Influence of bioresonance therapy in swing mode on the process of normalization of the leukocyte count of peripheral blood and changes in the cellular composition of the bone marrow of white mice on the model leukopenia L.A. Boqueriaone, N.T. Saliaone, O. L. Boqueriaone, L.T. Mikadzeone, M.Yu. Gotovsky2, D.V. Dzidziguri3 (oneNTSSSH them. A.N. Bakuleva,2Center "IMEDIS", Moscow, Russia, 3TSU named after Yves. Javakhishvili, Tbilisi, Georgia)

Leukopenia, a decrease in the absolute content of leukocytes in the unit blood volume, develops in infectious diseases (flu, visceral leishmaniasis, typhoid fever, malaria, etc.), as well as in diseases of the blood system. Along with viral infections and diseases of the hematopoietic organs, the cause of leukopenia can be the toxic side effect of medications on the bone marrow. As a result, leukopenia develops through allergic mechanisms [1, 2].

In a similar way, chemotherapy and radiation therapy for cancer patients also lead to leukopenia. The prognosis of treatment directly depends on the sensitivity of the tumor to drugs, the amount and dose of radiation used in chemotherapy and radiation therapy. Therefore, in treatment (chemotherapy and radiation therapy), it is often necessary to use highly toxic drugs and large doses, which by themselves, contribute to immunosuppression (leukopenia appears), and thereby bring the relapse of the disease closer and can

pose a threat to life. accompanying With these radical treatments, immune therapy aimed at

restoration of blood formula and life support systems affected by side effects of anticancer drugs [3–6].

Currently, there is an urgent need for agents that could reduce the toxic effect of cytostatics and radiation on white blood cells and multipotent bone marrow stem cells and slow down the development of leukopenia. This is essential

would expand the possibilities of chemotherapy and therapy, and in some cases, radiation would allow to achieve complete remission, therefore, increase life expectancy of patients [7-9].

We have previously shown on the model of leukopenia in white mice [10, 11] that BRT sessions according to strategy IV, carried out every other day for two weeks, contribute to the restoration of the peripheral blood formula of animals. In particular, it was shown that BRT according to the IV strategy contributes to the normalization of the leukocyte count of the peripheral blood of mice with pronounced leukopenia. Correction of disorders of the leukocyte count of the blood of mice, characteristic of leukopenia, is achieved by increasing the number of immature leukocytes. However, the mechanism of action of BRT remains unclear. In particular, it is not known: BRT accelerates the differentiation of immature progenitor cells, or stimulates the division of progenitor cells in the bone marrow.

Based on the above, aim of the present work was: the study of the effect of bioresonance therapy (BRT) on the process of normalization of the peripheral blood formula and the cellular composition of the bone marrow of white mice on the background of immunosuppression (caused by the introduction of cyclophosphamide).

In this regard, the following were delivered tasks:

1. Set the influence of BRT in the swing mode along all meridians on the process of normalization of the leukocyte count of the peripheral blood of white mice after a single injection of cyclophosphamide.

2. Establish the influence of BRT in swing mode along all meridians with unloading (cyclophosphamide in an inverse container) on the process of normalizing the leukocyte count of peripheral blood after a single injection of cyclophosphamide.

3. Establish the mechanisms of influence of BRT in the rocking mode for all meridians and BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container) on changes in the cellular composition of the bone marrow of white mice after a single injection of cyclophosphamide.

Research objects

1. Sexually mature white mice weighing 25–30 g, n = 270.

Material and research methods

Peripheral blood and bone marrow of experimental animals (mice).

Simulation of leukopenia in animals.

The criterion for the effectiveness of the therapy used was:

- determination of the total number of leukocytes in the peripheral blood (Goryaev's camera);

- determination of the leukocyte formula on peripheral blood smears;

- determination of changes in the cellular composition of the bone marrow of white mice.

Model immunosuppression (leukopenia) was called one-time intraperitoneal administration of cyclophosphamide at a dose of 280 mg per 1 kg of body weight. Blood was taken from the tails of animals. The total number of leukocytes was counted in the Goryaev chamber using a light microscope (magnification 10 × 15). In parallel, smears of peripheral blood and bone marrow were prepared. Preparations (blood and bone marrow smears) after fixation for 5 min. in the Main-Grunwald fixative, washed with distilled water. Giemsa dye (EUROTURBO,

DELTALAB, Spain). The colored preparations were transferred into running water. The leukocyte count and bone marrow were calculated in a light microscope (magnification 90 × 10). The reliability of the data obtained was assessed by the Student's test.

Results and its discussion

In order to study the effect of BRT in the rocking mode along all meridians on the process of normalization of the leukocyte formula of peripheral blood and changes in the cellular composition of the bone marrow of white mice, the animals were divided into three groups: I - intact mice (control group), II - animals after a single injection of cyclophosphamide; III - animals that, after a single injection of cyclophosphamide, underwent BRT in the rocking mode. all meridians every other day for two weeks (task number 1).

Studies have shown that on the 4th day after a single injection of cyclophosphamide, the total number of leukocytes in peripheral blood of white mice in groups II and III. An increase in the number of leukocytes in the peripheral blood of mice of both groups mentioned above is achieved on the 8th day after the injection of cyclophosphamide (Fig. 1).



Rice. one.Influence of BRT in the rocking mode along all meridians on the process of normalization of the leukocyte count of peripheral blood in white mice

However, as can be seen from Fig. 1, in the blood of animals of group III, an increase in the number of leukocytes in comparison with group II, already from the fourth day after the injection of cyclophosphamide, occurs more intensively. Moreover, the indicators of the total number of leukocytes in group III, on the eighth day, significantly exceed the corresponding indicators of group II, from which it follows that repeated exposure to BRT in swing mode along all meridians accelerates an increase in the total number of leukocytes in the blood of animals of the experimental group (Fig. 1).

The results of this series of experiments confirm the previously established corrective effect of bioresonance therapy according to strategy IV [10]. However, a comparative analysis of the curves shown in Fig. 1 and 1a, showed that, in contrast to BRT according to strategy IV (Fig.1a), the effect of BRT in the rocking mode along all meridians is manifested a week earlier after the injection of cyclophosphamide (Fig. 1).



Rice. 1a.Influence of BRT according to strategy IV on the process of normalization of the peripheral blood formula in mice after a single administration of cyclophosphamide

At the same time, the stimulating effect, i.e. the effect of BRT in the rocking mode along all meridians lasts up to two weeks. Fig. 1 also it follows that both in the II and III experimental groups, the increased indicators of the total number of leukocytes by the 30th day are close to the norm. In general, from the analysis of the curves shown in Fig. 1, it follows thatconducting BRT sessions in swing mode along all meridians causes acceleration the process of restoring the total number of leukocytes in the blood of mice after a single injection of cyclophosphamide.

In parallel, the analysis of peripheral blood smears of animals of all groups was carried out. On the 4th day after the injection of cyclophosphamide, the percentage of leukocytes in the peripheral blood of the animals of the studied groups slightly changed. For example, on blood smears of mice of group II, stab neutrophils and eosinophils are not visible, the number of segmented neutrophils decreases. In addition, the presence of mononuclear cells was revealed in experimental groups II and III. By the eighth day, against the background of a sharp decrease in lymphocytes, the number of segmented leukocytes and mononuclear cells. Accordingly, the percentage of

segmented leukocytes and mononuclear cells. Accordingly, the percentage of different types of cells is violated.

In the peripheral blood of animals of the experimental groups, the percentage of different types of cells, which sharply differs from the control, changes even more after two weeks. Attention is drawn to the increase in the number of mononuclear cells and immature, stab neutrophils. In addition, spontaneous restoration of the number of leukocytes (group II) occurs due to a sharp increase in stab forms of neutrophils, while in group III, there is mainly an increase in mature neutrophils, as well as mononuclear cells.

By the twenty-second day, as noted above, the total number of leukocytes returns to normal. The analysis of smears showed that stab and segmented neutrophils, i.e. there is a tendency to normalize the blood count. However, not only immature neutrophils, but also mononuclear cells are still present in the peripheral blood of mice of both experimental groups. These cell forms are present in the peripheral blood of experimental animals for 30 days after the administration of cyclophosphamide. Based on the results obtained, it can be concluded that BRT in the rocking mode along all meridians stimulates the release of both mature and immature forms of leukocytes into the peripheral blood of mice with pronounced leukopenia. Significant increase in the number segmented neutrophils in the peripheral blood of animals of group III after BRT in the rocking mode along all meridians, in comparison with animals of group II, indicates that the differentiation of immature precursors in the bone marrow is stimulated experienced mice.

Problem number 2. Influence of BRT swing mode along all meridians with in unloading (cyclophosphamide in inverse container) on the process normalization of the leukocyte count of peripheral mice against the white blood background of leukopenia.

The animals were divided into three groups: I - intact mice (control group), II animals after a single injection of cyclophosphamide; III - animals that, after a single injection of cyclophosphamide, underwent BRT in a rocking mode along all meridians with unloading (cyclophosphamide in an inverted container) every other day for two weeks. Studies have shown that 4 days after a single injection of cyclophosphamide, there is an increase in the total number of leukocytes in the peripheral blood of white mice in groups II and III. The maximum number of leukocytes in the peripheral blood of mice of both groups mentioned above was found on the 15th day after the injection of cyclophosphamide (Fig. 2). However, a comparative analysis of the curves showed that the indicators of the total number of leukocytes in group III on the 15th day significantly exceed the corresponding indicators of group II, from which it follows thatthat repeated exposure



Rice. 2.Influence of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container) on the process of normalization of the leukocyte count of peripheral blood in white mice

BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container) enhances the process of increasing the total number of leukocytes in the blood of animals of experimental group III (rice. 2).

Despite these differences, the total number of leukocytes on the 22nd day after the injection of cyclophosphamide decreases to normal in both experimental groups (Fig. 2). The results of this series of experiments also confirm the previously established corrective effect of bioresonance therapy. Moreover, comparative analysis of curves, presented at rice. 2\_and 1a,

showed that when using BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container), it turns out the reproduced image of the BRT curve according to strategy IV (Fig. 1a and 2), in contrast to the BRT in the swing mode along all meridians, as already noted above (Fig. 1), the effect of which is manifested a week earlier (8th day) after cyclophosphamide injections.

Analysis of smears of all groups of animals in this series of the experiment also showed that a violation of the percentage of various types of cells in the peripheral blood of animals is detected after 4 days. Lymphocytes predominate.

On the eighth day after the administration of cyclophosphamide, the number of mononuclear cells and immature, stab neutrophils increases. In addition, spontaneous restoration of the number of leukocytes (group II) occurs mainly due to a sharp increase in mature forms of neutrophils, as well as mononuclear cells.

The percentage violation is even more pronounced after two weeks. In particular, the number of immature forms increases significantly - stab neutrophils.

By the twenty-second day, as noted above (Fig. 2), the total number of leukocytes returns to normal. Analysis of smears showed that, accordingly, the number of stab and segmented neutrophils decreases, i.e. there is a tendency to normalize the blood count. However, not only immature neutrophils, but also mononuclear cells are still present in the peripheral blood of mice of both experimental groups. These cell forms are present in the peripheral blood of experimental animals for 30 days after the administration of cyclophosphamide.

Based on the results obtained, it can be concluded that BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverse container) enhances the release of both mature and immature forms of leukocytes into the peripheral blood of mice with pronounced leukopenia. An increase in the number of segmented neutrophils in the peripheral blood of animals on the 15th day after BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container), compared with group II, indicates that differentiation of immature is stimulated in this series of experiments. precursors in the bone marrow of experimental mice.

In order to explain the difference in the actions of BRT in swing mode along all meridians and BRT in swing mode along all meridians with unloading (cyclophosphamide in an inverted container) on the normalization process In peripheral blood of white mice with pronounced leukopenia, an analysis was made of changes in the cellular composition of the bone marrow of white mice within 30 days after a single injection of cyclophosphamide.

Problem number 3. To establish the mechanisms of influence of BRT in the rocking mode along all the meridians and BRT in the rocking mode along all the meridians with unloading (cyclophosphamide in an inverted container) on changes in the cellular composition of the bone marrow of white mice after a single administration of cyclophosphamide.

Based on the foregoing, we analyzed bone marrow smears of animals from the above groups. In fig. 3, curves are presented, reflecting the change in the number of myeloblasts in the bone marrow of white mice in dynamics (within 30 days after the injection of cyclophosphamide).Fig. 3 it follows that BRT in the rocking mode along all meridians stimulates the division of myeloblasts.The peak of proliferative activity appears on the 4th day after the injection of cyclophosphamide and the start of therapy.



Rice. 3.Influence of BRT in the rocking mode along all meridians on the change in the quantity myeloblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)

The process of differentiation of the myeloid series is also accelerated. Within one month, the number of promyelocytes increases in comparison with the control indicator (Fig. 4), as well as of segmented cells on the 8th day (Fig. 5).



Rice. 4.Influence of BRT in the rocking mode along all meridians on the change in the quantity promyelocytes in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)



Rice. five.Influence of BRT in the rocking mode along all meridians on the change in the quantity metamyelocytes - segmented neutrophils in the bone marrow of white mice in dynamics (within 30 days after injection of cyclophosphamide)

Regarding the regulation of proliferative activity, it turned out that BRT in the rocking mode along all meridians stimulates only the division of blast forms of lymphocytes and megakaryocytes (Fig. 6 and Fig. 7).



Rice. 6.Influence of BRT in the rocking mode along all meridians on the change in the quantity lymphoblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)



Rice. 7.Influence of BRT in the rocking mode along all meridians on the change in the quantity megakaryoblasts - megakaryocytes in the bone marrow of white mice in dynamics (within 30 days after cyclophosphamide injection)

Interesting dynamics is also observed in red, the blood. In particular proliferation of erythroblasts increases (Fig. 8) and at the same time differentiation of basophilic and oxyphilic pronormocytes and release to the periphery of mature cells (Figs. 9 and 10).



Rice. eight.Influence of BRT in the rocking mode along all meridians on the change in the quantity erythroblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)



Rice. nine.Influence of BRT in the rocking mode along all meridians on the change in the quantity basophilic normocytes in the bone marrow of white mice in dynamics





Rice. 10.Influence of BRT in the rocking mode along all meridians on the change in the quantity oxyphilic normocytes in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)

Analysis of smears showed that BRT in the rocking mode along all meridians stimulates the proliferation of blast forms in the bone marrow of white mice. On the 4th day after the start of therapy, there is a pronounced increase in the number of myeloblasts, as well as segmented neutrophils (8th day, 22nd day) and causes acceleration normalization formulas peripheral blood, originating from an increase in the number of segmented neutrophils (fig. 1).

A different picture is observed in the case of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container). In this case, analysis of bone marrow smears showed that only the differentiation of myeloblasts is stimulated (Fig. 11).



Rice. eleven.Influence of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverse container) on the change in the amount myeloblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)

As a result, the number of myeloblasts and promyelocytes is below normal (Fig. 12).



Rice. 12.Influence of BRT in swing mode along all meridians with unloading (cyclophosphamide in inverse container) on the change quantity promyelocytes in the bone marrow white mice in dynamics (within 30 days after cyclophosphamide injection)

Therefore, increases number of metamyelocytes - segmented neutrophils and the release of mature cells into the peripheral blood. This can explain gain normalization of the blood count on the 15th day after the start of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container).

Analysis of smears also showed that when BRT was performed in a rocking mode along all meridians with unloading (cyclophosphamide in an inverted container), stimulation of proliferation and other blast forms of the bone marrow was not detected (Figs. 13, 14 and 15).



Rice. 13.Influence of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverse container) on the change in the amount lymphoblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)



Rice. fourteen.Influence of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverse container) on the change in the amount megakaryoblasts - megakaryocytes in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)



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Rice. fifteen.Influence of BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverse container) on the change in the amount erythroblasts in the bone marrow of white mice over time (within 30 days after the injection of cyclophosphamide)

## Conclusions:

1. BRT in the swing mode along all meridians calls acceleration process restoration of the total number of leukocytes in the blood of mice after a single injection of cyclophosphamide.

2 Acceleration of the growth of the total number of leukocytes is caused by the stimulating effect of BRT in the rocking mode along all meridians on the division of blast cells (especially myeloblasts) in the bone marrow of animals.

3. BRT in rocking mode along all meridians with unloading (cyclophosphamide in inverse container) enhances the growth of the total number of leukocytes in the blood of animals after a single administration of cyclophosphamide.

4. Strengthening the restoration of the total number of leukocytes in the blood of mice after a single administration of cyclophosphamide during BRT in the rocking mode along all meridians with unloading (cyclophosphamide in an inverted container), it is mediated by the stimulating effect of bioresonance therapy on the process of differentiation of blast cells and the release of some immature forms of leukocytes into the peripheral blood.

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