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Dinucleotide repeat polymorphism at the D10S178 locus

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Source/Description: A human genomic RsaI fragment was cloned into pBluescript KS+ and selected by hybridisation to poly(dC-dA).poly(dG-dT). The cloned fragment was designated Wis4. Sequencing was performed to determine primer sequences for the polymerase chain reaction. The predicted length of the amplified fragment was 154 bp.

Primer Sequences:
5′ GGCAACAGCTAGTAGTCATCT 3′ (CA Strand)
5′ CTTTGGTCTGTTCAGCGCTAC 3′ (CA Strand)

Frequency: Estimated from 66 chromosomes of unrelated CEPH family members and unrelated individuals. Observed heterozygosity = 0.55, PIC = 0.49.

Allele (bp)	Frequency	Allele (bp)	Frequency
168	0.015	160	0.090
166	0.030	156	0.015
164	0.075	154	0.685
162	0.075	152	0.015

Chromosomal Localization: Assigned to chromosome 10 by PCR of somatic cell hybrids.

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 55°C (1 min) and extension at 72°C (1 min) with a final extension step of 72°C for 5 min using 0.5 U AmpliTaq, 1× GeneAmp buffer and 200μm dNTPs (Perkin Elmer-Cetus), 100ng genomic DNA and 10 pmol of each primer in a volume of 15μl. Allele sizes were determined by comparison to the cloned sequence on 6% denaturing polyacrylamide gels with autoradiography. The sequenced repeat was $(CA)_{18}$. The sequence has been submitted to EMBL Data Library. Accession number = X62982.

Other Comments: The cosmid containing D10S178 overlaps with the D10S179 containing cosmid.

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Source/Description: A human genomic RsaI fragment was cloned into pBluescript KS+ and selected by hybridisation to poly(dC-dA).poly(dG-dT). The cloned fragment was designated Wis5. Sequencing was performed to determine primer sequences for the polymerase chain reaction. The predicted length of the amplified fragment was 121 bp.

Primer Sequences:
5′ GCCCACTTTCAGATTCCTGCT 3′ (CA Strand)
5′ GCAGGGAGAAGGACTATGCAT 3′ (GT Strand)

Frequency: Estimated from 64 chromosomes of unrelated CEPH family members and unrelated individuals. Observed heterozygosity = 0.82, PIC = 0.80.

Allele (bp)	Frequency	Allele (bp)	Frequency
141	0.078	125	0.015
139	0.078	123	0.204
137	0.094	121	0.328
135	0.094	119	0.015
131	0.064	117	0.015
127	0.015		

Chromosomal Localization: Assigned to chromosome 10 by PCR of somatic cell hybrids. Assigned to chromosome 10p by linkage analysis of three CEPH families which gave maximum two point LOD scores of 4.76 at $\theta = 0.077$ with D10S28 and 3.47 at $\theta = 0.065$ with D10S35.

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 53°C (1 min) and extension at 72°C (1 min) with a final extension step of 72°C for 5 min, using 0.5 U AmpliTaq, 1× GeneAmp buffer and 200μm dNTPs (Perkin Elmer-Cetus), 100ng genomic DNA and 10 pmol of each primer in a volume of 15μl. Allele sizes were determined by comparison to the cloned sequence on 6% denaturing polyacrylamide gels with autoradiography. The sequenced repeat was $(CT)_9(CA)_{18}$. The sequence has been submitted to EMBL Data Library. Accession number = X62983.

Other Comments: The cosmid containing D10S179 overlaps with the cosmid containing D10S178.

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