Relative serum amyloid A (SAA) values: the influence of SAA1 genotypes and corticosteroid treatment in Japanese patients with rheumatoid arthritis

T Yamada, Y Okuda, K Takasugi, K Itoh, J Igari

Abstract

Objectives—(1) To determine whether serum concentration of serum amyloid A (SAA) protein is influenced by the SAA1 allele in Japanese patients with rheumatoid arthritis (RA) as previously shown in a healthy control group; and (2) to analyse what factors, based on such an allelic bias, influence the relative SAA values of those patients.

Methods—SAA and C reactive protein (CRP) concentrations together with SAA1 genotypes were determined in 316 Japanese patients with RA. The relative SAA values were evaluated as an SAA/CRP ratio.

Results—Comparison of the three SAA1 homozygote groups showed that the SAA/CRP ratio was highest in the 1.5/1.5 group (mean 9.0, p<0.01 v the other two homozygote groups) followed by the 1.3/1.3 group (mean 7.2, NS v the 1.1/1.1 group) and the 1.1/1.1 group (mean 4.0). The SAA/CRP ratio was significantly higher in patients receiving corticosteroids regardless of the presence of allele 1.5. No clear differences in the ratio between patients with or without amyloidosis were found.

Conclusion—The SAA1.5 allele and corticosteroid treatment had a positive influence on SAA concentrations in serum. These findings are important when evaluating SAA concentration in inflammatory diseases and when considering the cause or treatment of amyloidosis.

Relative serum amyloid A values

125

‡CS = corticosteroid; DM = DMARDs; IS = immunosuppressive agents.

*Class = Steinbrocker functional class.

agglutination turbidimetric immunoassay,25

trations in serum were determined by a latex

subjects. Table 1 shows the frequency of use.

men, and doses of corticosteroids varied among

phamide, and tacrolimus (FK-506). DMARDs

phenylacetic acid). Immunosuppressive agents

salazosulfapyridine, and actarit (4-acetylamino-

test.

allele

Table 2 SAA1 allele frequency and amyloid involvement

<table>
<thead>
<tr>
<th>SAA1 allele</th>
<th>Amyloidosis (No (%))</th>
<th>χ²</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Absent</td>
<td>Present</td>
</tr>
<tr>
<td>1.1</td>
<td>162 (0.331)</td>
<td>23 (0.187)</td>
</tr>
<tr>
<td>1.3</td>
<td>184 (0.376)</td>
<td>59 (0.447)</td>
</tr>
<tr>
<td>1.5</td>
<td>144 (0.293)</td>
<td>50 (0.379)</td>
</tr>
</tbody>
</table>

Methods

PATIENTS

Three hundred and sixteen Japanese subjects (38 male, 278 female, aged 16–89), both inpatients and outpatients at the Centre for Rheumatism, Dohgo Spa Hospital, Matsuyama, Japan during the months of June and July 1999, took part in this study. Informed consent was obtained from all subjects. All patients met the 1987 revised RA criteria of the American Rheumatism Association.24 All patients had undergone gastroduodenal endoscopic biopsy within the past two years; amyloidosis was diagnosed histologically from the biopsy specimens. Therapeutic drugs were grouped into three categories: corticosteroids, disease modifying antirheumatic drugs (DMARDs), and immunosuppressive agents. Corticosteroid was given as oral prednisolone. DMARDs included intramuscular gold, d-penicillamine, auranofin, bucillamine, salazosulfapyridine, and actarit (4-acetylamino-

protein (CRP) concentrations classified by SAA1 genotype (table 1). There were no apparent differences in age, Steinbrocker functional class, duration of disease, use of corticosteroid (%), DMARDs (%), or immunosuppressive agents (%). The SAA1 allele distributions in patients with amyloidosis were compared with those in subjects without (table 2). The allele 1.1 was negatively associated with amyloidosis (p<0.001) while 1.5 was positively associated (p<0.01). There was no statistical difference in the 1.3 frequency between the groups.

INFLUENCE OF SAA1 GENOTYPES ON SAA AND CRP VALUES AND THE SAA/CRP RATIO

To assess the influence of the SAA1 allele on SAA and CRP values and on the SAA/CRP ratio, data from three homozygote groups were compared (table 3, fig 1). CRP concentrations did not differ between the three groups, whereas SAA concentrations and SAA/CRP ratios were significantly higher in the 1.5/1.5 than in the other groups.

FACTORS AFFECTING THE SAA/CRP RATIO

Because the allelic bias on the SAA/CRP ratio was proved, subjects were divided into two groups according to the presence or absence of the SAA1.5 allele and their data were analysed. Under these conditions the allele 1.2 was regarded as equivalent to the 1.5 as this allele is rare and has exon 3 structures, which distinguish the common alleles, identical to

Table 3 Mean serum amyloid A (SAA) and C reactive protein (CRP) concentrations classified by SAA1 genotype

<table>
<thead>
<tr>
<th>SAA1 genotype</th>
<th>No</th>
<th>SAA (mg/l)</th>
<th>CRP (mg/l)</th>
<th>SAA/CRP</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.1/1.1</td>
<td>27</td>
<td>96</td>
<td>23</td>
<td>4.0</td>
</tr>
<tr>
<td>1.1/1.2</td>
<td>3</td>
<td>185</td>
<td>35</td>
<td>8.7</td>
</tr>
<tr>
<td>1.1/1.3</td>
<td>73</td>
<td>104</td>
<td>39</td>
<td>4.1</td>
</tr>
<tr>
<td>1.1/1.5</td>
<td>55</td>
<td>193</td>
<td>29</td>
<td>6.8</td>
</tr>
<tr>
<td>1.2/1.3</td>
<td>4</td>
<td>60</td>
<td>14</td>
<td>3.7</td>
</tr>
<tr>
<td>1.2/1.5</td>
<td>3</td>
<td>195</td>
<td>11</td>
<td>14.8</td>
</tr>
<tr>
<td>1.3/1.3</td>
<td>44</td>
<td>71</td>
<td>19</td>
<td>7.2</td>
</tr>
<tr>
<td>1.3/1.5</td>
<td>78</td>
<td>105</td>
<td>16</td>
<td>11.0</td>
</tr>
<tr>
<td>1.5/1.5</td>
<td>29</td>
<td>232*</td>
<td>25</td>
<td>9.0†</td>
</tr>
</tbody>
</table>

* p<0.001 v group 1.1/1.1, p=0.009 v group 1.3/1.3.
† p=0.001 v group 1.1/1.1, p=0.007 v group 1.3/1.3.
‡ p=0.005, § p=0.0001 v group 1.5 (−).

Results

CLINICAL FEATURES OF SUBJECTS BY SAA1 GENOTYPES

Data were assessed according to SAA1 genotype (table 1). There were no apparent differences in age, Steinbrocker functional class, duration of disease, use of corticosteroid (%), DMARDs (%), or immunosuppressive agents (%). The SAA1 allele distributions in patients with amyloidosis were compared with those in subjects without (table 2). The allele 1.1 was negatively associated with amyloidosis (p<0.001) while 1.5 was positively associated (p<0.01). There was no statistical difference in the 1.3 frequency between the groups.

FACTORS AFFECTING THE SAA/CRP RATIO

Because the allelic bias on the SAA/CRP ratio was proved, subjects were divided into two groups according to the presence or absence of the SAA1.5 allele and their data were analysed. Under these conditions the allele 1.2 was regarded as equivalent to the 1.5 as this allele is rare and has exon 3 structures, which distinguish the common alleles, identical to

Table 3 Mean serum amyloid A (SAA) and C reactive protein (CRP) concentrations classified by SAA1 genotype

<table>
<thead>
<tr>
<th>SAA1 genotype</th>
<th>No</th>
<th>SAA (mg/l)</th>
<th>CRP (mg/l)</th>
<th>SAA/CRP</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.1/1.1</td>
<td>27</td>
<td>96</td>
<td>23</td>
<td>4.0</td>
</tr>
<tr>
<td>1.1/1.2</td>
<td>3</td>
<td>185</td>
<td>35</td>
<td>8.7</td>
</tr>
<tr>
<td>1.1/1.3</td>
<td>73</td>
<td>104</td>
<td>39</td>
<td>4.1</td>
</tr>
<tr>
<td>1.1/1.5</td>
<td>55</td>
<td>193</td>
<td>29</td>
<td>6.8</td>
</tr>
<tr>
<td>1.2/1.3</td>
<td>4</td>
<td>60</td>
<td>14</td>
<td>3.7</td>
</tr>
<tr>
<td>1.2/1.5</td>
<td>3</td>
<td>195</td>
<td>11</td>
<td>14.8</td>
</tr>
<tr>
<td>1.3/1.3</td>
<td>44</td>
<td>71</td>
<td>19</td>
<td>7.2</td>
</tr>
<tr>
<td>1.3/1.5</td>
<td>78</td>
<td>105</td>
<td>16</td>
<td>11.0</td>
</tr>
<tr>
<td>1.5/1.5</td>
<td>29</td>
<td>232*</td>
<td>25</td>
<td>9.0†</td>
</tr>
</tbody>
</table>

* p<0.02 v group 1.1/1.1, p=0.009 v group 1.3/1.3.
† p=0.001 v group 1.1/1.1, p=0.007 v group 1.3/1.3.
‡ p=0.005, § p=0.0001 v group 1.5 (−).

www.annrheumdis.com
the plot because of isolated data. Differences were shown by the use of DM or IS.

**Discussion**

Two previous reports, including one by us,

suggest that the SAA1.3 and SAA1.1 alleles are positive and negative risk factors for amyloidosis in Japanese patients with RA. One study showed an increased frequency, though not significant, of the SAA1.5 allele in subjects with amyloidosis. In the present study, similarly, a negative association of the allele 1.1 with amyloidosis was found. The increased frequency of 1.3, however, was not statistically significant. Instead, a significant association of 1.5 was shown. These controversial findings may be due to the subjects selected. Because this study did not aim at assessing susceptibility to amyloidosis, there may be some imbalance in the clinical profiles of the patients with or without amyloidosis. Nevertheless, the data show again the low frequency of 1.1. Perhaps, this may be a stronger genetic indication for amyloidogenesis in Japanese patients with RA than the high frequency of the allele 1.3.

In a previous report we described an allelic bias in plasma SAA concentrations of a healthy Japanese population sample. Our preliminary experiments with recombinant SAA1 isotypes suggested that the difference in plasma clearance among the allele-corresponding SAA phenotypes was responsible for this (unpublished findings). Whatever the mechanism, the possibility that such allelic bias is shown in subjects with abnormally raised SAA concentrations should be assessed. Because these concentrations change according to the inflammatory activity present when the blood sample is drawn, it was difficult to evaluate the relation between SAA values and SAA1 genotypes. Therefore, we introduced the SAA/CRP ratio. CRP correlated markedly with SAA in most of the inflammatory disorders. To date, no study has reported the participation of a genetic factor in producing a variation in serum CRP concentrations. By using this ratio, it was shown that SAA responds more sensitively than CRP to a kidney allograft rejection.

The clinical profiles of the patients with RA in this study showed no apparent differences among the six major genotype groups. In addition, no statistical difference in CRP values was noted among the groups, suggesting that disease activity was also equally distributed. To
examine the allelic influence on relative SAA values, the SAA/CRP ratios were compared in three homozygote groups: SAA1.1/1.1, SAA1.3/1.3, and SAA1.5/1.5. The results showed that SAA1.5 homozygotes had higher SAA/CRP ratios than the others. Consequently, the positive influence by allele 1.5 was confirmed not only in healthy (or minimally pathological) states but also in inflammatory conditions.

The above results indicate the need to evaluate SAA concentrations in the light of genetic information about subjects. A recent finding suggests that SAA is a better prognostic marker in early RA than CRP.26 Combining SAA1 genotypic information about subjects. A recent finding confirmed not only in healthy (or minimally SAA1.3/1.3, and SAA1.5/1.5. The results examine the allelic influence on relative SAA concentrations in the light of genetic conditions.

In conclusion, our study confirms a positive influence of the SAA1.5 allele and corticosteroid treatment on relative SAA concentrations. These findings are important when evaluating SAA concentrations in diseases and when considering treatment in amyloidosis. Whether the susceptibility to amyloidosis is due to the allele regulated SAA levels or other factors should be further investigated.

19 Booth DR, Booth SE, Gillmore JD, Hawkins PN, Penys MB. SAA1 alleles as risk factors in reactive systemic amyloidosis. Amyloid 1998;5:262-5.

www.amrheumdis.com