Dinucleotide repeat polymorphism at the D11S490 locus

X.Y.Luo, G.A.Evans¹ and M.Litt*

Oregon Health Sciences University, Portland, OR 97201
and ¹Salk Institute, La Jolla, CA 92138, USA

Source and Description of Clone: Cosmid 1,16 was from a human chromosome 11q-specific library (1). p1,16-2, a subclone of this cosmid, was sequenced and the sequences flanking a (GT)₁₄ repeat (EMBL accession no. X56514) were used to design PCR primers.

PCR Primers:

(#22314)-5'-CACAAACATTGGCGCAT-3'

(#41919)-5'-TTCTGGGTCACGGTGCTTCA-3'

Polymorphism: Allelic fragments were detected by autoradiography after separation on denaturing polyacrylamide gels. Lengths (nt) were: A1 = 167, A2 = 165, A3 = 163, A4 = 161, A5 = 159, A6 = 157, A7 = 155, A8 = 153, A9 = 147.

Frequencies: from 50 unrelated European Caucasians. A1 = .01, A2 = .07, A3 = .12, A4 = .07, A5 = .09, A6 = .43, A7 = .01, A8 = .02, A9 = .18. The PIC calculated from these frequencies is 0.72.

Chromosomal Localization: 11q23.3, by fluorescence in-situ hybridization (1).

Mendelian Inheritance: Mendelian inheritance was observed in 2 informative CEPH families with a total of 17 children.

PCR Conditions: PCR was performed according to reference (2), using 5' end labeled primer #22314 and an annealing temperature of 54°, 35 cycles. Amplified products are resolved on denaturing acrylamide gels of this type used for DNA sequencing and detected by autoradiography.

Acknowledgements: This work was supported by Grants GM-32500 and HD 17449 from the National Institutes of Health.

References: 1) Lichter, P., Tang, C.C., Call, K., Hermanson, G., Evans, G.A., Housman, D. and Ward, D.C. (1990) *Science* **24**, 64-69. 2) Luty, J.A., Guo, Z., Willard, H.F., Ledbetter, D.H., Ledbetter, S. and Litt, M. (1990) *Am. J. Hum. Genet.* **46**, 776-783.

Dinucleotide repeat polymorphism at the D21S13E locus

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

Oregon Health Sciences University, Portland, OR 97201, USA

Source/Description: Cosmid ICRFc102B11128 was from a flow-sorted human chromosome 21-specific library. DNA sequences flanking an (AC)₁₈ repeat within AluI subclone C21G-K23A of this cosmid (EMBL accession number X54391) were used to design PCR primers.

PCR Primers: (50)-5'-ATCTGATGCCTTTGAATCT-3'

(51)-5'-TATGCCATTCACTGTGATA-3'

Polymorphism: Allelic fragments were resolved on DNA sequencing gels. Lengths of allelic fragments (nt) were: A1 = 125, A2 = 123, A3 = 121, A4 = 117, A5 = 111.

Frequencies: Allele frequencies in 62 unrelated CEPH parents: F1 = .024, F2 = .024, F3 = .69, F4 = .024, F5 = .23. PIC = 0.42. This system is in linkage equilibrium with the TaqI RFLP revealed by probe D21K9 (1) ($\chi^2 = 2.73$, df = 4, $p > 0.5$). With the combined systems, 9 haplotypes were observed in 55 unrelated CEPH parents and the combined PIC was 0.69.

Chromosomal Localization and Mendelian Inheritance: Cosmid ICRFc102B11128 was identified by screening with the D21S13E clone D21K9. Linkage analysis in 8 CEPH families showed no recombination in 46 informative meioses with the TaqI RFLP identified by D21K9 (1) (previously localized to 21q11.2 (2)), giving a maximum LOD score of 13.2 at theta = 0. Mendelian inheritance was observed in all cases.

PCR Conditions: We carry out PCR in a total volume of 25 μ l containing: 25 ng genomic DNA, 5 pmoles of each primer, 1.5 mM MgCl₂, 200 μ M 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 54°C for 1 min and extension at 72°C for 30 sec.

References: 1) Davies, K.E., Harper, K., Bonthron, D., Krumlauf, R., Polkey, A., Pembrey, M.E. and Williamson, R. (1984) *Hum. Genet.* **66**, 54-56. 2) Gardiner, K., Horisberger, M., Kraus, J., Tantravahi, U., Korenberg, J., Rao, V., Reddy, S. and Patterson, D. (1990) *EMBO J.* **9**, 25-34.

Acknowledgements: This work was supported by Grants GM-32500 and HG-00022 from the National Institutes of Health. We thank H.Lehrach for providing us with ICRFc102B11128.

* To whom correspondence should be addressed

* To whom correspondence should be addressed