concentration was three times greater than that in the adult at 0.5 h and the difference between the two ages became greater as time passed, rising to tenfold at 8 h. The mean lung concentrations in adult and neonate were similar at 0.5, 1 and 2 h but at 4 and 8 h neonatal gentamicin levels were greater than those in the adult. Gentamicin was detectable in adult brain at 0.5- and 1 h only, values thereafter being insignificantly different from zero. In contrast the mean brain gentamicin concentration of the neonates rose progressively to a peak of 1.26 ng/mg at 4 h and had not fallen significantly at 8 h. In all tissues the extracellular space was significantly greater in the neonate but the degree of difference varied markedly, being greatest in the liver and least in the lung. The renal concentration of gentamicin in both adult and neonate was always in excess of the extracellular space.

At 4 and 8 h neonatal lung gentamicin levels were greater than the adult but at 8 h the adult lung concentration remained stable at 1.21-2.32 ng/mg. In liver 1 intracellular' gentamicin was first detected at 4 h when the mean concentration of 0.59 ng/mg, but at 8 h this had risen to 3.31 ng/mg. Cardiac 'intracellular' gentamicin was present at 8 h in a low but significant concentration of 0.59 ng/mg.

83. AN EXAMINATION OF THE PROTEINASE CONTENT OF HUMAN LYMPHOCYTES, MONOCYTES AND POLYMORPHONUCLEAR LEUCOCYTES

K. T. HUGHES, E. SANDERS, G. A. COLES and M. DAVIES
Kruf Institute of Renal Disease, Royal Infirmary, Cardiff

Azurophil granules of polymorphonuclear (PMN) leucocytes contain at least three neutral proteinases, two of which (elastase and cathepsin G) may play a role in the pathogenesis of glomerulonephritis (Davies et al., 1978, Clinical Science and Molecular Medicine, 54, 233; Sanders, Davies & Coles, 1978, ibid, in press). Recently Schreiner et al. (1978, Journal of Experimental Medicine, 147, 369) have reported that monocytes may also be involved in the disease process. Furthermore Atkins et al. (1976, Lancet, i, 830) found monocytes/macrophages in renal biopsies from patients with rapidly progressive glomerulonephritis. In order to determine whether monocytes mediate glomerular basement membrane (GBM) damage in a similar manner to PMN leucocytes we have examined the lysosomal proteinase(s) content of monocytes.

Leucocytes of human peripheral blood from normal volunteers were separated into lymphocytes, PMN leucocytes and monocytes, sonicated and examined for total neutral proteinase (NP) (azocasein), elastase (Z-Ala-2-0 Nap and \(^{14} \)H elastin), cathepsin G (BZ-DL-Phe-2-0 Nap), cathepsin B (BZ-DL-Arg-2-naphthylamide) and cathepsin D (haemoglobin at pH 3.2). The results are shown in Table 1.

Lymphocytes contain no NP or cathepsin B activity. PMN leucocytes contain large amounts of neutral proteinase(s) but no cathepsin B and therefore have the ability to degrade GBM only at physiological pH. Monocytes possess NP activity at a level considerably lower than PMN leucocytes. This activity contained both elastase and cathepsin G. In addition monocytes contained small amounts of cathepsin B, a thiol-dependent acid proteinase which also degrades GBM in vitro. Monocytes therefore contain proteinases which are capable of degrading GBM at acid and physiological pH. These findings suggest that the monocyte may also contribute to GBM damage in glomerulonephritis.

84. EVIDENCE FOR A FUNCTIONAL ADP RECEPTOR IN PLATELETS IN GLANZMANN’S THROMbasthena

L. C. BEST, M. B. MCGUERE, F. E. PRESTON and R. G. G. RUSELL

Departments of Chemical Pathology and Haematology, University of Sheffield Medical School, Sheffield

Glanzmann’s thrombasthena is a bleeding disorder in which platelets do not aggregate in response to ADP added in vitro. It has been postulated that a defect may exist in the ADP receptor. Although a deficiency of a specific glycoprotein has been demonstrated in isolated platelet membranes (Nurden & Caen, 1974, British Journal of Haematology, 28, 253), the binding of ADP to platelet membranes appears to be normal, so that the nature of the postulated receptor defect requires further clarification.

Since the addition of ADP to platelets is associated with a series of readily measurable biochemical changes, we decided to examine the biochemical responses to ADP in Glanzmann’s platelets.

Two patients with Glanzmann’s disease were studied. Their platelets did not aggregate in response to ADP, adrenaline, collagen or the 11,9-epoxymethano-analogue of prostaglandin H\(_2\). The platelets also exhibited an impaired release of \(^{14} \)C-labelled serotonin in response to ADP. However, the cyclic nucleotide responses were indistinguishable from normal. Thus a rise in platelet 3':5 '-cyclic AMP was observed in response to prostaglandin E\(_1\) (PGE\(_1\)), and, despite the lack of aggregatory responses to ADP and adrenaline, both these agents caused a fall in cyclic AMP levels in platelets previously exposed to prostaglandin E\(_1\). Moreover, the thrombin- and ADP-induced rises in malonyldialdehyde (MDA), a breakdown product of prostaglandin endoperoxides and thromboxane A\(_2\), were also present in Glanzmann’s platelets. These results suggest that the receptor mechanism for ADP is at least partially intact in Glanzmann’s platelets, since the cyclic nucleotide and MDA responses to ADP are still present. The defect seems more likely to reside in the later events in platelet aggregation that occur after generation of thromboxane A\(_2\).

**Table 1. Proteinase activities of neutrophils**

Enzyme activity is shown as units/10\(^6\) cells ± SD. n.d., not detected.

<table>
<thead>
<tr>
<th>Cell type</th>
<th>Total NP activity (1489 - 406.8 (9) 1-01 x 10(^6) dpm ± 0-57 x 10(^5) (5)</th>
<th>Elastase (Z-Ala-2-O Nap)</th>
<th>Elastase ((^{14} )H Elastin)</th>
<th>Cathepsin G</th>
<th>Acid proteinase (Hb, pH 3-2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PMN leucocyte</td>
<td>1223.9 ± 150.6 (9)</td>
<td>n.d.</td>
<td>n.d.</td>
<td>35.7 ± 31.3 (9)</td>
<td>n.d.</td>
</tr>
<tr>
<td>Lymphocytes</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>n.d.</td>
<td>84-6 ± 20-7 (4)</td>
</tr>
<tr>
<td>Monocytes</td>
<td>95-3 ± 43.2 (5)</td>
<td>77-7 ± 65-5 (5)</td>
<td>0-88 x 10(^4) dpm ± 0-17 x 10(^3) (3)</td>
<td>17-9 ± 1-9 (5)</td>
<td>13-6 ± 2-2 (3)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>3-8 ± 1-6 (3)</td>
</tr>
</tbody>
</table>