Ultrastructural Changes in Erythrocytes in Patients with Mental Disorders

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Transmission electron microscopy of peripheral blood erythrocytes from patients with paranoid and residual schizophrenia, mental retardation, and neurotic disturbances revealed nonspecific ultrastructural changes in the membrane and matrix of red blood cells.

Key Words: *mental disorders; erythrocyte; transmission electron microscopy*

Systemic disorders accompanying mental diseases attract much attention. Recent studies revealed multilevel disturbances in intra- and intercellular interactions [9,10,12,13]. Molecular mechanisms of these disturbances and their role in the pathogenesis of mental disorders are poorly understood. The pathogenesis of these diseases should be studied at various levels. Here we evaluated general and specific ultrastructural changes in peripheral blood erythrocytes from patients with various mental disorders.

MATERIALS AND METHODS

We performed an electron microscopy study of peripheral blood erythrocytes from 53 patients with mental disorders (17-52 years): paranoid (F20.0, ICD-10) and residual schizophrenia (F20.5, n=40), mental retardation (F71, n=7), and neurotic disturbances (F4, n=6). These patients were in a satisfactory somatic state and received no psychotropic drugs for more than 6 months. The control group included 12 healthy donors.

Ultrastructure of erythrocytes was studied by transmission electron microscopy [2]. To prepare ultrathin sections peripheral blood samples were fixed with 2.5% glutaraldehyde and 1% OsO_4 in cacodylate buffer (pH 7.4), dehydrated in alcohols of increasing concentrations and acetone, and embedded into the Epon-Araldite mixture. After targeted ultratomy, 30-60-nm slices were contrasted with uranyl acetate and lead citrate. The samples were examined under a JEM-100 CXII electron microscope (aperture diaphragm 25-30 μ, accelerating potential 80 kV). We examined 1000 erythrocytes in each preparation. The incidence of morphological changes was estimated. The results were analyzed by Mann—Whitney test.

RESULTS

Erythrocyte from healthy donors had well-contoured membranes and regular fine-grained electron-dense matrix. In some erythrocytes membrane destruction was manifested in its peeling from the stroma, local thinning or thickening, and formation of microexovesicles. Erythrocytes poorly contrasted with OsO_4 and cells with irregularly distributed hemoglobin granules and endovesicles were only occasionally seen. This morphological heterogeneity of erythrocytes from healthy donors probably reflects cell aging.

In patients with schizophrenia, the count of structurally abnormal erythrocytes increased. These cells were characterized by local and extensive defects of the cytolemma: loosening, thinning, thickening, breaks, fragmentation, peeling, and formation of spaces, exovesicles, endovesicles, and vacuoles containing small and large osmiophilic particles (Fig. 1). In patients

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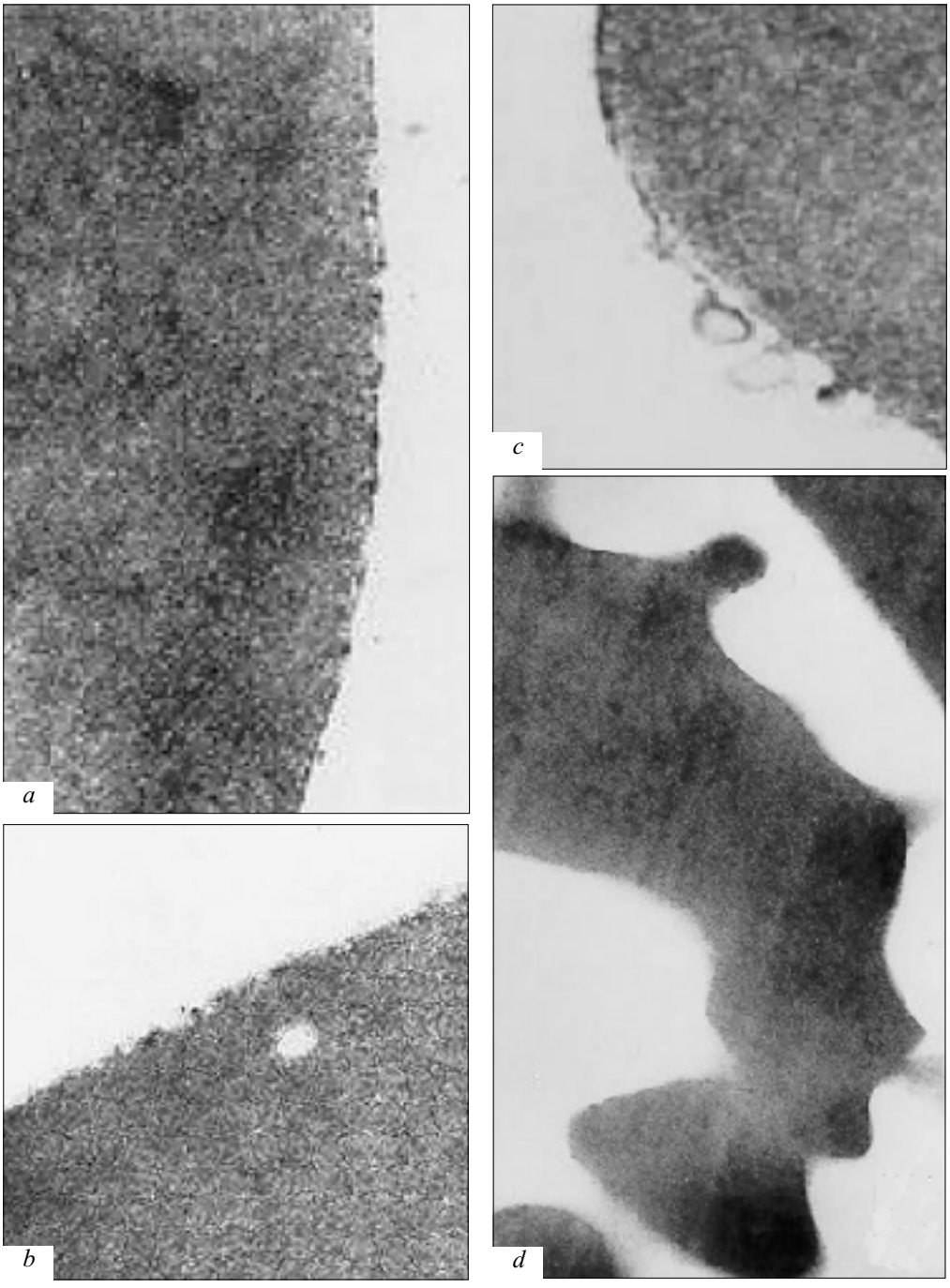

Fig. 1. Electron microphotographs of peripheral blood erythrocytes from a schizophrenic patient. Transmission electron microscopy (×58,000, *a-c*; and ×24,000, *d*). *a*) defects of erythrocyte membrane (breaks), *b*) endovesicle in the erythrocyte stroma, *c*) erythrocyte with exovesicles, cell with club-like processes (*d*).

with schizophrenia, the sum of ranks characterizing the count of cells with cytoplasmic endovesicles was 428 (67 in the control, $p<0.01$). The sum of ranks characterizing the count of cells with micro- and macrovesicles increased from 79 to 417 ($p<0.05$), which indicated pronounced exovesiculation of erythrocytes in patients with mental disorders.

These ultrastructural signs reflecting destruction of erythrocyte membranes and matrix are not specific for patients with schizophrenia. Similar ultrastructural changes were found in eritrocytes from patients with mental retardation resulting from disontogenesis of the brain caused by pre-, intra-, and postnatal pathogenic factors. In these patients we found not only erythrocytes with the homogenous cytoplasm, but also cells with irregularly distributed hemoglobin granules and focal destruction of stroma (endovesicles). In patients with mental retardation the sum of ranks characteri-

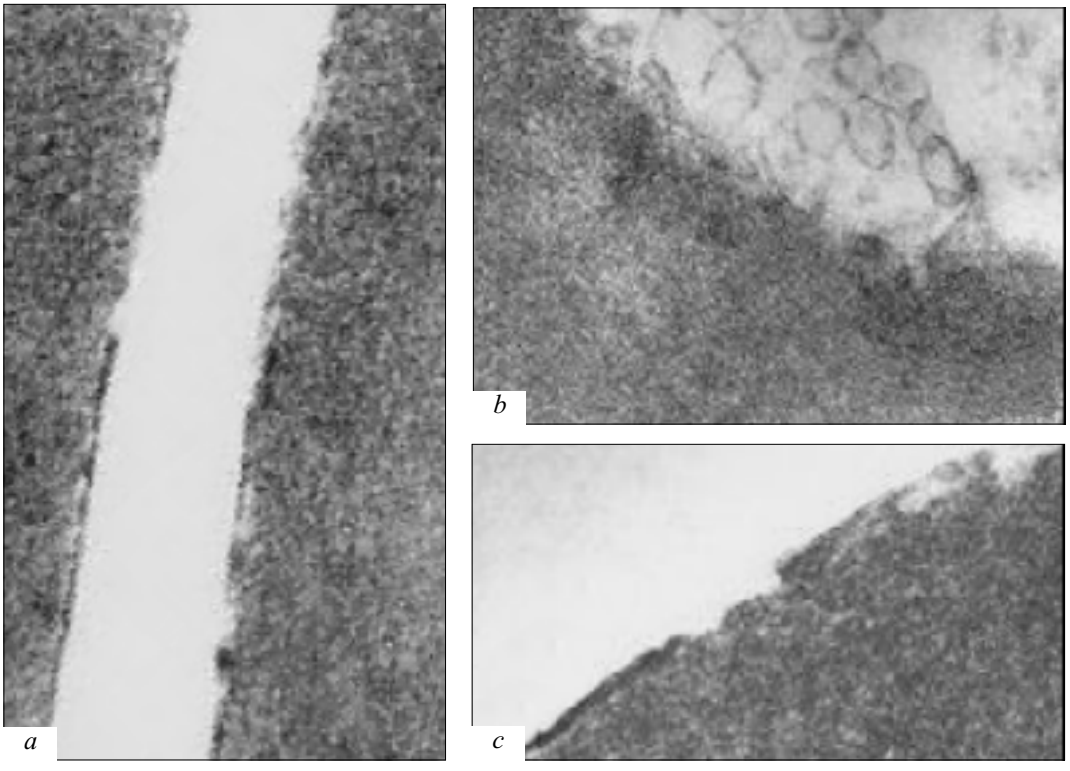

Fig. 2. Electron microphotographs of peripheral blood erythrocytes from a patient with neurasthenia. Transmission electron microscopy (×58,000). Local and extensive defects of erythrocyte membranes: thinning, breaks, and loosening (*a*). Numerous macro- and microexovesicles (*b*).

zing the count of erythrocytes with endovesicles was 81 *vs.* 39 in the control ($p<0.01$). In these patients the number of cells with micro- and macrovesicles markedly surpassed the control (74 and 46, respectively). However, exovesiculation of erythrocytes in patients with mental retardation was less pronounced than in patients with schizophrenia. The sum of ranks characterizing the count of cells with membrane and perimembrane exovesicles in patients with mental retardation and schizophrenia was 102 and 363, respectively ($p<0.05$).

In patients with neurotic disorders (neurasthenia and adaptive disturbances) we found ultrastructural signs of pathological changes in erythrocyte membranes: formation of local and extensive defects, thinning, breaks, fragmentation, and perforations. Cells with micro- and macroexovesicles (Fig. 2) were also revealed. The phenomenon of vesiculation is associated with erythrocytes aging [6,7]. Previous studies showed that erythrocytes respond to stress factors (including binding of immune complexes) by exocytosis, which prevents cell destruction [11]. The loss of membrane materials through exovesiculation is probably associated with abnormal lipid and protein composition of the cytolemma. This leads to changes in the surface tension determined by interactions in the outer and inner membrane monolayers [8], separation of the membrane cytoskeleton and plasmalemma during activation of phospholipases, and release of membrane vesicles [7].

Our experiments revealed nonspecific ultrastructural changes in erythrocyte membrane and matrix in patients with schizophrenia, mental retardation, and neurotic disturbances. The same ultrastructural changes in erythrocytes were found in patients with malignant neoplasms [4], myocardial infarction, acute viral hepatitis [3], thermal traumas [5], staphylococcal toxicosis, and postischemic syndrome [1], which attests to universal nature of ultrastructural morphological changes in peripheral elements of the erythron during etiologically and pathogenetically different diseases.

REFERENCES

1. N. B. Zakharova and G. P. Titova, *Pat. Fiziol*., No. 2, 50-52 (1992).
2. V. Ya. Karupu, *Electron Microscopy* [in Russian], Kiev (1984).
3. V. N. Kidalov and V. F. Lysak, *Lab. Delo*, No. 8, 36-40 (1989).
4. V. V. Novitskii, E. A. Stepanovaya, V. E. Gol'dberg, *et al*., *Erythrocytes and Malignant Neoplasms* [in Russian], Tomsk (2000).
5. N. V. Ryazantseva, V. V. Novitskii, and V. P. Ryazantsev, *Klin. Lab. Diagn*., No. 6, 23-34 (2000).
6. E. I. Slobozhanina, I. E. Fedorovich, E. A. Chernitskii, *et al*., *Biofizika*, **36**, No. 4, 621-623 (1991).
7. V. I. Sorokovoi, N. N. Mochenova, and G. M. Nikitina, *Byull. Eksp. Biol. Med.*, **117**, No. 5, 555-558 (1994).

8. V. I. Chernyshov, T. P. Smekhova, Yu. S. Tarakhovskii, and V. L. Borovyagin, *Biol. Membrany*, No. 5, 516-528 (1989).
9. A. Bertolino, A. Breier, J. H. Callicott, *et al*., *Neuropsychopharmacology*, **22**, No. 2, 125-132 (2000).
10. D. F. Horrobin, A. I. Glen, and K. Vaddadi, *Schizophr. Res*., **13**, No. 3, 195-207 (1994).
11. K. Iida, M. B. Whitlow, and V. Nussenzweig, *J. Immunol*., **147**, No. 8, 2638-2642 (1991).
12. R. Sandyk, *Int. J. Neurosci*., **68**, No. 1-2, 1-10 (1993).
13. C. A. Tamminga, *Crit. Rev. Neurobiol*., **12**, No. 1-2, 21-36 (1998).