Evidence for linkage disequilibrium between \textit{D16S94} and the adult onset polycystic kidney disease (PKD1) gene

S E Pound, A D Carothers, P M Pignatelli, A M Macnicol, M L Watson, A F Wright

Genetic mapping of the PKD1 gene in chromosomal region 16p13.3 has depended on the identification of closely linked markers and rare recombinants that suggest the order of loci in relation to meiotic crossover points.\textsuperscript{12} This has led to the proposal that the PKD1 gene lies in an approximately 750 kb region close to marker loci \textit{D16S84}, \textit{D16S125}, and \textit{D16S94}, and proximal to the hypervariable probe \textit{D16S85} that first detected linkage to \textit{PKD1}.\textsuperscript{3,4} However, genetic (meiotic) mapping can sometimes lead to inconsistent localisations, as with the Huntington's disease gene,\textsuperscript{3,5} owing to factors such as rare double crossovers, gene conversion events, gonadal mosaicism, genetic heterogeneity, misdiagnosis, mistyping, and non-paternity. While many of these factors can be minimised or excluded, some are very difficult to exclude and, although rare, become significant compared with the frequency of rare recombination events separating markers from disease genes. In the absence of patients with chromosomal rearrangements that can help to localise the PKD1 gene physically, genetic methods of mapping are critical.

A means of genetically mapping a disease gene, other than by linkage analysis, is by the identification of non-random allelic association (linkage disequilibrium) between a marker and disease locus. This can occur if a mutation causing disease arises in close association with a relatively uncommon marker allele or haplotype, which can then become over-represented in the patient group relative to the general population. The finding of linkage disequilibrium has served to refine the mapping both of the cystic fibrosis gene\textsuperscript{6} and of the Huntington's disease gene.\textsuperscript{5,7} However, linkage disequilibrium is expected to occur only if the mutation rate is relatively low or if the frequency of a particular chromosomal haplotype is inflated in the population of affected subjects as a result of previous selective advantage, migration, or population stratification. It requires that many or most affected subjects carry the same original mutation.

It has been suggested that the mutation rate in ADPKD is high,\textsuperscript{8} based on the apparently high frequency of unaffected parents of ADPKD subjects and on the high population prevalence. This study was carried out at a time when methods of presymptomatic diagnosis were less sensitive than with ultrasonography and when paternity testing was less readily available. We therefore tested a genetically homogeneous set of 33 ADPKD families for evidence of linkage disequilibrium between the PKD1 locus and closely linked markers \textit{D16S84}, \textit{D16S125}, and \textit{D16S94}.

A set of 34 ADPKD families was ascertained through medical renal units in Edinburgh and Glasgow and the diagnoses established as described.\textsuperscript{6,11} Family members were typed for the markers \textit{D16S85} (\textit{3' HVR}), \textit{D16S84} (\textit{CMH65}), \textit{D16S125} (\textit{26-6PROX}), and \textit{D16S94} (\textit{VK5}). Multipoint heterogeneity analysis was carried out to determine the proportion of families unlinked to the PKD1 region of chromosome 16.\textsuperscript{10} The sample was found to show evidence of linkage heterogeneity but 90 to 95% of the kindreds were linked to \textit{PKD1}. A single unequivocally unlinked family was removed from the analysis, leaving 33 families for genetic mapping of the PKD1 locus.\textsuperscript{11} In 28 families the haplotypes could be determined unambiguously, in one family the haplotype could not be established, and in the remaining four this was done by minimising the number of crossovers. The normal chromosomes used to establish control allele frequencies were derived from the unaffected spouses of family members.

The results are shown in the table. No evidence of allelic association was found between \textit{PKD1} and either \textit{D16S84} or \textit{D16S125}, both of which show close genetic linkage to \textit{PKD1}.\textsuperscript{12,11} In the case of \textit{D16S94}, there is evidence of linkage disequilibrium, with the affected haplotypes in seven kindreds showing the 1-6 kb allele (allele 1) and in 22 kindreds the 1-3 kb allele (allele 2) compared with the control population, who showed 38 1-6 kb alleles and 34 1-3 kb alleles. This result is significant at the 2% level (Yates's corrected \(x^2 = 3.75\)). The allele frequencies in the control population are in good agreement with those published previously.\textsuperscript{7}

The suggestion of linkage disequilibrium between \textit{PKD1} and \textit{D16S94} has implications for the genetic localisation of \textit{PKD1}. Any initial disequilibrium occurring when a mutation arises is dissipated at the rate of \((1-r)^n\) in \(n\) generations, where \(r\) is the recombination
### Frequency of marker alleles in haplotypes of ADPKD (PKD1) chromosomes compared with the general population. Yates's association coefficient provides a measure of the degree of disequilibrium. $\chi^2$ values were derived using Yates's correction.

<table>
<thead>
<tr>
<th>Allele</th>
<th>Affected</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>D16S84</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
<td>13</td>
<td>28</td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>38</td>
</tr>
<tr>
<td>$\chi^2$</td>
<td>0.020</td>
<td></td>
</tr>
<tr>
<td>$\Delta$</td>
<td>0.081 ± 0.225</td>
<td>0.387 ± 0.223</td>
</tr>
<tr>
<td>$p$</td>
<td>NS</td>
<td>NS</td>
</tr>
</tbody>
</table>

A high mutation rate but since the reproductive fitness of ADPKD patients is also high, this is not necessarily so. Additional factors, such as reproductive compensation, previous selective advantage, and genetic drift, may also contribute to the high prevalence. Linkage disequilibrium is not expected to occur if the mutation rate is high since each new mutation is likely to occur on a different chromosomal background. The results described in these ADPKD families therefore add support to the idea that the high prevalence is maintained by a high reproductive fitness or other factors referred to above, in the presence of an intermediate or low mutation rate. These issues await further clarification when the gene is isolated.

The financial support of The Wellcome Trust is gratefully acknowledged. We would also like to thank Drs J D Briggs and B J Junor for access to their patients. SEP is in receipt of an MRC Training Fellowship.

Evidence for linkage disequilibrium between D16S94 and the adult onset polycystic kidney disease (PKD1) gene.

S E Pound, A D Carothers, P M Pignatelli, A M Macnicol, M L Watson and A F Wright

doi: 10.1136/jmg.29.4.247

Updated information and services can be found at:
http://jmg.bmj.com/content/29/4/247.citation

These include:
Receiving free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

Notes

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/