The results are thus evidence that splenic lymphocytes from mice of different ages give different cytolytic effects against cells cytogenetically modified by streptolysin-O. A weaker cytolytic effect was exhibited by lymphocytes of old animals, and in our opinion this can be attributed to the long preservation of a high level of cells with cytogenetic disturbances induced in man and mice by infectious factors [3, 5]. At the same time, our results show that even in cell cultures obtained from mice of different ages, the spontaneous and induced levels of cytogenetically aberrant cells differ, evidently due to ontogenetic differences in the control of genetic homeostasis at the subcellular level and, in particular, differences perhaps in activity of the DNA-repair systems.

Splenic lymphocytes, it will be noted, eliminate only aneuploid cells and do not change the number of cells with structural chromosomal disturbances. Structural chromosomal disturbances evidently do not change the antigenic structure of the cell membrane and these cells cannot be "recognized" and eliminated by effector cells.

Thus, a considerable increase in the number of cytogenetic aberrations is observed in cultures of kidney cells, obtained from mice of different ages, and "infected" with streptolysin-O. On the addition of syngeneic splenic lymphocytes from newborn and middle-aged animals to these cultures, the number of aneuploid cells present in them is significantly reduced. Splenic lymphocytes of old mice under similar conditions did not cause a significant decrease in the number of cells with cytogenetic disturbances. No antimutagenic cytolytic activity likewise was observed in lymphocytes obtained from nonsyngeneic mice.

LITERATURE CITED

1. O. G. Andzhaparidze and V. I. Gavrilov, Tissue Culture in Virologic Research [in Russian], Moscow (1962).
2. F. M. Burnet, Cellular Immunology [Russian translation], Moscow (1971).
3. N. N. Il'inskikh, Tsitologiya, No. 5, 564 (1981).
4. N. N. Il'inskikh, Tsitologiya, No. 6, 724 (1982).
5. N. N. Il'inskikh, E. F. Bocharov, and I. N. Il'inskikh, Infectious Mutagenesis [in Russian], Novosibirsk (1984).
6. Yu. Ya. Kerkis and S. I. Radzhabli, Tsitologiya, No. 2, 282 (1966).
7. N. P. Kuleshov, Tsitologiya, No. 11, 1368 (1972).
8. D. K. Novikov and V. I. Novikova, Cellular Methods of Immunodiagnosis [in Russian], Minsk (1979).
9. O. A. Syseova, Effect of Ionizing Radiation on Heredity [in Russian], Moscow (1966), pp. 78-84.

COMPARATIVE ABILITY OF EARLY AND LATE CFU-S TO RECOVER AFTER SUBLETHAL RADIATION DAMAGE

T. V. Todriya

UDC 616-001.28-092.9-07:616.411-003.9

KEY WORDS: intracellular repair after radiation damage; fractional irradiation; exogenous splenic colonies; early and late CFU-S

High ability to repair after sublethal radiation damage is a property of splenic colony-forming units (CFU-S). This property is possessed by CFU-S forming colonies both in an endogenous test and on transplantation of bone marrow into an irradiated recipient [10]. We know that CFU-S constitute a heterogeneous class of cells, which produce early ($CFU\text{-}S_{7\ days}$) and late ($CFU\text{-}S_{12\ days}$) colonies in the spleen of irradiated recipients [8, 9]. These two subpopulations of CFU-S differ not only in the time of formation of macroscopically visible colonies, but also with respect to certain other characteristics: their self-maintaining ability [3-7, 11], their response to cycle-specific agents [4-7], position in the cell cy-

Laboratory of Developmental Biology, Tbilisi University.

(Presented by Academician of the Academy of Medical Sciences of the USSR M. M. Khananashvili.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 105, No. 5, pp. 597-599, May, 1988. Original article submitted May 18, 1987.

TABLE 1. Time Course of Exogenous Colony Formation by Bone Marrow Cells Irradiated with a Single or with Fractional Doses (M ± m)

Type of irradiation	Time after irradiation and injection of bone marrow, days						
	6	7	8	9	11	12	13
Single dose	0,25±0,23	0,71±0,51	4,37±0,69	5,87±1,13	8,12±1,01	10,00±0,97	7,75±1,18
Fractional doses	0,29±0,15	6,14±1,47*	10,13±1,56*	16,00±1,50*	9,14±1,63	11,10±1,57	10,80±1,39
RI	1,16	8,65	2,32	2,73	1,13	1,11	1,39

Legend. Data for experiment No. 1 are given. Here and in Table 2: RI estimated as the ratio of the number of colonies formed in groups with fractional irradiation to the number in groups with irradiation in a single dose; asterisk indicates increase in number of colonies is significant compared with that in groups with irradiation in a single dose.

cle [3], and composition of histological types of colonies [8]. It has been suggested that CFU-$S_{12\ days}$ occupy a higher position than CFU-$S_{7\ days}$ in the histogenetic order of hematopoietic stem cells, and for that reason CFU-$S_{12\ days}$ are precursors of CFU-$S_{7\ days}$.

The aim of this investigation was to compare the ability of the earlier and later stem cells to undergo intracellular repair after radiation damage.

EXPERIMENTAL METHOD

Experiments were carried out on (CBA × C57BL)F_1 mice of both sexes. Ability to undergo intracellular repair, defined as the repair index (RI), was studied by fractional irradiation of bone marrow [2]. The recipient mice were irradiated with a total dose of 10.5 Gy on the IPK ^{137}Cs gamma-source with a dose rate of 0.20 Gy/min, and bone marrow cells for injection ($5 \cdot 10^6$ per mouse) received either a single dose of 6 Gy for the same dose divided into two equal halves (each 3 Gy) separated by an interval of 5 h [2]. Colonies were counted in the spleens from the 6th through the 13th days or only on the 8th and 11th days after injection of the bone marrow cell suspension into the mice. In each case seven to nine recipients were used for counting the colonies.

EXPERIMENTAL RESULTS

In experiment No. 1 (Table 1, Fig. 1) the ability of irradiated bone marrow, given in one injection or in fractions, to form colonies was investigated over a period of time: from the 6th through the 13th days. By arranging the investigation in this way, it was possible on the one hand to assess the ability of cells surviving after a single irradiation in a dose of 6 Gy to proliferate and, on the other hand, to damage during the interval of 5 h between the first and second doses of irradiation, i.e., to detect recovery leading to an increase in the survival rate of the cells. In the next experiments (Table 2) ability to undergo intracellular repair only of 8- and 11-day CFU-S was compared.

TABLE 2. RI and Number of 8-11-Day CFU-S Formed by Bone Marrow Cells Irradiated Once or Fractionally (M ± m)

No. of experiment	Type of irradiation	Time after irradiation and injection of bone marrow, days	
		8	11
2	Single dose	7,83±1,14	8,50±1,20
	Fractional doses	15,33±1,85*	10,63±0,89
	RI	1,98	1,25
3	Single dose	4,86±0,83	8,4±1,62
	Fractional doses	7,86±1,81	9,8±1,67
	RI	1,62	1,17
4	Single dose	4,0±0,79	10,57±1,00
	Fractional doses	11,75±2,93*	12,50±1,45
	RI	2,94	1,18

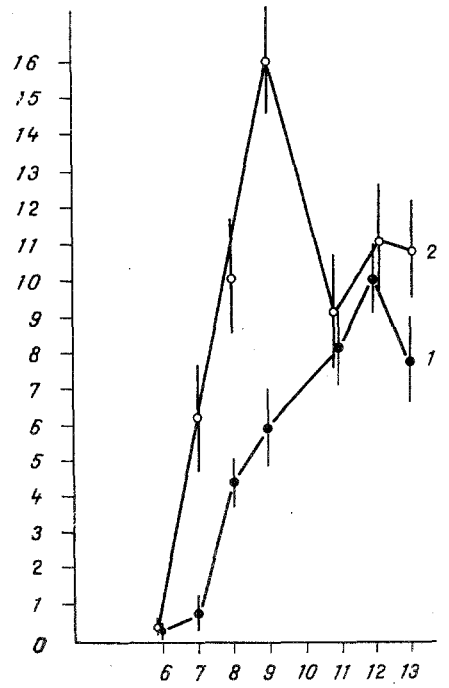

Fig. 1. Characteristic changes in number of colonies formed by bone marrow cells irradiated in a single dose and with fractional doses. Abscissa, time after irradiation and injection of bone marrow cells (in days); ordinate, number of exogenous colonies. 1) CFU-S formed by bone marrow irradiated in a single dose; 2) CFU-S formed by bone marrow irradiated fractionally.

As a result of irradiation of the transplanted cells in a single dose (Table 1, Fig. 1) fewer than one colony per group of mice was formed on the 6th and 7th days. The number of colonies detected after the 7th day increased gradually to reach a plateau by the 11th day; differences between the 11th, 12th, and 13th days were not significant. The number of CFU-$S_{8\ days}$ was only half the number of CFU-$S_{11\ days}$, evidence of more severe damage to late CFU-S, which belong to the rapidly proliferating subclass. The average increase in the number of colonies from the 8th through the 11th day was 1.25 colony per day. These results can be attributed either to the greater radioresistance of the CFU-$S_{11\ days}$ than of the CFU-$S_{7\ days}$ or to delay of proliferation of the progenies of the irradiated CFU-$S_{7\ days}$, or to the greater ability of CFU-$S_{11\ days}$ to undergo repair after potentially lethal damage.

Both after fractional irradiation of the donors' cells (Table 1, Fig. 1) and after irradiation in a single dose, fewer than one colony per group of mice was formed on the 6th day. Starting with the 7th day a sharp increase was observed in the number of colonies in the spleen of the irradiated recipients, to reach a maximum by the 9th day, when the number of colonies was about three times greater than after irradiation in a single dose. The increase in the number of colonies after fractional irradiation from the 6th through the 9th days was five daily. Meanwhile the number of colonies after 11-13 days in the case of fractional irradiation did not exceed that after irradiation in a single dose. Reduction of the number of colonies after the 9th day was probably connected with release of mature cells from the colonies into the circulation [8]. In the case of fractional irradiation the number of CFU-$S_{8\ days}$ was 230% of that after irradiation in a single dose.

After fractional irradiation an increased rate of survival of the CFU-$S_{8-9\ days}$ was thus observed, and this was confirmed by the values of RI: 2.32 and 2.73, respectively. On average for the results of four experiments (Fig. 1), RI for CFU-$S_{8\ days}$ was 2.22 ± 0.28, in agreement with data obtained previously [2, 10]. By contrast with the CFU-$S_{8\ days}$, the earlier procursors — CFU-$S_{11\ days}$ — were incapable of intracellular repair. On average for the results of four experiments (Tables 1 and 2) RI for CFU-Ss_{11} days was 1.18 ± 0.025.

The inability of CFU-$S_{11\ days}$ to undergo intracellular repair may be attributed to their greater radioresistance. It has in fact been shown that CFU-$S_{7\ days}$ from bone marrow of adult mice have greater radiosensitivity than CFU-S_{11} days [11]. Meanwhile, the quantitative assessment of recovery after sublethal radiation damage used in the present investigation (RI) may not completely characterize the ability of the early CFU-S to repair sublethals. It was shown previously [10] by the use of radiobiological tests and, in particular, by determination of D_0 and of the extrapolation number, that CFU-$S_{10\ days}$ can repair sublethal radiation lesions. However, with respect to proliferative activity and ability to maintain themselves, CFU-$S_{10\ days}$ are closer to CFU-$S_{8-9\ days}$ than to CFU-$S_{12-13\ days}$ [7].

The results on the whole are evidence that earlier CFU-S possess significantly less ability to repair after sublethal radiation damage than more mature CFU-S. This raises the question of whether ability to undergo intracellular repair appears only after the early precursors have reached a certain level of cellular differentiation. In particular, this may be linked with the appearance of sensitivity to thymic factors in the CFU-S, for it has been shown that CFU-$S_{8\ days}$ in thymectomized animals lose their ability to undergo intracellular repair [2].

The authors is grateful to Candidate of Biological Sciences O. I. Gan for help and valuable advice in the course of this research.

LITERATURE CITED

1. O. I. Gan, "Hematopoietic precursor cells in mouse liver culture," Author's Abstract of dissertation for the Degree of Candidate of Biological Sciences, Moscow (1984).
2. T. V. Todriya, Byull. Éksp. Biol. Med., No. 11, 584 (1978).
3. P. Bains and J. W. M. Visser, Exp. Hematol., 11, 701 (1983).
4. S. Hellman, L. E. Botnik, E. C. Hannon, and R. M. Vigneulle, Proc. Natl. Acad. Sci. USA, 75, 490 (1978).
5. G. S. Hodgson and T. R. Bradley, Nature, 281, 381 (1979).
6. G. S. Hodgson, T. R. Bradley, and J. M. Radley, Exp. Hetamol., 10, 26 (1982).
7. G. S. Hodgson and T. R. Bradley, Exp. Hematol., 12, 683 (1984).
8. M. C. Magli, N. N. Iscove, and N. Odartchenko, Nature, 295, 527 (1982).
9. G. V. Priestley and N. S. Wolf, Exp. Hematol., 13, 733 (1985).
10. J. E. Till and E. A. McCulloch, Radiat. Res., 18, 95 (1963).
11. R. G. Worton, E. A. McCulloch, and J. E. Till, J. Exp. Med., 130, 91 (1969).

REPAIR PROCESSES IN NERVE TISSUE AFTER BRAIN TRANSPLANTATION IN YOUNG RABBITS

I. I. Malyshev

UDC 616.831-089.843-092.9-07: 616.831-003.9

KEY WORDS: brain; transplantation; regeneration; CNS

Experimental brain transplantation has been widely undertaken in recent years. Besides problems of survival of the graft, the study of the effect of brain transplantation on regeneration of nerve tissue of the CNS has also aroused interest.

The aim of this investigation was to study repair processes in the nerve tissue of the brain after transplantation of large fragments of neocortex in young rabbits.

EXPERIMENTAL METHOD

Experiments were carried out on 168 rabbits of the same litter or obtained from different parents, and aged 2-4 days; transplantation of neocortical fragments into the parietal region of the right cerebral hemisphere was performed on the animals by the method described in [2]. The duration of the experiments was from 1 to 45 days. A particular feature of them was the nonobservance of the special conditions [3] that facilitate survival of the graft. The animals were killed by decapitation and the brain was embedded whole in paraffin wax. Histotopographical sections were stained with hematoxylin and eoxin, and by Van Gieson's, Nissl's, and Spielmeyer's methods; some sections were impregnated with silver by the Cajal and Bielschowsky–Gros method.

EXPERIMENTAL RESULTS

On the 1st day edematous, dystrophic, and necrobiotic changes predominated in the graft and in the recipient's brain. On the 3rd day, active proliferation of neurogliocytes de-

Department of Morbid Anatomy, No. 12 Hospital, Gor'kii. (Presented by Academician of the Academy of Medical Sciences of the USSR D. S. Sarkisov.) Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 105, No. 5, pp. 599-601, May, 1988. Original article submitted April 17, 1987.

 0007-4888/88/0005-0718$12.50