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Dinucleotide repeat polymorphism at the D17S518 locus

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Source/Description: A human genomic Sau3AI/EcoRI fragment was cloned into pBluescript KS+ and selected by hybridisation to poly(dC-dA).poly(dG-dT). The cloned fragment was designated Wis3. Sequencing was performed to determine primer sequences for the polymerase chain reaction. The predicted length of the amplified fragment was 90 bp.

Primer Sequences:

5' GATCCAGTGGAGACTCAGAG 3' (CA strand);

5' TAGTCTCTGGGACACCCAGA 3' (GT strand).

Frequency: Estimated from 58 chromosomes of unrelated CEPH family members.

Observed heterozygosity = 0.76, PIC = 0.67

Allele (bp)	Frequency	Allele (bp)	Frequency
100	0.017	90	0.328
98	0.086	88	0.310
94	0.259		

Chromosomal Localization: Assigned to chromosome 17q11.2-qter by PCR of somatic cell hybrid DNA. Linkage analysis of four CEPH families gave maximum two point LOD scores of 9.82, at $\theta = 0.023$ with NGFR and 6.25, at $\theta = 0.036$ with D17S37.

Mendelian Inheritance: Co-dominant segregation was observed in four informative two generation CEPH families.

PCR Conditions: Amplification was as follows, denaturation at 94°C (3 min), 32 cycles of denaturation at 94°C (1 min), annealing at 54°C (1 min) and extension at 70°C (1 min) using 0.5 U AmpliTaq, 1×GeneAmp buffer and 200 μM dNTPs (Perkin Elmer-Cetus), 100 ng genomic DNA and 10 pmol of each primer in a volume of 15 μl. Allele sizes were determined by comparison to the cloned sequence on 8% denaturing polyacrylamide gels with autoradiography. The sequenced repeat was (GT)₁₆. The sequence has been submitted to EMBL data Library. Accession number = X60690.

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Dinucleotide repeat polymorphism at the D8S161 locus

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Source/Description: A human genomic Sau3AI fragment was cloned into pBluescript KS+ and selected by hybridisation to poly(dC-dA).poly(dG-dT). The cloned fragment was designated Wis2. Sequencing was performed to determine primer sequences for the polymerase chain reaction. The predicted length of the amplified fragment was 157 bp.

Primer Sequences:

5' GATCAAGGAGCATCACATCT 3' (CA strand);

5' TAACATGTCCCCTCATTTGG 3' (GT strand).

Frequency: Estimated from 56 chromosomes of unrelated CEPH family members.

Observed heterozygosity = 0.75, PIC = 0.77

Allele (bp)	Frequency	Allele (bp)	Frequency
165	0.125	157	0.320
163	0.055	155	0.160
161	0.055	153	0.070
159	0.215		

Chromosomal Localization: Assigned to 8q by linkage analysis of CEPH families 1331, 1333 and 1341 which gave maximum two point LOD scores of 12.34 at $\theta = 0$ with D8S34 and 9.93 at $\theta = 0$ with D8S28.

Mendelian Inheritance: Co-dominant segregation was observed in three CEPH families.

PCR Conditions: Amplification was as follows, denaturation at 94°C (3 min), 32 cycles of denaturation at 94°C (1 min), annealing at 51°C (45 sec.) and extension at 70°C (1 min) using 0.5 U AmpliTaq, 1×GeneAmp buffer and 200 μM dNTPs (Perkin Elmer-Cetus), 100 ng genomic DNA and 10 pmol of each primer in a volume of 15 μl. Allele sizes were determined by comparison to the cloned sequence on 8% denaturing polyacrylamide gels with autoradiography. The sequenced repeat was (CA)₂₁. The sequence has been submitted to EMBL data Library. Accession number = X60689.

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