Two polymorphisms for amino acid substitutions in the APOA4 gene

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Description: Apolipoprotein A-IV is a structural component of chylomicron remnants and a cofactor for LCAT. Amplification of the 3' end of human apolipoprotein A-IV was carried out by the polymerase chain reaction (1) using the oligonucleotides 5'-CGGGTGGAGCCCTACGGGA-3' as the 5' primer and 5'-TGGGGCCTGTGCACAGGGG-3' as the 3' primer.

Polymorphism: By direct DNA sequence analysis, two polymorphic sites were identified within the amplified region, each one resulting in an amino acid substitution (2). Codon 347 may be ACT coding for Thr or TCT coding for Ser. Codon 360 may be CAG coding for Gln or CAT for His.

Frequency:

In 51 unrelated individuals: Codon 347 (Thr) = .784 and 347 (Ser) = .216

In 29 unrelated individuals: Codon 360 (Gln) = .914 and 360 (His) = .086

Chromosomal Localization: 11q13 to 11qter.

Mendelian Inheritance: Codominant segregation of both polymorphisms was observed in multiple families.

Other Comments: For the polymerase chain reaction denaturation was carried out for 1 min at 94°C and annealing and extension for 3 minutes at 59°C in the presence of 5% DMSO. The allele-specific oligonucleotides for typing variation at codon 347 were as previously described (2). The probe detecting the ACT(Thr) codon was GGACAAGTCTCTCTCCC; whereas the probe detecting the TCT(Ser) codon was GGACAAGACTCTCTCCC. Dot blots for detecting variation at codon 347 were washed at 55°C. The allele-specific oligonucleotides for typing variation at codon 360 were GGAACAGCAGCAGGAGC for the Gln codon and GGAACAGCATCAGGAGC for the His codon. Dot blots for detecting variation at codon 360 were washed at 54°C.

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A second NsiI RFLP at the CYBB locus

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Source and Description of Clone: The probe is a 3.5 kb PstI-KpnI cDNA fragment containing most of the transcript of the heavy chain of cytochrome b₅₅₈ in a gemini 4 plasmid (1).

Polymorphisms: In addition to the polymorphic fragments of 1.7 kb (allele A1) and 1.3 kb (allele A2) (2), NsiI reveals polymorphic fragments of 3.3 kb (allele B1) and 2.8 kb (allele B2). Invariant fragments are 7.7, 6.4, 4.1 and 0.95 kb.

Frequencies: 98 independent X chromosomes were analysed. The 3.3 kb B1 allele was present in 9/98 (0.09) and the 2.8 kb B2 allele in 89/98 (0.91) X-chromosomes. In our collection we observed a lower frequency of the 1.3 kb A2 allele (9/98; 0.09) than reported by Battat and Francke (2). The combined PIC is 0.27 compared to 0.15 for either RFLP alone.

Chromosomal Localization: The CYBB locus has been assigned to Xp21.1 (1).

Mendelian Inheritance: X-linked inheritance of the B1/B2 NsiI RFLP was observed in one three-generation family and 2 two-generation families segregating for chronic granulomatous disease (CGD).

Probe Availability: The probe is available from Stuart H.Orkin, Boston Childrens Hospital.

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