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Original article

PECULIARITIES OF USING LYMPHOCYTE TEST TO PREDICT THE SEVERITY OF ACUTE RADIATION INJURY

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Abstract. One of the methods of biological dosimetry is the use of information on the concentration of lymphocytes in the peripheral blood of victims in the first days after irradiation.
The aim of the study was to validate the lymphocyte test method for predicting the severity of acute radiation injury, taking into account the dose rate factor.
Materials and research methods. The method of investigation was a correlational analysis of clinical, dosimetric and laboratory data of the victims of the accident at the Chernobyl nuclear power plant in 1986 (n=65) and in radiation accidents with gamma-neutron irradiation (n=19). The data were taken from the database of acute radiation injuries in humans of A.I. Burnazyan Federal Medical Biophysical Center of the Federal Medical and Biological Agency of Russia.
Results of the study and their analysis. The results of correlation analysis indicated that average lymphocyte concentration in the range of 0.4-0.8×10^9/l on day 3-6 post-irradiation with dose rates greater than 2 Gy/h resulted in an average dose estimate which was 40.0% higher than that for dose rates of less than 2 Gy/h. Absolute error of dose estimation is (±1.0-1.5) Gy. For lymphocyte concentrations higher than 0.8×10^9/l the prognosis is uncertain: the range of dose assessment variability is 1-4 Gy. At a lymphocyte concentration of less than 0.4×10^9/l the average dose estimate is more than 4.0 Gy, corresponding to a severe or extremely severe degree of acute radiation disease. The predicted degree of severity of radiation injuries in the presence of the neutron component of radiation is lower in comparison with the predictions based on the data on the victims of the Chernobyl accident. It is concluded that the identified dependencies can be used for medical triage of the victims at advanced stages of medical evacuation. For the purpose of correct routing of medical evacuation to specialized centers, it is advisable to allocate 4 treatment-evacuation groups.

Key words: *acute radiation syndrome, disaster, dosimetry, emergency situation, lymphocyte tests, triage*

Conflict of interest. The authors declare no conflict of interest

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Оригинальная статья

ОСОБЕННОСТИ ИСПОЛЬЗОВАНИЯ ЛИМФОЦИТАРНОГО ТЕСТА ДЛЯ ПРОГНОЗИРОВАНИЯ СТЕПЕНИ ТЯЖЕСТИ ОСТРОГО ЛУЧЕВОГО ПОРАЖЕНИЯ

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Резюме. Одним из методов биологической дозиметрии является использование информации о концентрации лимфоцитов в периферической крови пострадавших в первые дни после облучения.
Цель исследования – валидация метода лимфоцитарного теста для прогнозирования степени тяжести острого лучевого поражения с учётом фактора мощности дозы.
Материалы и методы исследования. Метод исследования – корреляционный анализ клинико-дозиметрических и лабораторных данных пострадавших при аварии на Чернобыльской АЭС – ЧАЭС в 1986 г. (n=65) и при радиационных авариях с гамма-нейтронным облучением (n=19). Данные взяты из базы данных по острым лучевым поражениям человека Федерального медицинского биофизического центра им. А.И.Бурназяна ФМБА России.
Результаты исследования и их анализ. По результатам корреляционного анализа установлено, что при средней концентрации лимфоцитов в диапазоне 0,4–0,8×10^9/л на 3-и – 6-е сутки после облучения при мощности дозы более 2 Гр/ч в среднем оценка дозы оказывается выше на 40,0%, чем для мощности дозы менее 2 Гр/ч. Абсолютная погрешность оценки дозы – (±1,0–1,5) Гр. При концентрации лимфоцитов более 0,8×10^9/л прогноз оказывается неопределенным: диапазон вариабельности оценки дозы – 1–4 Гр. При концентрации лимфоцитов менее 0,4×10^9/л средняя оценка дозы составляет

более 4,0 Гр, что соответствует тяжелой или крайне тяжелой степени тяжести острой лучевой болезни (ОЛБ). Прогнозируемая степень тяжести лучевого поражения при наличии нейтронной компоненты излучения оказывается ниже по сравнению с прогнозом, основанным на данных о пострадавших при аварии на ЧАЭС. Сделаны выводы: выявленные зависимости можно использовать для медицинской сортировки пострадавших на передовых этапах медицинской эвакуации; в целях корректной маршрутизации медицинской эвакуации в специализированные центры целесообразно выделять 4 лечебно-эвакуационные группы.

Ключевые слова: *дозиметрия, лечебно-эвакуационные группы, лимфоцитарный тест, маршрутизация, медицинская сортировка, медицинская эвакуация, прогнозирование, радиационные аварии, степень тяжести лучевого поражения, чрезвычайные ситуации*

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Introduction

Despite tightening of requirements for protection of industrial facilities of nuclear power industry, actual risk of emergencies remains high. Liquidation of medical and sanitary consequences of large-scale radiation accidents has shown that organization of medical and evacuation measures is a priority task in the complex of measures to minimize accidents. Improvement of such measures is necessary for the early provision of emergency medical aid and optimal routing of the injured to the relevant specialized medical centers. For this purpose, in the advanced stages of medical evacuation, it is necessary to carry out medical triage of those affected by ionizing radiation. This triage has its own characteristics associated with determining the radiation dose and severity of acute radiation sickness. In particular, in the absence of data from individual dosimeters the doctor is forced to make a diagnosis and a triage decision based on clinical and hematological indicators — biological dosimetry.

Among the methods of biological dosimetry, the lymphocyte test is widely used because lymphocytes are the most radiologically sensitive blood cells available for analysis [1]. Strict correlation between radiation dose in the range of 1-10 Gy and the number of dead cells allows to use laboratory data on the number of lymphocytes in peripheral blood as a criterion for evaluation of acute radiation disease severity during first 7 days after irradiation [1-10]. The necessity of distinguishing the groups of patients according to the severity of acute radiation disease allows to create so called "medical-tactical corridor". The boundaries of this corridor define the main, most difficult for treatment, group of patients with acute radiation sickness of moderate and high severity.

Empirical dependence of the number of lymphocytes in blood tests on the radiation dose received in the first week after the injury has already been considered in various works [1, 4]. However, they deal with the dependence of only some averaged characteristics without taking into account uncertainty of results of lesion by dose of ±2 Gy. This uncertainty is due to natural biological variability. In isolated use of the lymphocyte test it allows only a very rough estimation of radiation injury severity at the level of statistically averaged values. The influence of radiation intensity (dose rate) was not taken into account in the aforementioned studies. However, the distribution of dose over time can affect the effects of exposure. In this paper we tried to establish the influence of dose rate on the prognostic assessment of acute radiation disease severity by the lymphocyte test, to identify the best correlations and to propose medical triage criteria in order to determine the routing of medical evacuation.

The aim of the study was to validate the lymphocyte test method for predicting the severity of acute radiation injury with assessment of the effect of dose rate.

Materials and methods of the study. The data for the analysis were taken from the registry of acute radiation injuries in humans of the State Research Center — A.I. Burnazyan Federal Medical Biophysical Center, Federal Medical and Biological Agency of Russia. Correlational analysis of laboratory data on the concentration of lymphocytes in the peripheral blood of the injured from the third day to the sixth day after exposure was used as the method of study. In previous studies we found that in this period the correlation coefficient between lymphocyte concentration and radiation dose was the highest. As a parameter of dose rate in the 1st and 2nd groups we took the "average dose rate during irradiation" (Gy/h) used for analysis in the works [10-13].

The study included clinical and dosimetric data of patients (n=65) involved in the Chernobyl nuclear power plant accident in 1986 and data on patients (n=19) involved in other radiation accidents with gamma-neutron radiation in the former USSR. Only cases with known duration of exposure and low lymphocyte concentration in peripheral blood in the first 24 hours after exposure — less than 1.0×10^9/l — were selected from the registry. The data for the analysis are given in Table 1.

Depending on the conditions of irradiation, the affected patients were divided into 3 groups:

— Group 1 (n=32) — Chernobyl accident victims with dose rate less than 2 Gy/h;

— Group 2 (n=33) — Chernobyl accident victims with a dose rate of more than 2 Gy/h;

— Group 3 (n=19) — victims of radiation incidents with a neutron component of radiation in the former USSR — in such incidents high-intensity irradiation of victims occurs in the first seconds.

Results of the study and their analysis. We revealed a correlation between the average lymphocyte count in peripheral blood samples taken from day 3 to 6 after exposure

Таблица 1/ Table No.1

Клинико-дозиметрические и лабораторные данные пораженных в радиационных авариях
Clinical, dosimetric and laboratory data of people affected in radiation accidents

УКН* UKN code*	Степень тяжести ОЛБ ARS severity	D, Гр D, Gy	Количество лимфоцитов, абс.** Lymphocytes count, abs**	УКН UKN code	Степень тяжести ОЛБ ARS severity	D, Гр D, Gy	Количество лимфоцитов, абс. Lymphocytes count, abs.	УКН UKN code	Степень тяжести ОЛБ ARS severity	D, Гр D, Gy	Количество лимфоцитов, абс. Lymphocytes count, abs.
Пораженные при аварии на ЧАЭС / Injured in the Chernobyl accident, 1986											
1001	IV	7,5	0,08	1025	IV	6,0	0,27	1053	II	2,8	0,81
1002	IV	10,0	0,14	1026	IV	14,0	0,10	1054	II	3,6	0,68
1003	IV	10,0	0,04	1027	III	9,3	0,24	1055	II	5,3	0,81
1004	IV	13,2	3,25	1028	IV	7,3	0,26	1056	II	3,6	0,57
1005	III	5,2	0,24	1029	IV	9,8	0,22	1057	II	3,0	0,57
1006	IV	8,5	0,30	1030	III	6,4	0,39	1058	II	3,0	0,57
1007	III	5,5	0,35	1031	III	7,7	0,35	1059	II	5,8	0,42
1008	IV	9,4	0,22	1032	II	4,2	0,59	1060	III	6,1	0,58
1009	IV	10,7	0,12	1033	II	3,9	0,39	1061	II	4,4	0,58
1010	IV	12,4	0,03	1034	III	6,7	0,33	1062	III	7,0	0,45
1011	III	6,3	0,25	1035	II	4,0	0,88	1063	I	1,1	0,48
1012	IV	10,4	0,14	1037	II	2,8	0,75	1064	I	1,0	0,91
1013	III	6,3	0,24	1039	II	4,3	0,57	1065	II	3,1	0,78
1014	IV	12,2	0,11	1040	I	1,7	0,89	1066	II	1,0	0,76
1015	IV	10,0	0,14	1041	II	3,1	0,66	1067	II	2,6	0,39
1016	IV	11,3	0,11	1042	III	6,3	0,45	1068	III	4,6	0,62
1023	IV	15,2	0,05	1043	II	4,7	0,58	1070	II	1,2	0,62
1018	II	2,7	0,34	1044	I	3,7	0,83	1071	III	5,4	0,53
1019	III	4,6	0,26	1047	III	3,2	0,53	1072	III	3,6	0,67
1020	IV	13,8	0,13	1048	I	2,0	1,62	1051	II	1,8	0,51
1021	III	4,7	0,34	1049	II	2,1	0,55	1103	I	1,9	0,84
1022	III	7,1	0,24	1050	II	3,3	0,40	1090	I	1,2	0,42
1024	IV	2,3	0,31	1052	III	4,3	0,35	1140	I	0,3	1,65
1135	I	0,3	1,87	1091	I	1,2	1,41	1105	I	1,5	0,79
1083	I	1,9	0,76	1092	II	2,7	0,63	1106	I	2,3	0,66
1084	I	1,1	1,28	1094	III	3,6	0,47	1107	I	0,7	4,0
1085	II	3,3	0,55	1095	II	2,2	0,58	1108	II	2,3	0,67
1087	III	3,5	0,95	1096	II	3,7	0,47	1115	I	0,7	2,53
1088	II	2,7	2,29	1097	I	1,0	4,87	1121	I	0,8	1,36
1089	II	1,7	2,36	1098	I	2,0	3,22	1123	II	4,3	0,81
1085	II	3,3	0,55	1099	II	5,6	0,97	1129	II	4,0	0,60
1087	III	3,5	0,95	1100	I	2,6	0,94	1131	I	3,7	0,77
1088	II	2,7	2,29	1101	II	3,2	0,68	–	–	–	–
1089	II	1,7	2,36	1102	I	1,2	3,63	–	–	–	–
Пораженные при инцидентах с облучением гамма-нейтронным излучением /Victims of gamma-neutron irradiation incidents											
3009	III	5,4	0,07	3010	I	0,9	1,00	3047	IV	14,0	0,10
3031	IV	10,0	0,07	3043	I	3,0	0,50	3073	III	5,0	0,14
3037	II	3,7	0,28	3045	I	5,5	0,55	3079	I	2,1	1,07
3108	I	2,4	0,74	3008	III	3,8	0,27	3030	II	3,6	0,35
3032	IV	>10, 0	0,03	3042	III	4,1	0,27	3036	II	3,3	0,49
3046	IV	7,4	0,08	3025	II	2,5	0,34	3071	I	3,7	0,66
3011	I	0,5	0,99	–	–	–	–	–	–	–	–

* УКН – Уникальный код пациента из базы данных по острым лучевым поражениям человека ФМБЦ им. А.И.Бурназяна / Unique code of the patient from the database of acute radiation lesions of A.I. Burnazyan Federal Medical and Biological Center

** Среднее значение количества лимфоцитов в периферической крови в период с третьих по 6-е сутки после облучения / Average peripheral blood lymphocyte count from 3 to 6 days after irradiation

and the radiation dose only for groups 1 and 3 under study (Figure). There was greater variability in the data for Group 2 (with low dose rate ≤2 Gy), and in this case we cannot demonstrate any clear dependence. The data of the 2nd group, in particular, are satisfactorily approximated by the following logarithmic function:

$$D = -4.59 \ln (C_{lymph}) + 2.01 \quad (1),$$

where D — dose, Gy; C_{lymph} — average value of lymphocyte count in peripheral blood for the period from the third to the sixth day after irradiation ($\times 10^9$/l).

To understand the uncertainty in assessing the dose and severity of radiation injury by lymphocyte concentration, all data were grouped (Table 2).

Comparison of the data in the figure and in Table 2 shows the effect of the dose rate on the prognosis of the radiation injury severity. It can be seen that in the 1st group the radiation dose received was lower than in the 2nd group with the same lymphocyte counts. Accordingly, the probability of the severity of the developing acute radiation disease in Group 2 was more high.

Таблица 2/Table No. 2

Оценка неопределенности прогноза полученной дозы и степени тяжести лучевого поражения по концентрации лимфоцитов в периферической крови в 1-й – 3-й группах

Assessment of the uncertainty in the prediction of the received dose (Gy) and the severity of radiation injury from the concentration of lymphocytes in the peripheral blood in groups 1 to 3

Концентрация лимфоцитов (10^9/л) Lymphocytes concentration (10^9/l)	Усредненная оценка дозы облучения (Гр) / диапазон дозы облучения / прогнозная оценка степени тяжести ОЛБ Averaged estimate of radiation dose (Gy) / range of radiation dose / predictive estimate of ALS severity		
	1-я группа – пораженные при аварии на ЧАЭС с мощностью дозы менее 2 Гр/ч Group 1 – affected by the Chernobyl accident with a dose rate of less than 2 Gy/h	2-я группа – пораженные при аварии на ЧАЭС с мощностью дозы более 2 Гр/ч Group 2 – affected by the Chernobyl accident with a dose rate of more than 2 Gy/h	3-я группа – пораженные гамма-нейтронным излучением Group 3 – victims of gamma-neutron radiation
Менее 0,2	–	IV	III-IV
0,3	–	7,5 (6,0-9,0) IV	3,9 (2,7-5,1) II-III
0,4	4,1 (2,1-6,0) II-III	6,2 (4,7-7,7) III-IV	3,3 (2,1-4,5) II-III
0,5	3,6 (1,0-5,5) I-III	5,2 (3,7-6,7) II-IV	3,0 (1,8-4,2) I-III
0,6	3,1 (1,0-5,0) I-III	4,4 (2,9-5,9) II-III	2,7 (1,5-3,9) I-II
0,8	2,6 (1,0-4,5) I-II	3,0 (2,0-4,5) II-III	2,2 (1,0-3,4) I-II
1,0	2,1 (0,7-4,0) до II 2,1 (0,7-4,0) to II	до II up to II	1,8 (0,6-3,0) до II 1,8 (0,6-3,0) to II

At a lymphocyte concentration of 0.4-0.8×10^9/l at high dose rate (P_0>2 Gy/h), its assessment by the lymphocyte test is higher than at low dose rate (P_0≤2 Gy/h), by an average of 40.0%. At lymphocyte levels above 0.8×10^9/l, dose prognosis becomes more uncertain in all three groups. When peripheral blood lymphocyte counts fall to less than 0.4×10^9/L, the average dose estimate is more than 4.0 Gy for Groups 1 and 2. This corresponds to a severe or extremely severe degree of acute radiation sickness. Thus, the most accurate prognosis according to the lymphocyte test is possible in the range of lymphocyte concentrations of 0.4-0.8×10^9/l.

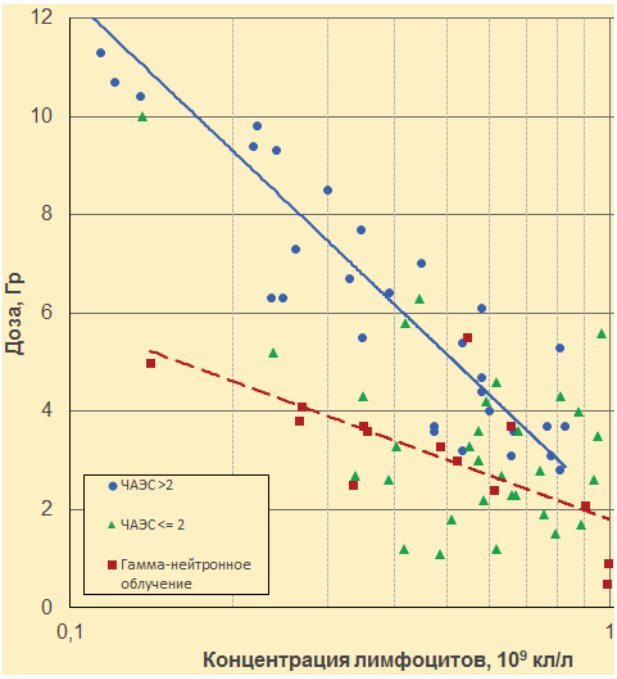

Рисунок. Зависимость среднего значения количества лимфоцитов в периферической крови в период с третьих по 6-е сутки после облучения от накопленной дозы облучения. Обозначения: треугольник зеленого цвета – данные 1-й группы; сплошная синяя линия – линия тренда по данным 2-й группы; пунктирная красная линия – линия тренда по данным 3-й группы

Figure. Dependence of the average number of lymphocyte count in the peripheral blood on the accumulated dose of irradiation from 3 to 6 days after irradiation. Note: green triangles — data of the 1st group; solid blue line — trend line in the data of the 2nd group; dashed red line — trend line in the data of the 3rd group

Independent estimates for former radiation accident victims, made without regard to radiation intensity, give an average dose estimate 30.0% lower than for "Chernobyl" patients irradiated at low dose rates (P_0≤2 Gy/h).

The predicted degree of severity of radiation injuries with the presence of neutron component of radiation — with lymphocyte concentrations less than 1.0×10^9/l — is lower compared to the predictions based on the data on the Chernobyl victims. Moreover, the difference in dose estimates progressively increases with decreasing lymphocyte counts. This result contradicts the conclusions of R.E.Goans, E.C.Holloway, M.E.Berger, R.C.Ricks, whose work declares that the lymphocyte test is insensitive to the gamma-neutron radiation spectrum [1].

Thus, it is necessary to assess the severity of radiation injuries according to the lymphocytic test, taking into account the intensity of irradiation. In this case the following typical medical and tactical solution is proposed.

The patients with lymphopenia in the range of 0.4-0.8×10^9/l who received acute high-intensity irradiation are the first to be evacuated from Level 1 – Level 2 medical treatment facilities to Level 3 medical treatment facilities. These patients constitute a difficult, but potentially curable group of medium and severe patients. In this group, provided rapid initiation of pathogenetic therapy, survival is possible.

The second line is to evacuate those affected with lymphopenia in the range above 0.8-1.0×10^9/l. Within these values, the dose prognosis becomes more uncertain. Therefore, dose estimates and prognosis should be reviewed over several days in order to identify patients with moderate severity of acute radiation sickness in a timely manner.

Those affected with a lymphocyte count above 1.0×10^9/l are classified as easily affected. Their treatment does not require the use of specialized methods and therefore they can be left in level 2 medical institutions.

Those affected with lymphopenia less than 0.4×10^9/l — irrespective of the conditions of exposure — are evacuated to Level 3 medical treatment organizations in the second line. When there is a mass admission of patients affected by ionizing radiation and under conditions of a heavy workload

of medical treatment organizations on the evacuation route such patients are not evacuated. They are given symptomatic care, since their survival is unlikely, even if pathogenetic therapy is started in time.

Conclusion

Uncertainty in dose estimation for conditions of irradiation with high dose rate (P_0>2 Gy/h) can reach (±1.0-1.5) Gy. It means that in the first approximation the lymphocyte test is of practical prognostic value in the range of lymphocyte concentrations from 0.4-0.8×10^9/l at high dose rate. When lymphocyte concentration is less than 0.4×10^9/l, acute radiation syndrome of extremely grave degree is predicted with high confidence. At low dose rates (P_0≤2 Gy/h) the prognosis is highly uncertain in the range of lymphocyte concentrations above 0.6×10^9/l. In such cases, the estimated dose is 1-4 Gy, which corresponds to mild to moderate severity of acute radiation sickness.

The identified dependencies can be used for medical triage of the injured in the advanced stages of medical evacuation, in order to adjust their routing to specialized medical centers. Thus, the following criteria should be used for mass loading of medical treatment facilities in the accident zone:

— when peripheral blood lymphocyte concentration is less than 0.4×10^9/l under any exposure conditions, acute radiation sickness of severe and extremely severe degree is predicted; such patients can be referred to the "agonizing" group;

— in the range of lymphocyte concentrations 0.4-0.8×10^9/l the assessment of dose and severity of acute radiation disease under high dose rate conditions is most accurate. These patients are referred to the category of moderate severity, whose treatment should be started as soon as possible in a specialized hospital — a level 3 medical treatment organization;

— peripheral blood lymphocyte concentrations in the range of 0.8-1.0×10^9/l predicts mild acute radiation sickness — those affected may be temporarily left for treatment in a Level 1 or Level 2 medical facility with daily adjustments of clinical and laboratory data and prognosis;

— If peripheral blood lymphocyte concentration is higher than 1.0×10^9/l, acute radiation sickness stage I is predicted.

This paper is the first to analyze the dependence of lymphocyte concentration on accumulated dose for cases with gamma-neutron irradiation compared to similar data on those affected by the Chernobyl accident. With the same values of lymphocytes, the predicted severity of acute radiation sickness is lower.

The lymphocyte test is appropriate when information about individual exposure conditions and the results of dose assessment by other methods of biological dosimetry are available.

It should be noted that in the victims of radiation accidents with hypovolemia, against the background of infusion and transfusion therapy, overestimation of the informativeness of some hematological parameters, including the number of lymphocytes, is possible. Therefore, when making a preliminary diagnosis, relying only on the lymphocyte test without taking this factor into account can lead to overdiagnosis. These peculiarities require dynamic control of laboratory parameters.

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