PLATELET MORPHOLOGY IN PATIENTS WITH ENDOGENOUS DEPRESSIONS DURING ANTIDEPRESSANT THERAPY

A. I. Kiktenko, K. Trunte, G. P. Zlobina, and O. S. Brusov

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The study of peripheral blood platelets in patients with endogenous depressions is urgent on two grounds. First, platelets are a convenient peripheral model of serotonin synapses, for most of the elements responsible for serotonin turnover, identical with those on central serotoninergic neurons, are present on them [4, 8]. Meanwhile the pathogenesis of endogenous depressions is linked with a disturbance of the central serotoninergic system [2, 7]. Second, chronic antidepressant therapy of some patients leads to the appearance of side effects associated with their cytotoxic action on myocardial and blood cells [9]. The action of antidepressants, taken by a patient, on different types of cells has received little study. The importance of this action to the therapeutic effect, and also to the development of side effects during long-term treatment is not clear. However, we know that platelets are very sensitive to various physical and chemical influences [5]. Because they have a contractile system they can actively change the shape of their surface by forming pseudopodia, and they can thus respond precisely to changes in the environment. There is therefore reason to suppose that under the influence of antidepressants changes will be observed not only in the functional properties of platelets, but also in their morphologic structure.

It was therefore decided to study changes in platelet morphology of patients with endogenous depressions receiving antidepressant treatment, by scanning electron microscopy, enabling a three-dimensional image of the object to be obtained and giving an objective impression of relations between particular formations of biological objects.

EXPERIMENTAL METHOD

Altogether 18 women patients aged from 17 to 57 years were studied. Only patients with endogenous depression in the category of affective psychosis of monopolar and bipolar type [11] took part in the investigation. Three blood samples were taken: on admission to hospital, 1-3 days after the beginning of treatment, and in the stage of initial clinical improvement (after 37-42 days). The patients were not given any psychotropic drugs, except small doses of benzodiazepines, for at least 10 days before the first blood sample was taken. The patients then started on a course of treatment with one of the following antidepressants: melipramiae, amitriptyline, maprotiline (Ludiomil), and yirlindol (Pirazidol). Platelet enriched plasma (PEP) was obtained by centrifugation of blood at 280*g* for 15 min at room temperature. A platelet suspension free from plasma proteins was obtained by gel-filtration of PEP on sepharose CL-2B on a column (2.5 × 15 cm), equilibrated beforehand in buffer: 50 mM Tris-HCl, 120 mM NaCl, 5 mM KCl, 7 mM citric acid, 8 mM Na citrate, 1.1 mM glucose, pH 7.5 [3] (buffer A). To investigate the surface structure of the platelets by scanning electron microscopy platelet suspension was fixed in 2.5% glutaraldehyde solution in buffer A for 30 min at room temperature. The platelets were then sedimented by centrifugation at 300*g* for 10 min on aluminum foil and dehydrated by ethanol solutions of increasing concentrations: 30, 50, 70, 90,

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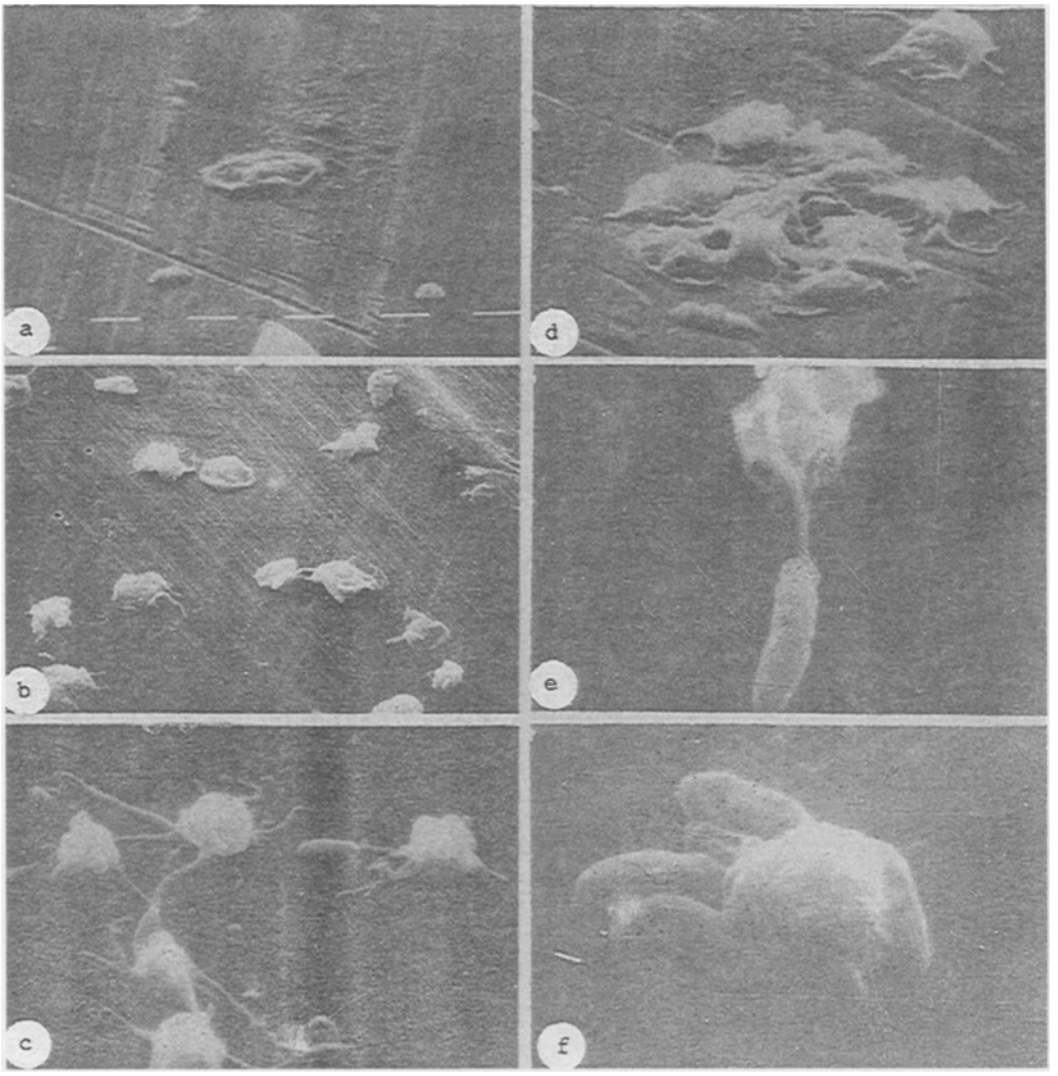

Fig. 1. Surface structure of peripheral blood platelets of patients with endogenous depressions, obtained by scanning electron microscopy: a) disk-shaped platelets, 10,000×; b) platelets with short pseudopodia, 5000×; c) platelets with long pseudopodia, 10,000×; d) appearance of platelets in a state of aggregation, 10,000×; e, f) rod-shaped platelets, 20,000×.

100%, for 3 min at room temperature. After dehydration the platelets, together with the aluminum foil, were secured to the microscope stages by means of special current-conducting glue (Polaron, Great Britain), dried in air, and covered with gold. The investigation was carried out on a Philips 501 SEM scanning electron microscope (The Netherlands).

EXPERIMENTAL RESULTS

The morphologic study of the peripheral blood platelets of patients with endogenous depressions at different stages in the course of the disease revealed the following types of cells: type 1: cells shaped approximately like a regular disk with no outgrowth. The diameter of these cells is about 3 μ (Fig. 1a). Type 2: cells of this type are irregular in shape and have a folded surface. Another feature of platelets of this type is that they have short outgrowths, not larger than the cell body (Fig. 1b). Type 3: these cells have more marked changes. The platelets may have long outgrowths, which are wider at the cell body end and taper toward their distal end. These outgrowths are

2-3 times longer than the diameter of the cells, and exceed five in number. An important feature of platelets of this type is their tendency to form large aggregates. Formations of this kind consist of an aggregate of many platelets. These cells are generally shapeless (Figs. 1c, d). Type 4: cells of this type are characteristically rod-shaped. The direct transition of the body of a platelet into one such rod is illustrated in Fig. 1e; the rods themselves are relatively small (about 3 μ long) and regular in shape. In some cases they lay separately, in others they were arranged around a cell (Fig. 1f). The distribution of these types of platelets during treatment was as follows. At the 1st point of investigation, on admission of the patients to the clinic, in six of nine cases the cells belonged to type 2. They showed few signs of activity. Three patients exhibited strong activation. Only cells of type 3 were observed. No rod-shaped forms were found. After 3 days of treatment (the 2nd point) the pattern of distribution of the various types of platelets was altered. Cells of type 1 were found in nine of 11 patients, but only in two cases were type 3 cells (activated form) found. By contrast with the 1st point of the investigation, rod-shaped forms appeared in three patients. In the stage of initial clinical improvement of the patients' condition, the morphologic picture of platelet structure revealed certain distinguishing features: in six of seven cases the cells were highly active (type 3). A low level of activation was found in one patient (type 2). Rod-shaped forms were found in all patients.

With the above findings in mind, the morphologic structure of the patients platelets before the beginning of treatment, when the depressive state was well marked and no influence of psychotropic drugs was present, is undoubtedly interesting. The picture was clearly heterogeneous. Activation was present in all cases, mainly mild. A change in the morphologic structure of the platelets took place 3 days after the beginning of treatment: in nine of 11 cases (over 80%) inactivated platelets were observed, and cells of type 3 were found in only two cases. Such changes during the 1st days of treatment can be explained by the stabilizing effect of the antidepressants on the platelet membrane when present in a low concentration on the cell surface [10]. During long-term therapy (37-42 days after the beginning of treatment) significant changes were observed. All the cells were activated, mostly strongly (cells of type 3). Besides, a new rod-shaped type of cell could be seen to have appeared in all patients. Incidentally, this morphologic structure differed more from the state of the patient's platelets before the beginning of treatment (at the 1st point) than at the 2nd point of investigation, namely 3-4 days after starting antidepressants. Antidepressant therapy led to the formation of a hitherto unknown rod-shaped type of platelet. It can be tentatively suggested that the appearance of these cells was due to the ability of the antidepressants to be adsorbed on the cell surface. Imipramine is known to have this property [6]. Presumably during long-term treatment antidepressants accumulate on the membrane, leading to a selective or general increase in its permeability and to manifestation of cytotoxicity, and so to the appearance of "spent" platelets together with these "rod-shaped" structures [12].

The investigation thus revealed that platelets of patients with endogenous depressions, not receiving antidepressants, are activated, and that the pattern of activation is heterogeneous. During long-term treatment more marked activation was observed, with the appearance of a new type of rod-shaped forms of platelets.

Changes in the morphologic structure of the platelets under the influence of antidepressants are associated with disturbance of the parameters of the serotonin system of these cells. The writers showed previously that mechanical activation of platelets causes an increase in the rate of serotonin reuptake and in the number of detectable imipramine receptors [1]. Unfortunately we do not yet know the role of the changes discovered in the morphologic structure of the platelets and disturbances of the functional activity of these cells connected with them in the mechanism of action of antidepressants. Most probably these changes make their own contribution to the development of side effects that are observed during antidepressant therapy and, in particular, to the realization of the cardiotoxic effects of these drugs [9].

REFERENCES

1. A. I. Kiktenko, G. P. Zlobina, M. R. Shurin, et al., Byull. Éksp. Biol. Med., **112**, 485 (1981).
2. H. Agren, Psychiat. Res., **3**, 211 (1980).
3. M. Asberg, B. Eriksson, B. Martensson, et al., J. Clin. Psychiat., **47**, suppl., 23 (1986).
4. M. Baron, A. J. Barkai, R. Gruen, et al., Biol. Psychiat., **18**, 1403 (1983).
5. D. Bottechia, G. Fantin, G. Nassuato, et al., Platelets: A Multidisciplinary Approach, New York (1977), p. 111.

6. D. Boulin and R. O'Brien, J. Pharm. Pharmacol., **20**, 583 (1968).
7. N. Briley, Neuropharmacology of Serotonin, Oxford (1985).
8. A. Coppen, C. Swade, and R. Wood, Clin. Chim. Acta, **87**, 166 (1978).
9. A. H. Glassman, B. T. Walsh, S. P. Roose, et al., J. Clin. Psychiat., **45**, 35 (1982).
10. J. D. Goode and V. H. Meltzer, Biochem. Pharmacol., **23**, 2629 (1974),
11. International Classification of Disease. 8th Revision. WHO Publication No. 8921, Geneva (1974); New York (1975).
12. I. Ishigoka, V. Shizu, U. Watanabe, et al., Biol. Psychiat., **20**, 866 (1985).