**Dinucleotide repeat polymorphism at the locus D15S222**

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Source/Description: Inverse PCR (1) revealed a (GT)₂₁ repeat in the end of the left arm of YAC clone 944B11 (CEPH libraries), EMBL accession X72860. The predicted size of the amplified sequence was 194 bp.

Primer Sequences:

T38.a: 5'-CCTCAGCGTCCrCTCTTG-3'
T38.b: 5'-CTGGTCACTGTCTGTCCTGT-3'

Polymorphism and Frequency: Estimated from 100 chromosomes of unrelated individuals in the CEPH reference family panel. The observed heterozygote frequency is 0.78.

<table>
<thead>
<tr>
<th>Allele</th>
<th>Size (bp)</th>
<th>Frequency</th>
<th>Allele</th>
<th>Size (bp)</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>A1</td>
<td>198</td>
<td>0.03</td>
<td>A5</td>
<td>190</td>
<td>0.14</td>
</tr>
<tr>
<td>A2</td>
<td>196</td>
<td>0.09</td>
<td>A6</td>
<td>188</td>
<td>0.06</td>
</tr>
<tr>
<td>A3</td>
<td>194</td>
<td>0.45</td>
<td>A7</td>
<td>186</td>
<td>0.02</td>
</tr>
<tr>
<td>A4</td>
<td>192</td>
<td>0.13</td>
<td>A8</td>
<td>184</td>
<td>0.08</td>
</tr>
</tbody>
</table>

Two other alleles were found among the population of Isle de la Reunion.

Reference Genotypes: CEPH individual 141801 is A1, A3; 141302 is A2, A4; 141814 is A5 and 10201 is A6, A8.

Chromosomal Localization: The cytogenetic localization of YAC clone 944B11 was inferred as 15q15.3 by reference with other neighbouring YAC clones mapped by FISH (Fougerousse et al. in preparation).

Mendelian Inheritance: Co-dominant segregation was demonstrated in 16 CEPH families.

PCR Conditions: The PCR reaction was performed using 200 ng genomic DNA, 10 pmoles of each primer and 0.75 units of Taq polymerase in a final volume of 50 μl. Samples were subjected to 30 cycles consisting of 40 seconds at 92°C, 30 seconds at 52°C and 30 seconds at 72°C.

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**A 14 bp deletion polymorphism in the HLA-G gene**

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Source/Description: By aligning HLA-G DNA sequences obtained from GenBank, we identified an insertion of 1 bp (T) and a deletion of 14 bp (ATTTGTTCATGCCT) at the same site in exon 8. Of the 9 HLA-G sequences available, 2 had the 1 bp insertion and another 5 had the 14 bp deletion. Oligonucleotide primers were designed for regions flanking this site which had as little similarity as possible to other HLA class I sequences and also flanked a HLA-G locus specific deletion of approximately 50 bp. When these primers were used to amplify genomic DNA from 73 unrelated individuals, products of only two sizes were observed on a denaturing sequencing gel. These products correspond to the presence (141 bp) or absence (155 bp) of a 14 bp deletion only.

Primer Sequences:

HLAG-1: GTAGTGTGAAACAGCTGCCC
HLAG-2: AAGGAATGCAGTTCAGCATGA

Frequency: Estimated from 146 chromosomes of 73 unrelated individuals from a Caucasoid population. Observed heterozygosity = 0.41.

<table>
<thead>
<tr>
<th>Allele</th>
<th>Length (nt)</th>
<th>Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td>Del +</td>
<td>141</td>
<td>0.58</td>
</tr>
<tr>
<td>Del −</td>
<td>155</td>
<td>0.42</td>
</tr>
</tbody>
</table>

Chromosomal Localization: The HLA-G gene has been mapped to the human MHC region of 6p21.3 (1).

Mendelian Inheritance: Co-dominant segregation was observed in 9 pedigrees of 2-3 generations.

PCR Conditions: PCR reactions were carried out in a 20 μl volume containing 50 ng of genomic DNA, 125 μM each dCTP, dGTP and dTTP, 5 μM dATP, 1 μCi [α-32P]-dATP at 3000 Ci/mmol, 25 pmoles of each primer, 2.5 mM MgCl₂, 50 mM KCl, 10 mM Tris-HCl (pH 9.0), 0.01 % Triton X-100 and 1 unit of Taq polymerase. After an initial denaturation step of 94°C for 3 min, 30 cycles of 94°C for 1 min, 62°C for 1.5 min and 72°C for 2 min were carried out followed by a final extension at 72°C for 10 min. The reaction products were electrophoresed on a 5% denaturing sequencing gel and visualised by autoradiography.

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