A hypervariable RFLP within the ABR gene

Deborah DiSilvestre, Christine Morris, Nora Heisterkamp and John Groffen

Section of Molecular Diagnosis, Department of Pathology, Childrens Hospital of Los Angeles, 4650 Sunset Boulevard, Los Angeles, CA 90027, USA

Source and Description of Clone: A 0.6 kb TaqI fragment was isolated from plasmid pVNTR-B; pVNTR-B represents a subcloned fragment from the ABR gene (1).

Polymorphism: TaqI cuts within VNTR-B of the ABR gene generating 1–4 different sized alleles per individual when hybridized with the 0.6 kb probe (see figure). HinfI also cuts within the VNTR. The enzymes BamHI, BglII, BstEII, EcoRI, HindIII and SstI cut outside of the VNTR generating one fragment per allele (1). However, the large sizes of the alleles make it difficult to differentiate between different-sized alleles.

Frequency: Conventional frequency calculations are not possible when the number of different sized fragments seen per individual is variable. Frequencies are given as the fraction of individuals in whom the band was present without regard to intensity (2). Calculations are based on DNAs from 49 unrelated individuals digested with TaqI. 2.05 kb = 0.041, 1.75 = 0.16, 1.51 = 0.10, 1.35 = 0.061, 1.25 = 0.28, 1.05 = 0.12, 0.7 = 0.33, 0.67 = 0.14, 0.54 = 0.02, 0.47 = 0.84, 0.37 = 0.082 and 0.28 = 0.041.

Heterozygosity: A heterozygosity of 78% was observed in 49 unrelated individuals. Cannot be determined; see frequency.

Chromosomal Localization: Initially ABR was localized to 17p13 (1). More detailed *in situ* hybridization to normal and chromosome 17 derivative metaphase spreads support a localization to 17p13.3 (C.Morris, unpublished results).

Mendelian Inheritance: Co-dominant segregation was observed in 3 pedigrees.

Probe Availability: The probe is available through the ATCC.

Other Comments: Post-hybridization washing to a stringency of $0.3 \times \text{SSC}$ at 65°C.

Acknowledgement: Supported by NIH grant 1 R01-CA 47456.

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Dinucleotide repeat polymorphism at the D21S168 locus

Z.Guo, V.Sharma, D.Patterson¹ and M.Litt*

Oregon Health Sciences University, Portland, OR 97201 and ¹Eleanor Roosevelt Institute for Cancer Research, Denver, CO 80206, USA

Source and Description: Cosmid ICRFc102D0494 was from a flow-sorted human chromosome 21-specific library. DNA sequences flanking a (GT)₁₉ repeat within AluI subclone C21G-94A of this cosmid (EMBL accession number X53367) were used to design PCR primers.

PCR Primers: (#334)-5'-ATGCAATGTTATGTAGGCTG-3' (#335)-5'-CGGCATCACAGTCTGATAAA-3'

Polymorphism: Allelic fragments were resolved on DNA sequencing gels. Lengths of allelic fragments (nt) were: A1=118, A2=116, A3=114, A4=112, A5=110, A6=108, A7=106, A8=104.

Frequencies: Allele frequencies in 32 unrelated European Caucasians: A1=.047, A2=.063, A3=.25, A4=.34, A5=.22, A6=.047, A7=.016, A8=.016, PIC=.073.

Chromosomal Localization and Mendelian Inheritance: PCR of genomic DNAs from a somatic cell hybrid panel (1) indicated localization to Region C1, corresponding to 21q22.3. Linkage analysis in 2 CEPH families showed no recombination in 14 informative meioses with D21S3 (previously localized to 21q22.3 (1)), giving a maximum LOD score of 3.0 at theta=0. Mendelian inheritance was observed in all cases.

PCR Conditions: We carry out PCR in a total volume of 25 µl containing: 50 ng genomic DNA, 5 pmoles of each primer, 1.5 mM MgCl₂, 200 µM dNTPs, 50 mM KCl, 10 mM Tris-Cl⁻, pH 8.3, 0.6 units Taq polymerase (Perkin-Elmer/Cetus) and 0.01% gelatin. Amplification is for 30 cycles with denaturation at 94°C for 1 min, annealing at 53°C for 1 min and extension at 72°C for 30 sec.

Acknowledgements: This work was supported by Grants GM-32500 and HD17449 from the National Institutes of Health. We thank H.Lehrach for providing us with ICRFc102D0494.

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* To whom correspondence should be addressed