

Use of Human Lymphocytes for Radiation Risk Assessment

T. Größer, P. Hessel and S. Ritter (GSI, Darmstadt), E. Nasonova (JINR, Dubna, Russia),
E. Gudowska-Novak (Institute of Physics, Krakow, Poland)

Chromosome aberrations are regarded as the most sensitive indicator of radiation-induced genetic alterations. In particular the frequency of aberrations in peripheral blood lymphocytes (PBLs) is used to estimate the dose to which an individual has been exposed. Moreover, aberrations in PBLs are considered as a promising tool in predictive assay research aiming to develop a test for individual normal tissue tolerance.

According to the standard protocol cytogenetic damage is analysed in first generation metaphases collected at one sampling interval. In the case of PBLs, metaphases are routinely harvested 48h after initiation of the culture. Yet, there is increasing evidence that radiation-induced cell cycle delays affect the aberration yield. In particular after high LET exposure a drastic increase in chromosomal damage with sampling time has been observed, i.e. heavily damaged cells have been found to enter mitosis later than slightly damaged cells. Consequently, depending on the subpopulation of cells which reaches mitosis at that particular sampling time chosen for the analysis, the delivered dose can be substantially under- or overestimated (e.g. [1,2]). To contribute to this issue in first experiments the cell cycle progression of PBLs obtained from 5 healthy donors has been measured. Exemplarily in figure 1 the fraction of mitotic cells found 48h after initiation of the culture is plotted indicating a pronounced intra- and inter-donor variability. Interestingly, the data obtained for donor C do not show a significant intra-donor variation.

Further, more detailed investigations of the proliferation of PBLs revealed a marked difference in the cell cycle transition time of untreated control cells, i.e. PBLs needed up to 96h to reach the first mitosis after initiation of the culture (see fig. 2). Following irradiation a dose-dependent delay in the entrance time to mitosis, reflected in the shift of the maximum of the mitotic indices, was found. Also, a general depression of the mitotic activity occurred as exemplarily shown in figure 2. Again, all cell cycle studies performed up to now demonstrate a pronounced intra- and interdonor variability.

As expected (3) similar variations are found on chromosomal level. For example, following X-irradiation cells of donor A were more sensitive to aberration induction than those of donor B (fig. 3). The same trend was observed after exposure to 400 MeV u C ions. Interestingly, neither after X-irradiation nor high energy C ion exposure a significant fluctuation in the aberration yield with sampling time has been detected. In contrast, in first experiments where PBLs of donor A were exposed to high LET particles, i.e. 200 MeV/u Fe ions with a LET of 500 keV/ μ m, a dramatic increase in the frequency of lesions with time has been observed. Thus, after high LET exposure the restriction of cytogenetic studies to only one sampling time will unavoidable lead to misleading results. Moreover, intra- and interdonor variability, which effect significantly the yield of radiation-induced aberrations has to be considered.

References

- [1] Nasonova, E. et al., Phys. Med. XVII, 198-201 (2001)
- [2] Ritter, S. et al., GSI Scientific Report 2000, 154 (2001)
- [3] Virsik-Peukert, P. et al., Radiat. Res. 148, 209-215 (1997)

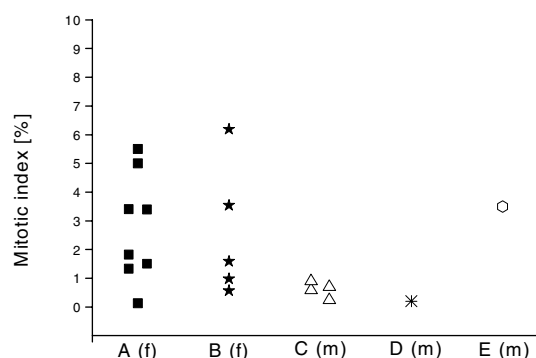


Fig. 1: Inter- and intradonor variability in the fraction of PBLs reaching mitosis by 48h. Cells have been obtained from healthy non-smoking volunteers (age: 38 to 50, f=female, m=male). Measurements were performed within a period of 3 years.

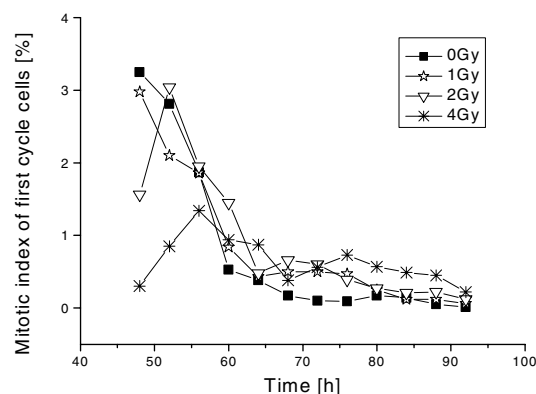


Fig. 2: Proportion of cells reaching the first post-irradiation mitosis as a function of sampling time. In this experiment PBLs of donor B have been exposed to 100 MeV/u C ions.



Fig. 3: Yields of X-ray induced dicentric chromosomes. PBLs of donor A (2 experiments) and B (1 experiment) were exposed in vitro and dicentrics were scored in first generation metaphases 48h post-irradiation. For comparison data from reference [3] are plotted.