Detection of a length polymorphism for human GIP gene by polymerase chain reaction

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Source and Description: Takeda et al. (1) reported a cDNA sequence for human GIP. We selected primers for polymerase chain reaction (PCR) from the 3' end of the coding sequence and extending into the untranslated region of the human GIP gene. The forward primer is 5'-CACAATGGGCTCGACTTAGCTAA-3', the reverse primer is 5'-CTTGCTGGATCAGAACCCTG-3'. The expected amplification product based on cDNA sequence was 165 bp, however, a 1800 bp product was observed, implying these primers span an intron.

Protocol: Each amplification reaction consisted of 100 ng genomic DNA, 10 ng of each primer, 1× reaction buffer as supplied by manufacturer (Promega), 200 µM of each nucleotide triphosphate, and 1.2 units Taq DNA polymerase (Promega) in a reaction volume of 20 µl. Each reaction mixture was denatured at 95 °C for 5 minutes, followed by 30 cycles of PCR with primer annealing at 60 °C for 2 minutes, elongation at 72 °C for 3 minutes and denaturation at 94 °C for 1 minute. The final cycle consisted of a 10 minute 72 °C elongation followed by rapid cooling to 4 °C. PCR products were electrophoresed on 0.8% agarose gels.

Polymorphism: Allele A1 produces a PCR product of approximately 1950 bp, allele A2 produces a product of approximately 1800 bp, and allele A3 produces a product of approximately 1650 bp.

Frequency: In 24 unrelated individuals the frequency of allele 1 was 0.44, the frequency of allele 2 was 0.50, and the frequency of allele 3 was 0.06.

Chromosomal Localization: 17q21.3-q22.

Mendelian Inheritance: Co-dominant inheritance was observed in one three generation family with 8 members.

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Dral1 and Xmn1 polymorphisms at the human parathyroid hormone locus

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Source/Description: The plasmid p20.36 which contains an EcoRI fragment encompassing the human parathyroid hormone gene was utilized as a probe (1).

Polymorphisms: Dral1 identifies a two allele polymorphism (allele C1: 15.0 kb, allele C2: 16.5 kb). Xmn1 (or Asp700I) identifies a two allele polymorphism (allele D1: 4.5 kb, allele D2: 6.0 kb).

Frequency: Estimated from 120 chromosomes of unrelated individuals for Dral1:
C1: 0.86
C2: 0.14
Estimated from 88 chromosomes of unrelated individuals for Xmn1:
D1: 0.60
D2: 0.40

Also Polymorphic For: PstI and TaqI (1).

Chromosomal Localization: 11p15 (2, 3).

Mendelian Inheritance: Analysis of a large CEPH family (8 children) yielded results indicative of co-dominant inheritance.

Probe Availability: American Type Culture Collection.


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