectual levels were estimated to have been normal in all the triplets. At the time of the present examination, approximately 14 years after falling ill, and when receiving neuroleptic maintenance treatment (see above) all three patients were found to be clearly impaired in several neuropsychological functions. In Fig. 5 the individual results are shown. In general terms the triplet

had a global impairment of neuropsychological functions. The differences between the brothers were small and the rank order differed in a non-systematic way in different tests. Impairment of attentional, mnestic, and executive functions were prominent in all the subjects. The mother had a profile close to the average after normalization for age and gender (Fig. 5).

Discussion

In the present paper we describe results from clinical, chromosomal, MRI, otological, audiological, and neuropsychological assessments of a set of male monozygotic schizophrenic triplets and their parents.

All the triplets had a clinically similar form of schizophrenia. Neither the father nor the mother showed any obvious schizophrenic symptoms during the interviews. There were no signs of lifetime schizophrenic disorder according to the interviews and available medical records. However, the father had a somewhat anxious, withdrawn, and partly paranoid behavior. He denied us to complete a full SCID interview with him. He interrupted the contact with the research team soon after initiation of the neuropsychological assessment. The reason for his behavior is not clear. The father's behavior may be a normal variant. However, it cannot be excluded that the father carries personality traits which may be associated with schizophrenia or neuropsychological abberances that may explain his behavior.

Interestingly, all the triplets as well as the father shared an extra band on chromosome 15p detectable with C-banding. This strongly indicates that this chromosomal abberation was transferred from the father. The nature of this extra band is not clear. The FISH investigations could not establish that the band is an inversion or translocation involving the centromers of chromosomes 13, 14, 15, 21, 22, or the proximal part of 15q. Microdissection of the aberrant 15p region indicated that the extra C-band probably only contains repetitive material with as yet no known effect on the phenotype. However, the resolution of the method used is approximately 1 kb. Thus, the remote possibility remains that the extra band reflects a pathological mechanism, e.g., a small inversion from a very proximal part of chromosome 15p or a small translocation from other chromosomal regions not traced by the used FISH probes.

Magnetic resonance imaging of the brain demonstrated that all the triplets tended to have a general widening of the ventricular system. Thus, their EIs were at the upper range of the normal variations. Moreover, there was a widening of subarachnoid spaces in frontal, parietal, and basal areas. All triplets also showed signs of an "empty sella," a pathological expansion of the suprasellar cistern into the sella turcica. Neither the father nor the mother showed any evidence of enlarged ventricles. However, the father's frontal and basal subarachnoidal spaces were widened similar to the triplet. The mother had an "empty sella" but was otherwise within normal range. Thus, all the triplets as well as both parents showed some but variable evidence for widened subarachnoid spaces.

The triplets all showed evidence of right-sided conductive hearing loss. Two of the three triplets displayed nystagmus at examination. The father and the mother had normal outer and inner ear function. Severe hearing loss has been associated with psychosis and schizophrenia, whereas slight hearing impairment was not overrepresented among preschizophrenics in a recent Swedish study (Cooper 1976; David et al. 1995). Interestingly, also middle ear disease has been reported associated with schizophrenia (Mason and Winton 1995). No clinically middle ear or other ear disease was apparent in the triplets before the discovery of the right-sided hearing loss at a routine control at the age of 12 years. The changes in the ossicular chain in the triplets, on the same side and with a similar anatomy, makes a genetic rather than an infectious inducement of the malformation more likely. It should be pointed out that such a genetic influence is unilateral. Interestingly, Crow (1990) has suggested that genes altering brain laterality may be of importance to the development of schizophrenia. Thus, in line with the neurodevelopmental hypothesis of schizophrenia (Murray et al. 1992; Waddington 1993; Weinberger 1995) it cannot be excluded that common mechanisms may be responsible for the association between middle ear disease and schizophrenia as well as the unilateral right-sided ear malformation found in the triplet. Interestingly, it was recently reported that homeobox Msx1 deficient mice exhibit among other abnormalities changes in the malleus (Satokata and Maas 1994).

Nystagmus was found in two of the triplets; in one of them nystagmus occurred spontaneously; in the other nystagmus was provoked after the head was turned to the right in supine position. It may be questioned whether the presence of nystagmus is a sign of a peripheral vestibular lesion occurring in connection to the abnormality of the ossicular chain. As an argument against this hypothesis stands the fact that other vestibular symptoms have not been present in any of the triplets. A long-standing impairment of the vestibular system, such as one of congenital origin, should long ago have been compensated, i.e., no nystagmus should be expected to have been demonstrated. Theoretically, neuroleptic drugs could either decompensate a lesion or suppress a nystagmus caused by a peripheral vestibular lesion. However, the inner ear function as reflected by the bone conduction threshold in the audiogram in each of the triplets was completely normal which argues against a nystagmus of otogenic origin. The pathological nystagmus found in some patients with schizophrenia may be caused by drugs acting on the central vestibular system. For instance, the combination of neuroleptics and lithium has been suggested to induce downbeat nystagmus (Williams et al. 1988). At the present investigation the triplets were medicated with different neuroleptics. However, none of them has ever been treated with lithium. The occurrence of spontaneous nystagmus in one of the triplets, position-based nystagmus in a second, whereas no nystagmus was present in the third,

suggests that environmental influences are involved in the genesis of nystagmus in the triplets. Interestingly, the two triplets who displayed nystagmus were both treated with zuclopenthixol, whereas the triplet without nystagmus was treated with sulpiride, a compound known to reduce nystagmus in humans (Mulch 1976). This does not exclude the possibility that all the triplets under similar environmental circumstances may display nystagmus, but that this sign is suppressed in various ways by the medication. In such a case also the nystagmus would rely upon a common genetic basis, although modified by environmental stimuli. Interestingly, a balanced 7;15 translocation has been found to cosegregate with an autosomal dominant congenital form of nystagmus without significant loss of visual function (Patton et al. 1993).

Neuropsychological assessment demonstrated a similar degree of general reduction of attentional, mnestic, and executive functions in all the triplets. The mother performed within normal limits for age and gender. The father refused to participate. These results are in line with previous research where a generalized cognitive impairment has been found in schizophrenic patients when compared with control subjects (Nyman 1992; Saykin et al. 1994).

Although the cytogenetic extra band may well be without functional significance, it is tempting to speculate in possible connections between this band and the results of the other assessments. If the cytogenetic abnormality indeed reflects pathology, it is possible that any of the breakpoints may interrupt a DNA sequence giving rise to an abnormal or untranslated protein. In such a case the extra band may reflect a gene involved in brain maturation or growth, since the most attenuated signs of widened subarachnoid spaces were found in both the triplets and the father, who all carried the cytogenetic abberation. However, the empty sella, also a sign of widened subarachnoid spaces, found in the mother makes such an interpretation less likely, as the mother lacked the extra band. Interestingly, an association between a genetic marker (D5S111 on chromosme 5p14.1–13.1), linked to schizophrenia (Silverman et al. 1996), and enlarged lateral ventricles and frontal and parietal atrophy was recently reported (Shihabuddin et al. 1996). Similarly to the present results, evidence of enlarged cerebrospinal fluid spaces were found not only in individuals diagnosed with schizophrenia spectrum disorders but also in one subject without such a diagnosis. Recently, midgestational influenza exposure has been reported to be associated with increased cerebrospinal fluid spaces in schizophrenic patients (Takei et al. 1996). This is of interest as the triplets were exposed to influenza in utero during the first trimester according to the obstetric records of the mother (Härnryd et al. 1995).

If the extra band would indicate an inversion from the most proximal part of chromosome 15q, it is of interest that psychoses have been reported in patients with Prader-Willi syndrome, a heritable disease located at the proximal part of 15q (Clarke 1993). Furthermore, a partial trisomy of 15pter-q13.3 and 18q23-qter, as well as a balanced chromosomal translocation (15;8), have been found in patients with bipolar, and in the first case also schizoaffective, disorder (Kunugi et al. 1995; Calzolari et al. 1996). Both these disorders have been suggested to be part of a continuum spectrum of psychoses, including schizophrenia, relying on a common genetic basis (Crow 1990). One may also speculate that the father has traits of some psychiatric disturbance potentially associated with schizophrenia, e.g., paranoid or schizotypal personality disorder. If so, it is possible that the extra band at 15p may indicate relevance also for schizophrenia spectrum disorders. Assuming fully penetrant dominant disorders to be associated with the chromosomal abberation makes schizophrenia, empty sella, the conductive hearing defect, and the neuropsychological deficits not attributable to the extra C-band at 15p. However, assuming the effect of many genes as well as environmental factors to influence the systems mentioned, the possibility for a modifying effect of a putative disturbed gene associated with the extra chromosomal band cannot been ruled out. A possibility is also that the schizophrenia, the widened subarachnoidal spaces, the otological deviances, and the neuropsychological deficits are related to a common DNA abberation, although this is too small to be detected by cytogenetic methods. It is also possible that all or some of these events have occurred independently by chance.

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