α₁-Antitrypsin-levels and phenotypes in Crohn’s disease in the Netherlands

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SUMMARY A group of 310 unrelated patients suffering from Crohn’s disease has been screened for quantitative and electrophoretic variations of α₁-antitrypsin (α₁AT). A comparison was made between patients and healthy controls. The distribution of electrophoretic α₁AT variants in the patients showed no significant deviation from the controls. The α₁AT quantities are significantly higher in the Crohn’s disease population than in the controls.

Crohn’s disease, a chronic inflammatory disease of the intestine, is an uncommon condition mainly affecting young adults.

Ever since it was first described, attempts have been made to determine precise clinical and pathological criteria for the diagnosis and to look for factors which could influence or be the direct cause of the disease. Studies on the role of infectious and immunological factors in Crohn’s disease have not yet provided clear answers concerning the cause of the disease. Although genetic factors have been implicated in its pathogenesis, neither a genetic marker nor a biochemical parameter correlated with Crohn’s disease has so far been identified.  

α₁-Antitrypsin (α₁AT, locus Pi) is the main member of a species of inhibitors of proteolytic enzymes occurring in human serum. Genetic polymorphism was discovered by Fagerhol and Braend. The alleles Pi¹, Pi₂, Pi₃, Pi₄w, and Pi₅null are associated with decreased levels of α₁AT.

A deficiency of α₁AT was found to be associated with a variety of clinical conditions such as chronic obstructive lung disease (COLD), chronic cirrhosis of the liver, coeliac disease, arthritis, uveitis, and ankylosing spondylitis. These last three clinical conditions have been found to be associated with Crohn’s disease. Linkage is shown between α₁AT and immunoglobulin G(Gm), whereas other markers revealed no linkage.

We have screened a population of thoroughly investigated Crohn’s disease patients attending the in- and outpatient clinics of the department of gastroenterology at the University Hospital, Leiden, for both quantitative and electrophoretic variations of α₁AT in order to explore whether there is (1) a deviation of the distribution of electrophoretic variants of α₁AT in Crohn’s disease patients compared with controls and (2) any quantitative change in the α₁AT level in the Crohn’s disease patients.

Methods

Three hundred and ten unrelated patients suffering from Crohn’s disease were studied. In about 90% of all the cases the diagnosis was established on the basis of histological evidence; 258 patients have had at least one operation and the specimen resected met the histological criteria of Crohn’s disease; in 22 of 34 patients with Crohn’s disease in the colon determined through the endoscope biopsies showed epithelioid-cell granulomas. In the remaining 18 patients the diagnosis was based upon clinical and radiological findings.

The electrophoretic variations of α₁AT in Crohn’s disease patients were compared with those found in 708 healthy Dutch blood donors.

α₁AT was typed using two previously described methods: agarose-acrylamide electrophoresis for the initial typing and isoelectric focusing for verifying the variants and for subtyping the M-variant (a combination of the methods of Klasen et al. and Frants and Eriksson).

α₁AT was quantified using the standard radial immunodiffusion technique. The average values of α₁AT were expressed as percentage of the standard
value. The standard serum was a mixture of several sera from healthy blood donors with the phenotype M. The \( \alpha_1 \)AT quantity of our standard serum was compared by Dr M K Fagerhol with his serum pool and found to be the same. For comparing the \( \alpha_1 \)AT levels in the Crohn’s disease patients the quantitative data of the healthy controls of Fagerhol were used.

Results and discussion

**Typing**

Seven phenotypes with six different alleles were found in the Crohn’s disease population and, they were shown to be in Hardy-Weinberg equilibrium (Table 1).

The Crohn’s disease and blood donor populations showed similar frequencies of electrophoretic variants of \( \alpha_1 \)AT; none of the seven phenotypes showed a significant difference compared with the controls (Table 2).

**Quantification**

The level of \( \alpha_1 \)AT rose in the Crohn’s disease patients in all phenotypes, as can be seen in Table 3. In the phenotypes FM, M, and MS the difference reached significance.

<table>
<thead>
<tr>
<th>( \alpha_1 )AT allele</th>
<th>( \alpha_1 )AT gene frequency</th>
<th>( \alpha_1 )AT phenotype</th>
<th>Number</th>
<th>( \chi^2 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>F</td>
<td>0.00806</td>
<td>FM</td>
<td>5</td>
<td>0.0159</td>
</tr>
<tr>
<td>I</td>
<td>0.00323</td>
<td>IM</td>
<td>2</td>
<td>0.0064</td>
</tr>
<tr>
<td>M</td>
<td>0.94516</td>
<td>MZ</td>
<td>277</td>
<td>0.0000</td>
</tr>
<tr>
<td>S</td>
<td>0.03065</td>
<td>MS</td>
<td>17</td>
<td>0.0511</td>
</tr>
<tr>
<td>X</td>
<td>0.00161</td>
<td>MX</td>
<td>1</td>
<td>0.0032</td>
</tr>
<tr>
<td>Z</td>
<td>0.01129</td>
<td>MZ</td>
<td>7</td>
<td>0.0223</td>
</tr>
<tr>
<td>S</td>
<td>0.029</td>
<td>S</td>
<td>1</td>
<td>1.7260</td>
</tr>
<tr>
<td>Others</td>
<td>0.064</td>
<td>Others</td>
<td>1</td>
<td>0.6411</td>
</tr>
</tbody>
</table>

Table 1: Hardy-Weinberg analysis of electrophoretic variants of \( \alpha_1 \)AT in 310 unrelated patients suffering from Crohn’s disease.

For 1 degree of freedom: \( p = 0.12 \).

<table>
<thead>
<tr>
<th>Phenotype</th>
<th>Patients (N=310)</th>
<th>Controls (N=708)</th>
<th>( \chi^2 )</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td>FM</td>
<td>5</td>
<td>124</td>
<td>30.1</td>
<td>24</td>
</tr>
<tr>
<td>IM</td>
<td>2</td>
<td>111</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>M</td>
<td>277</td>
<td>127</td>
<td>33.1</td>
<td>203</td>
</tr>
<tr>
<td>MS</td>
<td>17</td>
<td>104</td>
<td>27.3</td>
<td>54</td>
</tr>
<tr>
<td>MX</td>
<td>1</td>
<td>183</td>
<td>8</td>
<td>8</td>
</tr>
<tr>
<td>MZ</td>
<td>7</td>
<td>81</td>
<td>36.5</td>
<td>12</td>
</tr>
<tr>
<td>S</td>
<td>1</td>
<td>81</td>
<td>8</td>
<td>8</td>
</tr>
</tbody>
</table>

Table 2: Distribution of electrophoretic variants of \( \alpha_1 \)AT among patients and controls.

Table 3 also shows that the phenotypes MS, S and MZ in the controls had a lower average \( \alpha_1 \)AT quantity than the other phenotypes, especially the M phenotype. This means that when the average quantity is compared between patients and controls the phenotypes should also be taken into account.

**Conclusion**

The search for linkage or associations of other genetic markers including HLA-DR with Crohn’s disease has not so far led to positive results. The present study reveals no association. Patients suffering from Crohn’s disease do not differ from the normal controls in their \( \alpha_1 \)AT phenotype distribution. As \( \alpha_1 \)AT is an acute phase reagent, the overall increase of the level of \( \alpha_1 \)AT in the patients is likely to be related to the activity of the disease.

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**References**

842 Klasen, Biemond, and Weterman


