EXPERIMENTAL METHODS FOR CLINICAL PRACTICE

Serotonin Insufficiency of Neutrophils

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The mobility of neutrophils is found to be depressed by serotonin antagonists (toxins and drugs) in patients with wound infection. Addition of serotonin eliminates serotonin insufficiency of neutrophils, which is expressed in acceleration of their mobility. Data are presented showing that serotonin is indispensable for maintaining neutrophil and platelet homeostasis.

Key Words: *serotonin; neutrophils; serotonin insufficiency of neutrophils*

Neutrophils (NP), representing 50 to 70% of the total number of adult human leukocytes, are partially responsible for nonspecific immunity. NP are known to possess an ameba-like mobility of their own which is lowered in various diseases; specifically, it is markedly depressed in patients with wound infection [3]. Normal mobility of NP is an integral parameter of their normal morphology and function. A decrease of NP mobility indicates disorders of biochemical processes and of the normal transformation of biochemical energy into mechanical energy.

We noted that with the appearance of serotonin (5-HT) antagonists in the organism serotonin relationships with 5-HT receptors are disrupted, this leading to dysfunction of the organs and systems in which these receptors are situated. Exogenous administration of 5-HT normalizes serotonin interactions with 5-HT receptors of smooth muscles and the central nervous system and simultaneously repairs their functions. In simulation experiments with depressed NP we proceeded from the hypothesis that if a 5-HT insufficiency of the whole organism is possible, organ and cellular 5-HT insufficiency is possible as well [10-12].

MATERIALS AND METHODS

Leukocytes were isolated from the blood routinely: 2-3 drops of capillary blood were put on a small slide and placed in an incubator in a humid chamber for 10-15-min incubation at 37°C, then washed from the clot and red cells by gentle rinsing in a glass with Hanks' solution, after which the small slide with the NP adhering to it was transferred onto a slide with a Vaseline ridge applied to it along the perimeter of the smaller slide. The space delimited by the Vaseline ridge was filled with Hanks' medium (pH 7.3) with 1% human serum albumin (Fluka). In this manner, the smaller slide with NP stuck to it was placed in a small ridge-confined volume of the above liquid. It was sealed with a large slide to create a closed 1-ml chamber that would not dry out.

The preparation was transferred to the thermostage of the microscope (Jena-Mikroscope-250-CF) and heated 10-15 min to 37°C. Under objective 6.3 in a 512 μ^2 image the count of NP in the visual field of the microscope varied from 30 to 70. The dark-field image was stored in a Magiscan 2A processing unit with an inter-image interval of 1 min. Twenty images were recorded to obtain a reliable statistical picture of cell behavior. Subsequent mathematical processing yielded information about the velocity of cell movement [3].

RESULTS

The mean velocity of a pool of NP (each pool consisting of 30 to 70 NP) was determined in 37 healthy donors. It varied from 7.0 to 10.5 μ/min. The mean

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velocity of all pools was 8.8 μ/min (the normal value). For control, the effect of 5-HT on the mobility of NP of 8 donors was examined. The mean velocity of NP of healthy donors after 5-HT was added in a concentration of 10^{-9} g/ml increased from 8.8 to 12.3 μ/min.

To assess the mobility of NP, we collected blood from patients with wound infection whose clinical status was assessed as grave or extremely grave. At first the initial mean velocity of a pool of NP of a patient was determined. If it was low, the next preparation was made from the same sample. 5-HT was added to the solution and the mean velocity of the NP pool was assessed by the same method. In studies of the mobility of NP, an officinal 1% solution of serotonin adipinate was used. 5-HT in a concentration of 1×10^{-7} g/ml increased the mean velocity of NP pools from all 7 patients (Table 1).

The appearance of substances interfering with the normal interactions between 5-HT and its receptors in the blood (environment) leads to a relative 5-HT insufficiency of NP, which manifests itself in disorders of their integral function — decrease of the velocity of movement. Exogenous addition of 5-HT to the NP environment abolishes their insufficiency, which is confirmed by an increase (normalization) of their mobility. For the elimination of 5-HT insufficiency of NP *in vitro* a much higher concentration of 5-HT is needed than in studies with normal NP.

Depression of the mobility of NP, in turn, disrupts their normal (physiological) function, that is, lowers the nonspecific immunity. Therapy with some drugs is known to lead to neutropenia and even agranulocytosis, whose mechanism is unknown [1,4,5,14].

Some scientists have described such complications of psychopharmacological therapy as impaired contractility of the intestinal and vascular smooth muscles (functional ileus and hypotonia), thrombocytopenia, and agranulocytosis [13,14]. On the other hand, 5-HT is known to be capable of causing neutrophilosis [1,9]. Moreover, hyperserotoninemia is known to lead to thrombocytosis, and release of 5-HT from platelets hastens their destruction [6,8,10]. In addition, some authorities [5] indicate that in aplastic anemias, characterized by neutropenia and thrombocytopenia, the content of 5-HT in the blood is sharply reduced and this agent is recommended in hypo- and aplastic anemias [6].

Hence, published data and our findings indicate that 5-HT is one of the substances essential for maintaining the homeostasis of NP and platelets.

Oxygen-independent systems are known to be the main source of energy for mature NP [2,7,15]. 5-HT exerts its effect on NP in a virtually oxygen-free medium, whereas on the smooth muscles it exerts a normalizing effect in the presence of oxygen [10,12].

TABLE 1. Effect of Serotonin on the Mobility of Depressed Neutrophils (NP) ($p<0.001$)

Patient	Mean velocity of 30-70 NP, μ/min	
	initial	with serotonin
1	3.4	11.0
2	3.9	8.3
3	4.1	6.4
4	5.3	9.5
5	7.8	10.6
6	6.8	8.9
7	4.3	9.5

The ability of 5-HT to eliminate 5-HT insufficiency in both the presence and absence of oxygen is important in the treatment of diseases involving tissue hypoxia.

Administration of 5-HT in diseases involving impaired mobility of NP and their accelerated destruction will help normalize their mobility and preserve their physiological properties in the early stages of disease. Measurement of NP mobility before, during, and after treatment is an informative test for assessing the efficacy of 5-HT therapy.

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REFERENCES

1. Sh. Berchanu, *Clinical Hematology*, Bucharest (1985).
2. O. K. Gavrilov, G. I. Kozinets, and N. B. Chernyak, *Bone Marrow and Peripheral Blood Cells* [in Russian], Moscow (1985).
3. A. A. Galkin, E. A. Tumanov, M. M. Filippov, *et al.*, in: *Biochemical Problems of Surgery* [in Russian], Moscow (1991), pp. 43-47.
4. E. D. Gol'dberg, *Handbook of Hematology* [in Russian], Tomsk (1989).
5. I. A. Kassirskii and G. A. Alekseev, *Clinical Hematology* [in Russian], Moscow (1970).
6. M. D. Mashkovskii, *Drugs* [in Russian], Moscow (1993).
7. A. N. Mayanskii and O. I. Pikuza, *Clinical Aspects of Phagocytosis* [in Russian], Kazan (1993).
8. V. V. Men'shikov, L. S. Bassalyk, and G. A. Shapiro, *The Carcinoid Syndrome* [in Russian], Moscow (1972).
9. E. I. Pukhal'skaya, "Serotonin and cell proliferation in health and disease," Author's synopsis of Doct. Med. Sci. Dissertation [in Russian], Moscow (1966).
10. A. P. Simonenkov, "Functional ileus, disseminated intravascular blood coagulation, and the syndrome developing during poisoning with psychotropic agents as a clinical manifestation of serotonin insufficiency," Author's synopsis of Doct. Med. Sci. Dissertation [in Russian], Moscow (1992).
11. A. P. Simonenkov, V. D. Fedorov, A. V. Fedorov, *et al.*, *Vestn. Rossiisk. Akad. Med. Nauk*, No. 6, 11-15 (1994).
12. A. P. Simonenkov, V. D. Fedorov, A. V. Fedorov, *et al.*, *Ibid.*, No. 6, 27-30 (1995).
13. L. I. Spivak, V. A. Raiskii, and B. S. Vilenskii, *Complications of Psychopharmacological Therapy* [in Russian], Leningrad (1988).
14. M. J. Denham and I. Chanarin, *Blood Disorders in the Elderly*, Edinburgh - London - Melbourne - New York (1985).
15. R. Schmidt and G. Thews, *Human Physiology*, Berlin - Heidelberg - New York (1983).