Hum Genet (1995) 96:355–356

SHORT COMMUNICATION

Catherine R. Begy · Herman A. Dierick · Jeffrey W. Innis
Thomas W. Glover

Two highly polymorphic CA repeats in the Menkes gene *(ATP7A)*

Received: 3 February 1995 / Revised: 21 March 1995

Abstract Two highly polymorphic CA repeats have been identified in the Menkes gene *(ATP7A)*. These repeats should be useful for prenatal diagnosis and carrier detection in families with Menkes disease and X-linked cutis laxa. The observed heterozygosity for these two repeats was 0.778 and 0.60 in Centre d'Etude du Polymorphisme Humaine (CEPH) families.

Source/description. Two CA repeats, a $(CA)_A$ and $(CA)_B$, were found in the Menkes (MNK) gene (locus *ATP7A*, Genome Data Base (GDB) accession nos. G00-392-040 and G00-437-244) located in introns 2 and 5, respectively (Dierick et al. 1995). The MNK gene encodes a predicted 190-kDa copper-transporting ATPase. Mutations in the gene are responsible for Menkes disease (Das et al. 1994; Kaler et al. 1994) and X-linked cutis laxa (Kaler et al. 1994). Both repeats were isolated by hybridization of a ^{32}P-labeled $(CA)_{11}$ probe to a gridded contig of lambda genomic clones spanning the Menkes gene (Mercer et al. 1993).

Polymorphism. Each repeat detected six different alleles in the Centre d'Etude du Polymorphisme Humaine (CEPH) parents.

Polymerase chain reaction (PCR) primers.

$(CA)_A$:

4512c: 5′-CTTAAATCTTCTGACTCCCAACCC-3′
4513c: 5′-TGTCATGATTGCATCTCTTTGGCT-3′

$(CA)_B$:

1578d: 5′-GCCAAGTATTATGAAGCAAGG-3′
2181d: 5′-TACCAGTCTTGACCCCAAACA-3′

C. R. Begy · H. A. Dierick · J. W. Innis · T. W. Glover (✉)
Department of Pediatrics and Department of Human Genetics,
4708 Medical Sciences II, Box 0618,
University of Michigan, Ann Arbor, MI 48109, USA

Frequency. $(CA)_A$: The observed heterozygosity at this locus was 0.778 in 71 unrelated CEPH parents with a polymorphic information content (PIC) value of 0.591. The allele observed for CEPH no. 133101 is 124 bp. The alleles observed for CEPH no. 133102 are 124 bp and 126 bp.

Allele	$(CA)_n$	Length	Frequency
A1	$n = 16$	114 bp	0.037
A2	$n = 20$	122 bp	0.009
A3	$n = 21$	124 bp	0.243
A4	$n = 22$	126 bp	0.505
A5	$n = 23$	128 bp	0.196
A6	$n = 25$	132 BP	0.009

$(CA)_B$: The observed heterozygosity at this locus was 0.60 in 20 unrelated CEPH parents with a PIC value of 0.446. The allele observed for CEPH no. 133101 is 190 bp. CEPH no. 133102 is homozygous for the 186-bp allele.

Allele	$(CA)_n$	Length	Frequency
B1	$n = 19$	180 bp	0.033
B2	$n = 20$	182 bp	0.033
B3	$n = 21$	184 bp	0.167
B4	$n = 22$	186 bp	0.700
B5	$n = 23$	188 bp	0.033
B6	$n = 24$	190 bp	0.033

PCR conditions. PCR was performed in a 20-µl reaction with 20 ng of CEPH parent DNA using 25 pmole of each primer (4513c and 2181d were end-labeled with gamma ^{32}P ATP), 1.5 m*M* MgCl2, 0.5 m*M* each dNTP, and 0.1 U of *Taq* polymerase (Amersham). For primer set 4512c/4513c, amplification was performed in a Perkin Elmer 480 for 35 cycles of denaturing at 94° C for 45 s, annealing at 65° C for 45 s extension at 72° C for 45 s with a final extension time at 72° C for 10 min. For primer set 1578d/2181d, the conditions of amplification were identical to that mentioned above except for the annealing temperature, which was changed to 57° C.

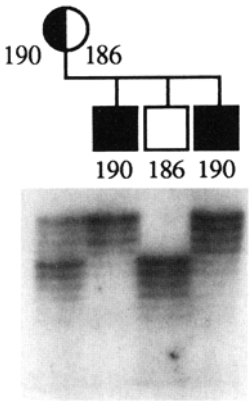

Fig. 1 Analysis of $(CA)_B$ in a family with X-linked cutis laxa. The 190-bp allele cosegregates with the mutant phenotype

Chromosomal localization. The ATP7A gene has been mapped to Xq13.3 (Verga et al. 1991).

Mendelian inheritance. The gene is X-linked.

Comments. Both the $(CA)_A$ and the $(CA)_B$ repeats can be used to determine segregation of disease related alleles in families with Menkes disease and X-linked cutis laxa. Figure 1 shows a pedigree of a family where two of three boys were clinically diagnosed with X-linked cutis laxa. The mother carries the 186-bp and 190-bp alleles of the $(CA)_B$ repeat. The normal son carries only the 186-bp allele, whereas the affected sons carry the 190-bp allele. In conjunction with direct mutation detection (Das et al. 1994), these repeats will be useful in carrier detection and prenatal diagnosis of Menkes disease and its variants.

Acknowledgements This study was supported by NIH grant DK44130 to T.W.G. J.W.I. is supported by a Junior Investigator Award from NICHD (HD28820).

References

Dierick HA, Ambrosini L, Spencer J, Glover TW, Mercer JFB (1995) Molecular structure of the Menkes disease gene (ATP7A). Genomics (in press)

Das S, Levinson B, Vulpe C, Whitney S, Gitschier J, Packman S (1994) Diverse mutations in patients with Menkes disease often lead to exon skipping. Am J Hum Genet 55:883–889

Kaler SG, Gallo LK, Proud VK, Percy AK, Mark Y, Segal NA, Goldstein DS, Holmes CS, Gahl WA (1994) Occipital horn syndrome and a mild Menkes phenotype associated with splice site mutations at the MNK locus. Nat Genet 8:195–202

Mercer JFB, Livingston J, Hall B, Paynter JA, Begy C, Chandrasekharappa S, Lockhart P, Grimes A, Bhave M, Siemieniak D, Glover TW (1993) Isolation of a partial candidate gene for Menkes disease by positional cloning. Nat Genet 3:20–25

Verga V, Hall BK, Wang S, Johnson S, Higgins JV, Glover TW (1991) Localization of the translocation breakpoint in a female with Menkes syndrome to Xq13.2–q13.3 proximal to PGK-1. Am J Hum Genet 48:1133–1138