AvaII RFLP detected by the anonymous DNA segment p10E5.SC1 [D11S806] on chromosome 11q22–23

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Source/Description: A single-copy 1.4 kb EcoRI/HindIII fragment from cosmid cSalk3–10E5 was subcloned into pUC19. Cosmid cSalk3–10E5 was isolated from a library prepared from the distal arm of chromosome 11 (1).

Polymorphism: AvaII identifies a two allele polymorphism:
A1:0.9 kb
A2:0.6 kb
constant band at 1.4 kb.

Frequency: Estimated from 46 unrelated Caucasians:
A1: 0.67
A2: 0.33
Observed heterozygosity = 0.40.

Not Polymorphic For: AccI, ApaI, BamHI, EcoRI, HindIII, KpnI, MspI, NcoI, NsiI, PstI, PvuII, RsaI, SacI, XbaI, XhoI.

Chromosome Localization: p10E5.SC1 was localized to chromosome 11q22–23 using somatic cell hybrids and linkage analysis.

Mendelian Inheritance: Codominant segregation has been observed in 30 two- and three-generation families.

Probe Availability: Available on request.

Other Comments: No background observed when filters were washed to a stringency of 0.5×SSC/0.1% SDS at 65°C. p10E5.SC1 also identifies a rare two allele polymorphism with TaqI (A1: 3.6 kb, A2: 2.2 kb; allele freq. A1:0.07, A2: 0.93 in 37 unrelated Caucasians).

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Reference: 1) Evans, G.A. and Lewis, K.A. (1989) *Proc. Natl. Acad. Sci. USA* **86**, 5030–5034.

PCR of a KpnI RFLP in the α -L-iduronidase (IDUA) gene

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Source/Description: Oligonucleotide primers were used to amplify a 450 bp region including exon III and part of introns II and III of the human α -L-iduronidase gene. The polymorphism can be used diagnostically in families with α -L-iduronidase deficiency (mucopolysaccharidosis type I) and Huntington Disease, which is closely linked (about 1 Mb) (1) to the IDUA locus.

PCR Primers:
ID-49, 5'-AGGTCCTGCCTGGCTCCTGA-3'
ID-36, 5'-GGCTGGGAGCAGAGCCCCACA-3'

Polymorphism: The 450 bp PCR product contains a polymorphic KpnI site caused by a T→C silent change in the first base of a Leu codon in the IDUA gene (2) and produces fragments of 390 bp and 60 bp after digestion.

Frequency: Studied in 114 chromosomes from 57 unrelated healthy individuals.

allele 1	(450 bp)	0.22
allele 2	(390 plus 60 bp)	0.78
Observed heterozygosity		37%

PCR Conditions: 250 ng of genomic DNA was amplified using 500 ng of each primer, 2 units of Taq polymerase (Cetus) in the recommended buffer, except that the reactions contained 400 μ M dNTPs, 2.5 mM MgCl₂ and 0.1% v/v Triton X-100. After 5 min at 95°C the reactions were subjected to 30 cycles of 95°C for 30 sec, 66°C for 45 sec and 72°C for 45 sec.

Chromosomal Localization: IDUA has recently been reassigned from chromosome 22 to chromosome 4p16.3 (3) and fine mapped to locus D4S111, 1100 kb from the telomere of chromosome 4p (1).

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References: 1) MacDonald, M.E. *et al.* (1991) *Somatic Cell Mol. Genet.* **17**, 421–425. 2) Scott, H.S. *et al.* (1991) *Proc. Natl. Acad. Sci. USA*, in press. 3) Scott, H.S. *et al.* (1990) *Am. J. Hum. Genet.* **47**, 802–807.

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