Dinucleotide repeat polymorphism in the IL-2Rβ gene

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Source/Description: Analysis of the published sequence of the human interleukin 2 receptor β (IL- 2Rβ) gene revealed a dinucleotide repeat of the form $(GT)_n$ (1). Using the DNA sequence, polymerase chain reaction (PCR) primers flanking the repeat were designed to yield an amplified fragment with a predicted length of approximately 153 bp.

Primer Sequences:
TG-01: 5'GAGAGGGAGGGCCTGCGTTC3'
TG-02B: 5'CACCCAGGGCCAGATAAAGA3'

Polymorphism and Frequency: Estimated from 11 unrelated individuals.
Heterozygosity = 91%

Allele (bp)	Frequency	Allele (bp)	Frequency
163	0.09	155	0.09
161	0.09	153	0.09
159	0.18	151	0.09
157	0.18	149	0.18

Chromosomal Localization: The IL-2Rβ gene was localized to 22q11.2-q12 by somatic cell hybrid mapping (1).

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

Other Comments: PCR amplification was performed in a total volume of 100 μL containing: 100 ng genomic DNA, 250 nM of each primer, 200 nM of each dNTP, 1.5 mM $MgCl_2$, 50 mM KCl, 10 mM Tris/HCl pH 9.0, 0.01% gelatin, 0.1% Triton X-100, and 1.25 units of *Taq* polymerase (Promega). Primer TG-01 was 5' end labeled with γ-^{32}P-dATP. After denaturation at 94°C for 3 min, amplification was carried out for 30 cycles with denaturation at 94°C for 20 sec, annealing at 55°C for 20 sec, and elongation at 72°C for 30 sec. PCR products were resolved on a 6% polyacrylamide gel and visualized by autoradiography. Fragment sizes were determined by comparison to an M13 sequencing ladder.

References: 1) Gnarra,J.R., Otani,H., Wang,M.G., McBride,O.W., Sharon,M. and Leonard,W.J. (1990) *Proc. Natl. Acad. Sci. USA* **87**, 3440–3444.

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A *NlaIV* polymorphism within the human Factor X gene

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Source/Description: We describe a sequence variant occurring in exon 7 of the gene encoding blood coagulation Factor X. The variant is found at residue 817 of the cDNA (numbering system of Leytus *et al.* [1]), where either a C or a T may be present. The polymorphism is silent at the amino acid level, since both triplets (ACT→'ACC) encode threonine. The latter sequence, but not the former, forms part of a restriction enzyme recognition site for the enzyme *NlaIV*, which recognizes the sequence GGNNCC.

Frequency: Estimated from *NlaIV* restriction digest of PCR-amplified 315 bp fragment containing all of exon 7. Oligonucleotides used to prime amplification were 5' GGATGG-AAGCTTACCGAAGAGGAC 3' and 5' CCTACCTTCG-AATGGCTTCTCCTG 3' (from intron sequences flanking exon 7). In the presence of the D1 allele (ACT), the 315 bp fragment remains uncut by *NlaIV*. In the presence of the D2 allele (A-CC), fragments of 171 and 144 bp are generated. The figure demonstrates homozygous D1, heterozygous and homozygous D2 samples.
Estimated from 17 unrelated Caucasians:
D1 (ACT) = 0.76
D2 (ACC) = 0.24
Estimated from 5 unrelated American Blacks:
D1 = 0.5
D2 = 0.5

Chromosomal Localization: The gene for human F.X has been mapped to 13q34.

Other Comments: The polymorphism may have utility for RFLP diagnosis in F.VII mutations, since F.VII and F.X are closely linked on chromosome 13. This polymorphism differs from others previously reported within the Factor X gene [2].

References: 1) Leytus. *et al.* (1986) *Biochemistry* **25**, 5098–5102. 2) Hassan *et al.* (1988) *Blood* **71**, 1353–1356.

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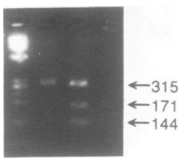

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