Exp Oncol 2015
37, 2, 135–138

Experimental ONCOLOGY

PREVENTION OF MYELOSUPPRESSION BY COMBINED TREATMENT WITH ENTEROSORBENT AND GRANULOCYTE COLONY-STIMULATING FACTOR

O.O. Shevchuk[1,], K.A. Posokhova[1], I.N. Todor[2], N.Yu. Lukianova[2], V.G. Nikolaev[2], V.F. Chekhun[2]*
[1]I.Ya. Horbachevsky Ternopil State Medical University, Ternopil 46001, Ukraine
[2]R.E. Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology, NAS of Ukraine, Kyiv 03022, Ukraine

Hematotoxicity and its complication are the prominent limiting factors for rational treatment of malignancies. Granulocyte colony-stimulating factor (G-CSF) is used to increase granulocyte production. It has been shown previously that enterosorption causes prominent myeloprotective activity also. Still, no trial was performed to combine both of them. ***Aim*****: To study the influence of combination of enterosorption and pharmaceutical analogue of naturally occurring G-CSF (filgrastim) on bone marrow protection and the growth of grafted tumor in a case of injection of melphalan (Mel).** ***Materials and Methods*****: Mel injections were used for promotion of bone marrow suppression in rats. Carbon granulated enterosorbent C2 (IEPOR) was used for providing of enteral sorption detoxifying therapy. Filgrastim was used to increase white blood cells (WBC) count.** ***Results*****: The simultaneous usage of enterosorption and filgrastim had maximum effectiveness for restoring of all types of blood cells. WBC count was higher by 138.3% compared with the Mel group. The increase of platelets count by 98.5% was also observed. In the group (Mel + C2 + filgrastim) the absolute neutrophils count was twofold higher, in comparison with rats of Mel group.** ***Conclusion*****: Simultaneous administration of G-CSF-analogue and carbonic enterosorbent C2 is a perspective approach for bone marrow protection, when the cytostatic drug melphalan is used. Such combination demonstrates prominent positive impact on restoring of all types of blood cells and had no influence on the antitumor efficacy.**
Key Words**: myelotoxicity, melphalan, enterosorption, granulocyte colony-stimulating factor.**

Uncontrolled cell division is one of the features of cancer cells. The common side effects of anti-cancer drugs, which have no selective activity against tumors, include damaging of all highly proliferative cells. The critical tissues and organs are the bone marrow, mucosa of gastrointestinal tract, ovaries and testis in this case.

Dose-dense and dose-intense chemotherapy may result in the suppression of activity of proliferating hematopoietic precursor cells, leading to deprivation of blood cells and incidence of life threatening hemorrhage and infections [1]. Hematotoxicity and its complication similar to febrile neutropenia and sepsis are the prominent limiting factors for successful treatment of malignancies. It often results in lengthy treatment delays and dose reductions, which have been shown to compromise the treatment.

The endogenous intoxication syndrome can develop in oncological patients as well. The main factors are: "tumor disposition"; tumor-dependent compression or tumor invasion; destruction of neoplastic tissues due to aggressive treatment; surgical trauma; hemorrhage; sepsis; prominent oxidative stress, etc. [2–4]. All these features decrease the activity of detoxifying systems of organism, i.e. elimination of waste products, inhibit the liver and kidney functions and disrupt the important biochemical process.

Over the past years, a new class of drugs has become available to boost marrow function — the recombinant hematopoietic growth factors. Erythropoietin is used to increase red blood cells (RBC) production; granulocyte colony-stimulating factor (G-CSF) and granulocyte-macrophage colony-stimulating factor — to increase granulocyte production; and interleukin-11 is administered to increase platelet count [1].

Finally, studies have shown that filgrastim, lenograstim and pegfilgrastim (G-CSF analogues) have clinical efficacy. The use of any of these agents was recommended to prevent febrile neutropenia and related complications. Despite the fact that G-CSF reduces the neutropenia duration for 1–2 days, it has no influence on such clinical parameters like fever severity, intensity of antibiotics therapy and lethality indices. However, additional G-CSF-therapy increases the cost of treatment notably [5–7].

Enteral sorption detoxifying therapy is used widely today to decrease the systemic signs of acute or chronic intoxication and oxidative stress in case of renal failure, hepatic insufficiency, burn toxicosis, and acute enteric infections [8–13], as well as complications of chemo- and radiotherapy of cancer [3, 12, 14–16]. It was shown previously that enterosorption causes prominent myeloprotective activity in case of applying of therapeutic doses of alkylating agent melphalan (Mel) and demonstrates some other positive effects [17].

Still, to the best of our knowledge, no trial was performed to combine this technique with myeloprotective cytokines. At the same time, it is known, that some metabolic corrections by antioxidants and vitamins can promote the tumor growth [18]. So, it is very important to understand what influence the different additive medicines might have on malignancy progress.

The aim of our investigation is to study the efficacy of combination of enterosorption and G-CSF analogue to protect the bone marrow in case of injection

Submitted: December 21, 2014.
*Correspondence: E-mail: doctor.oksana@rambler.ru
Tel.: +38 (0352) 52 39 87

Abbreviation used: BET – Brunauer – Emmett – Teller model of pore distribution calculation; G-CSF – granulocyte colony-stimulating factor; Mel – melphalan; RBC – red blood cells; WBC – white blood cells.

of high dose of alkylating bifunctional cytostatic drug of the bischloroethylamine type — Mel, which is widely used in the palliative treatment of patients with multiple myeloma, advanced ovarian cancer, etc. [19].

The influence of combined administration of enterosorption and G-CSF analogue filgrastim on the growth of grafted Guerin's carcinoma in rats is studied as well.

MATERIALS AND METHODS

Melphalan (Mel) was used for promotion of bone marrow suppression in rats. It is an alkylating anti-cancer drug Mel, also known as L-phenylalanine mustard (L-PAM), phenylalanine mustard, or L-sarcolysin.

Carbon granulated enterosorbent C2 (R.E. Kavetsky Institute of Experimental Pathology, Oncology and Radiobiology, NAS of Ukraine — IEPOR) was used for detoxifying therapy by enteral sorption therapy. Parameters of carbonic enterosorbent C2 were the following: a bulk density $\gamma = 0.18$ g/cm^3, a diameter of granules 0.15–0.25 mm, the BET pore surface — 2162 m^2/g.

Filgrastim was used to increase white blood cells (WBC) count. It is a G-CSF analogue which used to stimulate the proliferation and differentiation of granulocytes, which is produced by recombinant DNA technology.

Animals and experiment design. The first part of experiment was carried out on 65 healthy male inbred albino Wistar rats weighing 200 ± 20 g from the IEPOR animal facility. All procedures were performed according to the rules and requirements of European Convention for the Protection of Vertebrate Animals Used for Experimental and Other Scientific Purposes and a local Ethic Committee of IEPOR. Animals were randomly distributed into 5 groups: group 1 — intact rats (n = 25); group 2 — rats treated with Mel (n = 10, control group); group 3 — animals treated with Mel and carbonic enterosorbent C2 (Mel + C2, n = 10); group 4 — rats treated with Mel and filgrastim (Mel + filgrastim, n = 10); and group 5 — rats treated with Mel and both carbonic enterosorbent C2 and filgrastim (Mel + C2 + filgrastim, n=10).

Experiment was carried out to study the influence of administration of enterosorption and G-CSF on the dynamics of tumor growth. For this purpose, we used 35 female inbred albino Wistar rats with grafted tumors. Suspension of Guerin's carcinoma tumor tissues (23%, 0.4 ml) was applied subcutaneously to the back of animals. All rats were randomly distributed into 5 groups (n = 7 in each group) after formation of tumor in 8 days: group 1 — Guerin's carcinoma-bearing rats (*tumor control*); group 2 — Guerin's carcinoma-bearing rats treated with Mel (*tumor + Mel*); group 3 — treated with Mel and C2 (*tumor + Mel + C2*); group 4 — treated with Mel and filgrastim (*tumor + Mel + filgrastim*) and group 5 — with Mel, C2 and filgrastim (*tumor+ Mel + C2 + filgrastim*).

The dynamics of tumor growth in all groups was calculated by the formula:

$$V = a \cdot b \cdot c \cdot \pi/6,$$

where V — the volume of tumor (cm^3); a, b, c — orthogonal sizes of tumor (cm).

Mel has been injected intravenously as a single treatment in tail vein 4.0 mg/kg of body weight. The dose of alkylating cytostatic was 5.5 mg/kg for the rats with grafted Guerin's carcinoma. A suspension of carbonic enterosorbent (C2 dosage of 5 ml/1 kg of animals body weight, 900 mg of dry mass of enterosorbent) in appropriate quantity of distilled water was introduced via the tube into rat stomach during 3 days before the day of Mel injection and during 7 days after injection (1 time per 1 day). The filgrastim was injected once a day starting from the next day after Mel applying during 4 days in dose 50 mcg/kg. Rats of Mel group and intact group were given equivalent quantity of distilled water. Animals of intact group received intravenously equal quantities of saline instead of Mel.

The rats were weighted and blood was taken from the heart under Ketamine hydrochloride general anesthesia on the 8th day after Mel injection. We used automatic hematology analyzer BC-3000Plus Mindray for evaluation of complete blood cell count. Peripheral blood smears were made as well. Panoptic method of Pappenheim was used for the staining of cytological smears [20].

Statistical analysis. Since data were not normally distributed in all groups (Shapiro — Wilk Normality Test), non-parametric tests (Mann — Whitney U-test and one-way ANOVA test) were performed for analysis of all data (significant at pb 0.05). The data were expressed as the mean ± standard error of the mean (M ± m). All statistical calculations were performed on the separate data from each individual with using Origin 7.5 software (OriginLab Corporation, USA).

RESULTS

It was observed that Mel at a dose of the 4 mg/kg influenced negatively on blood cells. For example, WBC count decreased by 75.6%, while platelets count — by 59.9% compared with intact rats (Table 1). Decrease of red blood indices was not so prominent: erythrocyte count was lower by 10.3% and there were no changes of hemoglobin level. The enterosorbent C2 promoted the increase of WBC count by 43.8%, as well as the tendency of platelets concentration increase was noted too. Filgrastim caused the significant rising of WBC count by 57.0%. The most effective results were observed in case of combined use (C2 and filgrastim): amount of WBC was higher by 138.3% compared with the Mel group; and higher (by 65.8%) compared with Mel + C2 group. The leukocytes count was higher by 51.7% in rats which were administered sorbent C2 and filgrastim compared with usage of filgrastim alone. Interestingly, we observed the increase of platelets by 98.5% in case of combined treatment of enterosorbent C2 and filgrastim. This index is higher by 126.4% compared with Mel + filgrastim group.

As one can see in leucocytes formula (Table 2), Mel at dose of 4 mg·kg^{-1} caused the 89.0% increase of neutrophils, while the percent of lymphocytes decreased by 46.1%, and amount of monocytes was 3 times-fold higher (increased by 200.5%). The usage of enteral sorption therapy and filgrastim had no influence on percentage of leukocytes formula. Simultaneously, it was observed that absolute neutrophils count decreased

Table 1. Hematological indices in case of Mel injection at dose 4 mg·kg^{-1} and its correction by carbonic enterosorbent C2 and filgrastim (M ± m)

Parameters	Groups				
	Intact rats (n = 25)	Mel (n = 10)	Mel + C2 (n = 10)	Mel + filgrastim (n = 10)	Mel + C2 + filgrastim (n = 10)
Leukocytes, ×10^9/l	5.24 ± 0.29	1.28 ± 0.15*	1.84 ± 0.17**	2.01 ± 0.16$^{\#}$	3.05 ± 0.28**, $^{\#}$, $^{\#\#}$
Erythrocytes, ×10^{12}/l	7.34 ± 0.12	6.58 ± 0.17*	6.66 ± 0.20	6.85 ± 0.19	6.80 ± 0.15
Hemoglobin, g/l	134.32 ± 1.69	115.90 ± 3.46	122.10 ± 2.24	121.60 ± 2.84	122.0 ± 2.97
Platelets, ×10^9/l	634.84 ± 22.97	254.60 ± 45.59*	328.10 ± 65.65	223.20 ± 40.45	505.40 ± 70.68**, $^{\#\#}$

Notes: significance $p < 0.05$ comparatively with: *intact rats; **Mel treatment; $^{\#}$Mel + C2 group treatment; $^{\#\#}$Mel + filgrastim treatment.

Table 2. Leukocytes formula in case of Mel injection at dose 4 mg·kg^{-1} and its correction by carbonic enterosorbent C2 and filgrastim (M ± m, n = 10)

Parameters	Groups				
	Intact rats	Mel	Mel + C2	Mel + filgrastim	Mel + C2 + filgrastim
Promyelocytes, %	–	–	–	–	–
Myelocytes, %	–	–	–	–	–
Metamyelocytes, %	–	–	–	0.40 ± 0.27	0.22 ± 0.22
Neutrophils, %	25.90 ± 1.83	49.0 ± 3.76*	42.83 ± 3.19	51.30 ± 5.61	60.89 ± 5.19
Eosinophils, %	1.0 ± 0.45	–	–	–	–
Basophils, %	–	–	–	0.10 ± 0.10	–
Lymphocytes, %	68.4 ± 3.10	36.88 ± 4.42*	42.34 ± 3.07	36.0 ± 4.71	31.11 ± 4.05
Monocytes, %	4.70 ± 0.97	14.12 ± 2.12*	14.83 ± 2.61	12.20 ± 2.45	7.78 ± 2.09

Notes: significance $p < 0.05$ comparatively with: *intact rats; **Mel treatment; $^{\#}$Mel + C2 treatment; $^{\#\#}$Mel + filgrastim treatment.

Table 3. Absolute blood cells count in case of Mel injection in dose 4 mg·kg^{-1} and its correction by carbonic enterosorbent C2 and filgrastim (M ± m, n = 10)

Parameters	Groups				
	Intact rats	Mel	Mel + C2	Mel + filgrastim	Mel + C2 + filgrastim
Promyelocytes, ×10^9/l	–	–	–	–	–
Myelocytes, %	–	–	–	–	–
Metamyelocytes, %	–	–	–	0.01 ± 0.01	0.01 ± 0.01
Neutrophils, ×10^9/l	1.37 ± 0.09	0.63 ± 0.05*	0.79 ± 0.06	1.05 ± 0.12**	1.86 ± 0.16**, $^{\#}$, $^{\#\#}$
Eosinophils, ×10^9/l	0.05 ± 0.02	–	–	–	–
Basophils, ×10^9/l	–	–	–	–	–
Lymphocytes, ×10^9/l	3.58 ± 0.16	0.47 ± 0.06*	0.78 ± 0.06**	0.71 ± 0.10	0.94 ± 0.12**
Monocytes, ×10^9/l	0.24 ± 0.05	0.18 ± 0.03	0.27 ± 0.05	0.24 ± 0.05	0.24 ± 0.06

Notes: significance $p < 0.05$ comparatively with: *intact rats; **Mel group; $^{\#}$Mel + C2 group; $^{\#\#}$Mel + filgrastim group.

by 53.8% and absolute lymphocytes count — by 86.8% (Table 3).

The combined treatment with carbonic enterosorbent C2 and filgrastim had maximum effectiveness for restoring of all types of WBC. For example, in group Mel + C2 + filgrastim the neutrophils absolute count was higher by 196.1%, lymphocytes — by 101.0% compared with Mel group. Neutrophils absolute count in mentioned group was higher by 135.6% compared with usage of enterosorbent alone and higher by 77.3% compared with usage of filgrastim alone.

Analysis of Guerin's carcinoma growth in the groups of rats with grafted tumors has shown that administration of carbonic enterosorbent C2 or filgrastim, or its combination, had no effect on the antitumor efficacy of Mel. On the 13th day after tumor transplantation, the average sizes of tumor volume were the same in Guerin's carcinoma-bearing rats, which have got the enterosorbent and/or filgrastim besides the Mel, and in rats treated with Mel alone (Table 4).

Table 4. The dynamics of Guerin's carcinoma growth (volume, cm^3)

Group	Days after transplantation of Guerin's carcinoma			
	7	10	13	17
Tumor control (n = 7)	0.9 ± 0.1	3.7 ± 0.2	7.7 ± 0.2	11.3 ± 0.2
Tumor + Mel (n = 7)	0.9 ± 0.1	3.8 ± 0.3	5.8 ± 0.2*	6.6 ± 0.2*
Tumor + Mel+ filgrastim (n = 7)	0.8 ± 0.2	3.7 ± 0.2	5.8 ± 0.3*	6.4 ± 0.3*
Tumor + Mel + C2 (n = 7)	1.0 ± 0.1	3.5 ± 0.2	5.7 ± 0.3*	6.1 ± 0.2*
Tumor + Mel + filgrastim + C2 (n = 7)	1.0 ± 0.1	3.8 ± 0.3	5.7 ± 0.3*	6.0 ± 0.4*

Note: * the significance $p < 0.05$ comparatively with the tumor control.

The slight tendency to enhancement of Mel inhibiting action in case of simultaneous usage of enteral detoxifying sorption therapy and filgrastim was observed on day 17.

DISCUSSION

Myelosuppression is a common and expected side effect of antitumor treatment, especially using alkylating agents. Because of it, patients may experience anemia, neutropenia, and thrombocytopenia. Depending on their severity, these side effects can have a negative impact on patient's medical treatment and quality of life leading to therapy interruption or reduction and causing life-threatening complications. Neutropenia, including febrile neutropenia, can result in life-threatening infections. Overall mortality rates due to febrile neutropenia are about 5% in patients with solid tumors and as high as 11% in some hematological malignancies. Prognosis is the worst in patients with proven bacteremia, with mortality rates of 18% in Gram-negative and 5% in Gram-positive bacteremia [21, 22].

Today, G-CSF is being used prophylactically to support the dose-dense and dose-intense chemotherapy regimens, in spite of the insufficient data to assess the impact of G-CSF on disease-free and overall survival [23] and side effects of such treatment [24]. Results of this study show the potent myeloprotective effect of G-CSF as filgrastim caused the significant rising of WBC (by 57.0%). At the same time, there is no effect on other types of blood cells in case of its administration alone. Filgrastim did not show a potent impact on restoration of antioxidant-prooxidant balance also.

In case of enteral sorption detoxifying therapy, especially in combination with the filgrastim, one can see the 2.3-fold increase of platelets count, compared with Mel + filgrastim group. It is well known that endogenic intoxication is prominent negative factor in patients with malignant tumors [25]. Enterosorption demonstrates

the proved systemic effects in such cases, diminishing the disruption of metabolism [3, 11, 26].

The overall positive effect of simultaneous administration of enterosorption and G-CSF could be explained by their different effects on human organism. The hematopoietic growth factor stimulates the bone marrow directly, while there are systemic positive effects of adsorptive therapy, such as the removal of toxic substances, some intermediate metabolites, attenuation of "metabolic chaos" and oxidative stress. These factors improve the detoxifying function of body itself and, possibly, binding of some substances, which can limit the restoration of bone marrow function. Enteral sorption therapy and G-CSF do not cause any negative effect on the antitumor activity of cytostatic drug also, as was shown by results of our experiment.

CONCLUSIONS

Simultaneous administration of filgrastim, an analogue of G-CSF and carbonic enterosorbent C2 is a prospective approach for bone marrow protection in case of usage of cytostatic drug Mel.

In case of the cytostatic bone marrow suppression, simultaneous administration of the hematopoietic growth factor (G-CSF, filgrastim) and carbonic enterosorbent C2 demonstrated significant myeloprotective effect and synergy in comparison with single effects of each agent alone. We observed restoration of all types of blood cells and more pronounced increase of absolute neutrophils and lymphocytes counts. Interestingly, platelets count increased as well.

Filgrastim used in combination with the carbonic enterosorbent C2 and its alone administration had no effect on the antitumor efficacy of Mel.

REFERENCES

1. **Trillet-Lenoir V, Green J, Manegold C, *et al.*** Recombinant granulocyte colony stimulating factor reduces the infectious complications of cytotoxic chemotherapy. Eur J Cancer 1993; **29**: 319–24.

2. **Kavetsky RE.** Host and tumor interaction. K: Naukova dumka, 1977. 236 p. (in Russian).

3. **Nikolaev VG, Sakhno LA, Snezhkova EA, *et al.*** Carbon adsorbents in oncology: achievements and perspectives. Exp Oncol 2011; **33**: 2–8.

4. **Esper DH, Harb WA.** The cancer cachexia syndrome: a review of metabolic and clinical manifestations. Nutr Clin Pract 2005; **20**: 369–76.

5. **Aarts MJ, Grutters JP, Peters FP, *et al.*** Cost effectiveness of primary pegfilgrastim prophylaxis in patients with breast cancer at risk of febrile neutropenia. J Clin Oncol 2013; **31**: 4283–9.

6. **Girnius S, Seldin DC, Skinner M, *et al.*** Hepatic response after high-dose melphalan and stem cell transplantation in patients with AL amyloidosis associated liver disease. Haematologica 2009; **94**: 1029–32.

7. **Bijelic L, Sugarbaker PH, Stuart OA.** Hyperthermic intraperitoneal chemotherapy with melphalan: a summary of clinical and pharmacological data in 34 patients. Gastroenterol Res Pract 2012: Article ID 827534.

8. **Lukichev BG, Shostka GD, Strelko VV, *et al.*** 10-years' experience in using enterosorption for treating chronic kidney failure. Ter Arkh 1992; **64**: 52–6 (in Russian).

9. **Ryss ES, Riabov SI, Lutoshkin MB.** A comparative evaluation of the efficacy of the clinical use of SKN-4M-. SKT-6A- and polifepan-type sorbents in treating patients with chronic kidney failure (clinical and experimental studies). Ter Arkh 1996; **68**: 39–43 (in Russian).

10. **Lukichev BG, Panina IYu.** Prognosis of the effectiveness of enterosorption in chronic renal failure. Nephrology 2005; **9**: 89–94 (in Russian).

11. **Mikhalovsky SV, Sandeman SR, Howell CA, *et al.*** Biomedical applications of carbon adsorbents. Novel carbon adsorbents. *J Tascyn*, ed. First edition. Elsevier, 2012: 639–69.

12. **Nikolaev VG.** Enterosgel: a novel organosilicon enterosorbent with a wide range of medical application. Biodefence: advanced materials and methods for health protection. *S Mikhalovsky, AB Khajibaev,* eds. Springer, 2011: 199–222.

13. **Chernykhova EA, Anikhovskaya IA, Gataullin UK, *et al.*** Enterosorption as important mean of eliminating of chronic endotoxin aggression. Human Physiol 2007; **33**: 135–6 (in Russian).

14. **Sakhno LA, Yurchenko OV, Maslenniy VN, *et al.*** Enterosorption as a method to decrease the systemic toxicity of cisplatin. Exp Oncol 2013; **35**: 45–52.

15. **Manikhas GM, Akhytin VA, Fridman MKh, *et al.*** Effect of enterosorption on immunologic parameters of patients with colorectal cancer in the postoperative period. Vopr Onkol 2009; **55**: 66–71 (in Russian).

16. **Ponomarova OV, Pivnyuk VM, Nosko MM, *et al.*** Prophylaxis by coal enterosorbent of acute and extended emethogenic toxicity of cancer patient chemotherapy. Oncology 2008; **3**: 370–3 (in Ukrainian).

17. **Shevchuk OO, Posokhova KA, Sidorenko AS, *et al.*** The influence of enterosorption on some haematological and biochemical indices of the normal rats after single injection of melphalan. Exp Oncol 2014; **36**: 1–7.

18. **Sayin VI, Ibrahim MX, Larsson E, *et al.*** Antioxidants accelerate lung cancer progression in mice. Sci Transl Med 2014; **6**: 221ra15.

19. **Palumbo A, Cavallo F, Gay F, *et al.*** Autologous transplantation and maintenance therapy in multiple myeloma. N Engl J Med 2014; **371**: 895–905.

20. **Ruter A, Gunzer U.** Differentiation of granulocytes in Pappenheim stained blood cell smears using standardized cytophotometric analysis. Blut 1984; **48**: 307–20.

21. **Innes H, Lim SL, Hall A, *et al.*** Management of febrile neutropenia in solid tumours and lymphomas using the multinational association for supportive care in cancer (MASCC) risk index: feasibility and safety in routine clinical practice. Support Care Cancer 2008; **16**: 485–91.

22. **Hamalainen S, Kuittinen T, Matinlauri I, *et al.*** Severe sepsis in autologous stem cell transplant recipients: microbiological aetiology, risk factors and outcome. Scand J Infect Dis 2009; **41**: 14–20.

23. **Kuderer NM, Dale DC, Crawford J, *et al.*** Impact of primary prophylaxis with granulocyte colony-stimulating factor on febrile neutropenia and mortality in adult cancer patients receiving chemotherapy: a systematic review. J Clin Oncol 2007; **25**: 3158–67.

24. **Lei KIK, Leung WT, Johnson PJ.** Serious pulmonary complications in patients receiving recombinant granulocyte colony-stimulating factor during BACOP chemotherapy for aggressive non-Hodgkin's lymphoma. Br J Cancer 1994; **70**: 1009–13.

25. **Skipworth RJE, Stewart GD, Dejong CHC, *et al.*** Pathophysiology of cancer cachexia: much more than host — tumour interaction? Clin Nutr 2007; **26**: 667–76.

26. **Lukichev BG, Panina IYu.** Prognosis of the effectiveness of enterosorption in chronic renal failure. Nephrology 2005; **9**: 89–94 (in Russian).