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HAEMATOLOGY

Two Populations of Platelets

It has been suggested that the tail frequently observed in a platelet survival study could be explained by a young population of platelets having a longer life span than the remainder[1]. The following experiments demonstrated the presence of two morphologically different populations, following the exposure of human platelets to osmotic stress.

Platelets were obtained from whole blood collected in 2 per cent ethylenediamine tetraacetic acid (EDTA) by differential centrifugation and then centrifuged at 2,000 r.p.m. for 10 min; after removal of the plasma the platelets were re-suspended in 0·85 per cent NaCl and re-centrifuged.

The platelets were then equally divided; one half was placed in distilled water and the other in 0·85 per cent NaCl to act as a control. At various intervals ranging from 10 sec to 1 h, samples were taken from both and, in the case of the distilled water treatment, were added to an equal volume of 1·8 per cent NaCl to make the samples isotonic. The platelets were then centrifuged and fixed as a pellet in 1 per cent osmium tetroxide in Palade's buffer for 1 h. After dehydration through graded alcohols they were embedded in 'Araldite' and sections were cut on an L.K.B. 'Ultratome'. A Philips *E.M.* 200 electron microscope was used to examine the sections.

Fig. 1 shows the changes present in the platelets after 10 min in distilled water. It can be seen that they behaved in one of two ways: either they appeared pale, round and slightly expanded, frequently showing breaks in the cell membrane with an apparent loss of cytoplasmic organelles; or they became darker and slightly contracted, having a continuous cell membrane with many pseudopodia and the usual cytoplasmic organelles. The paler group of platelets became evident after 10 sec in water and the darker group after 60 sec. By 10 min no 'normal' platelets remained and the two distinct groups were clearly evident. No further morphological change could be noted at 30 and 60 min. Similarly, two types of platelets have been observed in a preliminary study using hypertonic saline.

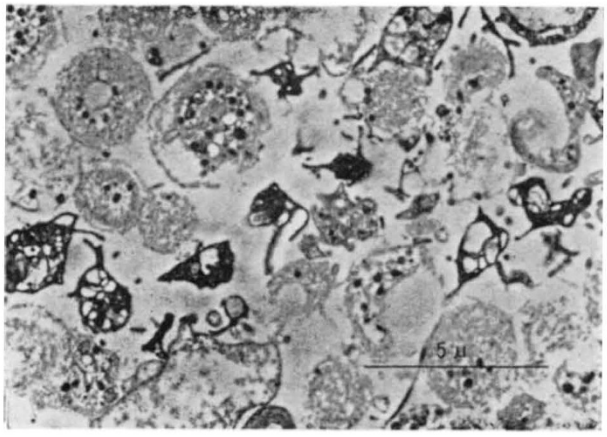

Fig. 1. Platelets after 10 min in distilled water

Serial sectioning was performed to prove that two distinct platelet populations were present. It seems likely that these changes may correspond to the 'spread' and 'sword' forms seen in whole platelets subjected to hypotonic solutions in studies of osmotic fragility[2,3].

The difference in response to hypotonicity that we have observed probably reflects biochemical differences in the platelet population. This may be due to (i) platelet age, (ii) two biochemically distinct but normally morphologically similar groups of platelets, or (iii) artefact induced in handling. The possibility of an artefact, however, is unlikely, as one patient with severe thrombocytopenia showed only one type of platelet, the 'paler' type, when tested in this way. Further studies are being undertaken in an attempt to determine whether or not these changes are due to age. The possibility that there are two platelet populations has obvious importance in studies of platelet physiology and pathology.

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Efficiency of Searching Gm Antigens for Identification of Blood-stains in Forensic Medicine

The recent communications of Brocteur and Moureau[1], and Nielsen and Henningsen[2], as well as the interest found in seric groups, lead us to report here a forensic investigation illustrating the importance of the Gm antigens for identification of blood-stains.

In September 1961, a suitcase containing a sum of money was stolen, and it was recovered on waste ground; its lid had been cut, probably with a sharp knife. A thorough examination revealed two very small brown-red stains, looking like blood, on the lid. A suspect arrested soon afterwards had a wound on his thigh, and his trousers were torn at the very spot where the knife must have wounded him.

The police reconstructed the facts as follows: the man had stolen the suitcase, had put it on his lap and, trying to cut it open with his knife, had implanted the blade in his thigh. To verify this hypothesis, we had to prove that the brown stains on the suitcase could have come from the man's blood and that they were similar to the blood-stains found on the trousers.

The difficulty in this investigation was that the stains were very small, making it impossible to determine the antigens of the ABO system or, of course, those of the P or Rh systems. So, after having shown the presence of peroxidase and human proteins in these stains, we tried to identify them simply by searching Gm (a) and Gm (x) antigens.

In fact, the subject was A_1 Rh + Gm (a + x −). Because of the lack of specificity of some anti-Gm (b) sera, we had refrained from using this system in a forensic investigation.