Observations suggest that lysosomal enzymes released from synovial cells or leucocytes may be important in the pathogenesis of the inflammatory process in the joints of patients with rheumatoid arthritis (Weissmann, 1966; Cochrane, 1968). A likely source of these lysosomal enzymes is a synovial fluid polymorphonuclear leucocyte (Hollander, McCarty, Astorga, and Castro-Murillo, 1965) which has been found to engulf rheumatoid factor, immunoglobulins, complement, and other materials (Barnhart, Riddle, and Bluhm, 1967; Malinin, Pekin, Zvaifler, 1967; Brandt, Cathcart, and Cohen, 1968; Vaughan, Barnett, Sobel, and Jacox, 1968). Various protease inhibitors are present in serum, including $\alpha_1$-macroglobulin, inter-alpha-trypsin inhibitor, $\alpha_2$-antitrypsin, $\alpha_1$-antitrypsin, and $\alpha_1$-antichymotrypsin (Schwick, Heimburger, and Haupt, 1968). However, the principal proteolytic enzyme inhibitor in human serum is $\alpha_1$-antitrypsin, a glycoprotein that inhibits trypsin (Kueppers, 1971), plasmin (Heimburger and Haupt, 1966), thrombin (Rimón, Shamash, and Shapiro, 1966), chymotrypsin (Kueppers, 1971), elastase (Janoff, 1972), and proteolytic enzymes from granulocytes (Kueppers and Bearn, 1966). In this study we investigated the role of $\alpha_1$-antitrypsin in regulating the joint inflammation of rheumatoid arthritis by measuring its concentration in sera and synovial fluid.

Material and methods

Serum was obtained from forty healthy blood donor controls and from 24 patients with classic and definite rheumatoid arthritis (diagnostic criteria of American Rheumatism Association) (Ropes, Bennett, Cobb, Jacox, and Jessar, 1959). Knee synovial fluid was obtained by needle aspiration from normal volunteers and from patients with degenerative arthritis, classic and definite rheumatoid arthritis, and other forms of joint disease. Both sera and synovial fluid were obtained on the same day from most of these patients.

The concentration of $\alpha_1$-antitrypsin in sera and synovial fluid was determined by the quantitative radial immunodiffusion method of Mancini, Carbonara, and Heremans (1965) by using specific antisera to human $\alpha_1$-antitrypsin (Beringwerke). This antisera gave a major band in the $\alpha$ region when analysed by immunoelectrophoresis with normal human serum; two other faint bands also were seen. Serum and synovial fluid $\alpha_1$-antitrypsin molecules gave a reaction of identity in Ouchterlony analysis (Ouchterlony, 1958) and had similar sedimentation coefficients on sucrose density gradient ultracentrifugation, so the same standards were used for the serum and synovial fluid measurements. Women taking hormones or oral contraceptives were excluded from this study. Total protein concentrations in synovial fluid were determined by the biuret method (Kabat and Mayer, 1961).

Results

Age and sex had little influence on serum $\alpha_1$-antitrypsin levels and were therefore not considered further in this study. $\alpha_1$-Antitrypsin levels were significantly elevated in both the serum and the synovial fluid of patients with classic and definite rheumatoid arthritis as compared with normal serum controls and normal and degenerative arthritis synovial fluids (Table). Comparison of $\alpha_1$-antitrypsin concentrations of ten patients (eight with rheumatoid arthritis) revealed a positive correlation ($r = 0.84$) between the level in synovial fluid and that in serum (Fig. 1). There was also a positive relationship ($r = 0.79$) between total protein level and $\alpha_1$-antitrypsin concentration in individual synovial fluid specimens from patients with various joint diseases (Fig. 2). In two patients with juvenile rheumatoid arthritis, the serum $\alpha_1$-antitrypsin concentrations were 525 mg./100 ml. and 495 mg./100 ml. respectively. $\alpha_1$-Antitrypsin concentrations (mg./100 ml.) in the synovial fluid of four patients with gout were 136, 141, 159, and 162 mg. and in three patients with systemic lupus erythematosus they were 218, 222, and 246 mg. These concentrations were all higher than the normal controls, which were less than 110 mg.
Table  Concentrations of $\alpha_1$-antitrypsin in serum and synovial fluid

<table>
<thead>
<tr>
<th>Source</th>
<th>No. of cases</th>
<th>$\alpha_1$-Antitrypsin (mg./100 ml)</th>
<th>Mean</th>
<th>SD</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal controls</td>
<td>40</td>
<td>191</td>
<td>44</td>
<td></td>
</tr>
<tr>
<td>Classic and definite RA</td>
<td>24</td>
<td>278</td>
<td>68</td>
<td></td>
</tr>
<tr>
<td>Synovial fluid</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Normal controls</td>
<td>5</td>
<td>78</td>
<td>17</td>
<td></td>
</tr>
<tr>
<td>Degenerative arthritis</td>
<td>15</td>
<td>84</td>
<td>26</td>
<td></td>
</tr>
<tr>
<td>Classic and definite RA</td>
<td>21</td>
<td>148</td>
<td>46</td>
<td></td>
</tr>
</tbody>
</table>

* Significantly different from controls (P < 0.001; t test).
† Significantly different from classic and definite RA (P < 0.0001; rank-sum test).
‡ Significantly different from controls (P < 0.002; rank-sum test).

Discussion

$\alpha_1$-Antitrypsin is a potent proteolytic enzyme inhibitor, and approximately 90 per cent. of the total inhibitory capacity of serum trypsin can be attributed to it (Kueppers, 1971). Laurell and Eriksson (1963) first described the association between chronic obstructive pulmonary disease beginning at an early age and serum $\alpha_1$-antitrypsin deficiency. A lack of $\alpha_1$-antitrypsin in patients with rheumatoid arthritis could allow inflammation to increase because of uninhibited lysosomal enzymes. Conversely, higher than normal concentrations of $\alpha_1$-antitrypsin could help to control tissue damage by effecting a response of these enzymes. Cleve and Behrend (1966) and Müller and Müller-von Voigt (1968) reported that $\alpha_1$-antitrypsin factor increased in serum from patients with rheumatoid arthritis. The absolute values differ from those which we have obtained because of methodological variations. They did not measure $\alpha_1$-antitrypsin in the synovial fluid.

Summary

To investigate the role of $\alpha_1$-antitrypsin in joint inflammation, we measured its concentrations in sera and synovial fluid from patients with rheumatoid arthritis, degenerative arthritis, and other joint diseases, and compared these to normal control levels. There was a significant elevation of $\alpha_1$-antitrypsin concentrations in both serum and synovial fluid from patients with rheumatoid arthritis. There was a positive relationship between the $\alpha_1$-antitrypsin concentration in serum and that in synovial fluid in individual patients and between the total protein of synovial fluid and the $\alpha_1$-antitrypsin concentration in individual specimens. These results indicate that the inflammation in rheumatoid arthritis brings about an increased concentration of $\alpha_1$-antitrypsin, presumably in response to the increased release of lysosomal enzymes.
References


Janoff, A. (1972) Amer. Rev. resp. Dis., 105, 121 (Inhibition of human granulocyte elastase by serum α1-antitrypsin)


Kueppers, F. (1971) Humangenetik, 11, 177 (Alpha1-antitrypsin: physiology, genetics and pathology)


Weissmann, G. (1966) Ibid., 9, 834 (Lysosomes and joint disease)
Alpha 1-antitrypsin in serum and synovial fluid in rheumatoid arthritis.

H A Swedlund, G G Hunder and G J Gleich

doi: 10.1136/ard.33.2.162