A frequent human CD20 (B1) differentiation antigen DNA polymorphism detected with MspI is located near 11q12–13

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Source/Description: 2.1 kb EcoRI fragment from the cDNA clone pB1–21A-29 which encodes the B1 (CD20) cell-surface differentiation antigen of human B lymphocytes (1).

Polymorphism: MspI (CCGG) detects a single two-allele polymorphism with bands at either 9.0 kb or 6.0 kb, as well as a monomorphic band of 2.8 kb.

Frequency: Estimated from 93 unrelated Caucasians.
MspI 9.0 kb allele: 0.47
6.0 kb allele: 0.53

Not Polymorphic For: BamHI, BglII, EcoRV, TaqI, PvuII, XbaI, RsaI, and KpnI using a panel of 10 unrelated individuals.

Chromosomal Location: Assigned to human chromosome 11q12–q13 by *in situ* hybridization and further confirmed by use of rodent/human somatic cell hybrids (2).

Mendelian Inheritance: Co-dominant segregation of the MspI alleles observed in 19 families (239 individuals).

Probe Availability: Request for probe to T. Tedder at the above address.

Other Comments: RFLPs were observed under normal hybridization and washing conditions.

References: 1. Tedder *et al.* (1988) *Proc. Natl. Acad. Sci. USA* **85**, 208–212, 2. Tedder *et al.* (1989) *J. Immunol.* **142**, 2555–2559.

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Detection of the XbaI RFLP within the retinoblastoma locus by PCR

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We previously identified an *XbaI* RFLP within the human retinoblastoma susceptibility gene (1). Here we report oligo primer sequences that can be used to analyze this polymorphism using the PCR. This method yields results in as little as 2 days and does not require a radiolabelled probe.

PCR Primers: Sense oligo–5'TTCCAATGAAGAACAAATGG-3'
Antisense oligo–5'-GCAATTGCACAAATCCAAGTT-3'

Polymorphism: *XbaI* digest of the amplified fragment identifies two alleles; A1 = ~945 bp and A2 = ~630 bp + ~315 bp.

Frequency: Allele frequencies were calculated from 50 unrelated individuals. A1 = 0.55 A2 = 0.45.

Chromosomal Localization: The polymorphic *XbaI* recognition site occurs within the 17th intron of the retinoblastoma susceptibility locus (13q14), approximately 21.8 kb downstream of exon 17.

Mendelian Inheritance: Co-dominant segregation patterns were observed in 12 pedigrees with retinoblastoma (see figure for example).

PCR Conditions: We carry out PCR in a total volume of 50 μ l containing: 500 ng of genomic DNA, 20 pmoles of each primer, 2 mM MgCl₂, 200 μ M dNTPs, 50 mM KCl, 20 mM Tris–pH 8.4, and 0.01 μ g/ μ l BSA. The amplification is performed for 35 cycles with an annealing temperature of 50°C. The amplified product is then passed through a 1 ml Sepharose CL-6B column and digested overnight with *XbaI*. DNA fragments are resolved by electrophoresis through a 2% agarose gel.

References: (1) Wiggs, J., Nordenskjöld, M., Yandell, D., Rapaport, J., Grondin, V., Janson, M., Werelius, B., Petersen, R., Craft, A., Riedel, K., Liberfarb, R., Walton, D., Wilson, W. and Dryja, T. P. (1988) *New Engl. J. Med.*, **318**, 151–157.

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