Tetranucleotide repeat polymorphism in the apolipoprotein B gene

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Source/Description: The polymorphic (TTTA)_n tandem repeat is located in intron 20 of the ApoB gene at the 3' end of an Alu sequence (1). Two oligonucleotides flanking the repeated sequence, GZ-9 and GZ-10, were used to selectively amplify the sequence from genomic DNA employing the polymerase chain reaction (PCR). Based in the published sequence the expected size of the fragment is 143 bp.

Primer Sequences:

GZ-9 = ACATTGATTCCTTGGAGTTTCTCTA

GZ-10 = GGGCAAAGAGCGAGACTCCATCTC

Frequency: Estimated in 20 unrelated Caucasian American individuals:

Allele (nt)	Number of (TTTA) Repeats	Frequency
151	6	0.25
147	5	0.40
143	4	0.35

The heterozygosity index was 66%.

Mendelian Inheritance: Co-dominant segregation was observed in two families with five informative meioses.

Chromosomal Localization: ApoB gene has been assigned to chromosome 2 (p23-p24).

Other Comments: The PCR reaction was performed on genomic DNA as previously described (3) using end-labeled oligonucleotide GZ-9 and unlabeled GZ-10 with the following modifications: 1) denaturation at 96°C for 1 min, 2) annealing and extension was performed at 68°C for 3 min, and 3) the number of cycles was 25. The PCR product was fractionated on 8% denaturing polyacrylamide gel. The size of the alleles was determined by comparison with end-labeled MspI digested pBR322 DNA.

References: 1) Huang, C.S. *et al.* (1989) *J. Biol. Chem.* **264**, 11394–11400. 2) Law, S.W. *et al.* (1985) *Proc. Natl. Acad. Sci. USA* **82**, 8340–8344. 3) Saiki, R.K. *et al.* (1988) *Science* **230**, 487–491.

Tetranucleotide repeat polymorphism in the apolipoprotein C-III gene

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Source/Description: A polymorphic (TTTC)_n repeat is located in the third intron of the ApoC-III gene at the 3' end of an Alu sequence (1). The length polymorphism can be detected by performing two consecutive polymerase chain reactions (PCR). In the first reaction, oligo GZ-11 and GZ-13 are used to amplify a 847 bp fragment. The reaction product is subjected to a second amplification using oligonucleotides GZ-11 and GZ-12 which generates a band of the expected length of 545 nt.

Primer Sequences:

GZ-11 = GAGTCCCAGGTGGCCAGCAGGCCAG

GZ-12 = CAGTGAGCCGAGATGGCACCACTGC

GZ-13 = GTCCACCACAAAAGTGGTCCCTGGTG

Mendelian Inheritance: Co-dominant segregation was observed in one family with nine informative meioses.

Chromosomal Localization: ApoC-III gene has been assigned to chromosome 11q23-qtr (2).

Other Comments: The PCR reaction was performed as previously described (3) with the following modifications; 1) the DNA was denatured at 96°C for 1 min, 2) annealing and extension was done at 68°C for 3 min, 3) the number of cycles was reduced to 25. In the first reaction 1 µg of genomic DNA was amplified in 50 µl using unlabeled oligos GZ-11 and GZ-13. One fifth of the amplification reaction was re-amplified using end-labeled oligo GZ-12 and unlabeled oligo GZ-11. The final product was fractionated on 4% denaturing polyacrylamide gel. Seven different alleles were found that differed in size by integrals of four nucleotides. The heterozygosity index was 83%.

References: 1) Protter, A.A. *et al.* (1984) *DNA* **3**, 449–456. 2) Cheung, P. *et al.* (1984) *Proc. Natl. Acad. Sci. USA* **81**, 508–511. 3) Saiki, R.K. *et al.* (1988) *Science* **230**, 487–491.