Ring chromosome 20 syndrome without deletions of the subtelomeric and CHRNA4—KCNQ2 genes loci

Hatem Elghezal a,*, Hanene Hannachi a, Soumaya Mougou a, Hassene Kammoun b, Chahnez Triki c, Ali Saad a

a Department of Cytogenetics and Reproductive Biology, Farhat Hached University Teaching Hospital, IbnEljazzar Street, Sousse 4000, Tunisia
b Department of Genetics, University Teaching Hospital of Sfax, Tunisia
c Department of Neurology, University Teaching Hospital of Sfax, Tunisia

Received 29 March 2007; accepted 2 July 2007
Available online 6 August 2007

Abstract

Ring chromosome 20 [r(20)] syndrome is a rare disease characterized by refractory epilepsy, moderate mental retardation and particular electroencephalographic disorder with non-convulsive status epilepticus. Here, we report a new case of r(20) syndrome in a 12 year old female who presented minimal dysmorphism, generalised tonic—clonic and absence seizures refractory to medical therapy and behavioural troubles.

Among 20 cytogenetically analysed cells, 14 (70%) exhibited a 46,XX,r(20)(p13q13.3) karyotype and 6 (30%) showed a normal 46,XX caryotype. Interphasic FISH using centromeric probe of chromosome 20 detects the presence of a chromosome 20 monosomy in 7% and a duplicated ring chromosome 20 in 8% of studied cells. Metaphase FISH using chromosome 20 telomeric probes and specific probes of CHRNA4 and KCNQ2 genes detects the absence of any deletion in the ring chromosome 20.

Clinical symptoms of r(20) syndrome are attributed to telomeric partial monosomy generated by ring chromosome and causing an haploinsufficiency of two epilepsy genes CHRNA4 and KCNQ2. However, our patient presents the typical epilepsy disorder but no detectable deletion in the ring chromosome 20. We speculate that clinical features of ring chromosome 20 syndrome are caused by low mosaicism of chromosome 20 monosomy caused by the loss of the ring chromosome 20.

© 2007 Elsevier Masson SAS. All rights reserved.

Keywords: Ring chromosome 20; Epilepsy; CHRNA4 gene; KCNQ2 gene; Mosaicism
1. Introduction

Ring chromosome 20 [r(20)] syndrome is a rare disease characterized by refractory epilepsy, mild to moderate mental retardation, facial dysmorphism and particular electroencephalographic disorder with non-convulsive status epilepticus [1,10].

Ring chromosomes are usually formed by fusion of the deleted telomere ends of both chromosome arms [7]. It is therefore reasonable to speculate that the epilepsy disorder in ring chromosome 20 might be caused by the haploinsufficiency of genes located in chromosome 20 telomeric regions. Two genes, CHRNA4 and KCNQ2, mapped to 20q13.2–13.3 have been identified to be implicated in the mechanism underlying seizure disorders and were considerate as candidate gene in the generation of neurologic manifestation of r(20) syndrome [11–13].

Here, we report a new case of r(20) syndrome in a patient with typical severe seizure phenotype but FISH analysis not detects any deletion in the telomeric regions of chromosome 20, nor in CHRNA4 and KCNQ2 genes loci.

2. Materials and methods

The patient, a 12 year old female, was the first child of an unrelated couple devoid of medical history. She was born after uneventful pregnancy and delivery. Birth weight was 3.2 kg, size was 50 cm and head circumference was 34 cm. Apgar scores were 9 and 10. Developmental milestones were mildly delayed and she began sitting at 8 months and walking at age 15 months. First generalised tonic seizures were observed at 2 years and at 6 years of age, absence seizures were observed several times per day. After many drug trials, she still experiences several seizures per week.

The patient’s electroencephalogram (EEG) showed diffuse high-voltage theta waves (4–5 Hz; 100–150 μV) and occasional spike and wave complexes consistent with the findings of non-convulsive status epilepticus in r(20) syndrome [2,5]. Cerebral MNR was normal.

R banded karyotype analysis was performed on P.H.A. stimulated blood culture using standard procedures. Interphase and metaphase FISH analysis was performed using commercially available Vysis (Downers Grove, IL) whole chromosome 20 painting probe, chromosome 20 centromeric probe and subtelomeric specific probes for the short and the long arms of chromosome 20. FISH study was also performed using two direct-labeled probes from bacterial artificial chromosomes (BACs) clones RP11-939M14 and RP11-358D14 containing, respectively, CHRNA4 and KCNQ2 genes.

3. Results

Among 20 cytogenetically analysed cells, 14 (70%) exhibited a 46,XX,r(20)(p13q13.3) karyotype and 6 (30%) showed a normal 46,XX caryotype. FISH analysis using a whole chromosome 20 painting probe confirmed that the ring chromosome was derived entirely from chromosome 20. Among 500 nuclei and mitosis explored by FISH using centromere probe of chromosome 20, we detect the presence of chromosome 20 monosomy in 7% of studied cells. A duplicated ring chromosome 20 was moreover detected in 8% of studied cells (Fig. 1). Using metaphase FISH with locus specific probes, any deletion was detected in both subtelomeric regions and in CHRNA4 and KCNQ2 genes loci (Fig. 1).
4. Discussion

Ring chromosome 20 is a rare chromosomal disorder characterized particularly by refractory epilepsy with frequent episodes of non-convulsive status epilepticus [2,5,6,9]. Epilepsy in r(20) syndrome may be caused by a partial monosomy since a ring chromosome is thought to arise from deletions of telomeric regions [3,7]. Two epilepsy genes CHRNA4 and KCNQ2 mapped to 20q13.2–13.3 and located within 1 Mbp of 20qter [11,12] were considered as responsible for epilepsy generation if deleted.

However, this is to the best of our knowledge the fourth described case of a ring chromosome 20 patient who has the typical severe epilepsy disorder but not deleted subtelomeric regions [2,4,14]. In previously reported cases of ring chromosome 20 syndrome without detectable telomeric deletion, authors speculated that phenotype can be caused by haploinsufficiency associated with an interstitial deletion including CHRNA4 and/or KCNQ2 genes. To exclude this hypothesis, in this case, FISH analyses were performed using specific probes of CHRNA4 and KCNQ2 genes and did not detect any deletion in these loci.

On the other hand, ring chromosomes are known to be instable and can be lost or duplicated during mitosis. However, r(20) is generally reported in its homogeneous form or in mosaic with normal cells. In this study and using FISH with centromeric and telomeric chromosome 20 probes we detect, in addition with 46,XX,r(20) and 46,XX normal cells, a low mosaicism with chromosome 20 monosomic and trisomic cells caused by the loss and the duplication.

Fig. 1. FISH results. (a) Intact subtelomeric regions of chromosome 20 in normal metaphases (commercial probes). (b) Intact subtelomeric regions in ring chromosome 20 (commercial probes). (c) Probes from BACs clones RP11-939M14 (red) and RP11-358D14 (green) indicate the absence of interstitial microdeletion in CHRNA4 and KCNQ2 genes loci. (d) Subtelomeric duplication in a duplicated ring chromosome 20. (For interpretation of the references to colour, the reader is referred to the web version of this article.)
of the ring chromosome 20, respectively, in 7% and 8% of analysed cells. A previous statistical analysis study of 58 reported cases of r(20) syndrome showed that mental development and age at seizure onset inversely correlated with the ratio of mosaicism between normal and r(20) cells [8]. However, the relationship between clinical manifestations and the ratio of additional low mosaicism with monosomic and trisomic 20 cells was not evaluated. We suggest that this form of mosaicism can be implicated in the phenotype of ring chromosome 20 syndrome and must be considered in the correlation studies between the genotype and the phenotype of this syndrome.

The absence of seizure disorders in previously reported cases of supernumerary ring chromosome 20 speculate that epilepsy in r(20) syndrome was not caused by trisomy 20 mosaicism [10]. Consequently, we consider that clinical features of ring chromosome 20 syndrome are caused essentially by the loss of the ring chromosome 20 involving mosaic chromosome 20 monosomy and not because the mere deletion of the two epilepsy-associated genes CHRNA4 and KCNQ2 located at 20q13.3.

In the future, high density array CGH analysis will be useful to confirm the absence of submicroscopic chromosome rearrangements in these cases and particularly microduplications undetectable using FISH.

Acknowledgements

We thank Dr. Mariano Rocchi (University of Bari, Italy), for the BAC clones. The authors thank Nabila Gaaloul, Jamila Romdhane and Leila Cherif for excellent technical assistance in the cytogenetic and FISH experiments.

References

