Influence of a P53 Mutation on the Radiation Sensitivity of Mouse Zygotes

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The aim of studies under way in our laboratory is to investigate whether heterozygous mutations in genes involved in such important cellular processes as cell cycle regulation, apoptosis and DNA repair may influence the radiation sensitivity of early mammalian embryos. The embryonic stage chosen for our first investigations is the zygote (first day of gestation). This stage occurs while women cannot be aware of pregnancy. Moreover, in contradiction with a long standing dogma of teratology, various kinds of malformations were previously found in mouse fetuses from particular (wild-type) strains after X-irradiation at the zygote stage. Our studies mainly concentrate on external congenital anomalies, cytokine secretion in the amniotic fluid and chromosomal instability. Measuring cytokines in the amniotic will enable us to determine whether developmental abnormalities are accompanied by changes in the levels of particular cytokines, as suggested by the few available data. On the other hand, chromosome instability has been recently reported in mouse fetuses from different strains, after x-irradiation at the zygote stage. The gene currently under study is P53, the "genome guardian". The P53 mutation was introduced in the CF1 strain, whose wild-type zygotes had been previously shown by us to be sensitive to radiation induction of congenital anomalies (Jacquet et al., Mutation Res., 332, 73-87, 1995). P53 (+/+ x -/-) or P53 (+/+ x +/-) matings were performed from 7.30 till 9.30 am and the females showing a vaginal plug were x-irradiated with either 0.2 or 0.4 Gy 2 h after presumed fertilization. A number of them were sacrificed on day 8 of gestation, their gastrula stage embryos were collected and their embryonic parts were cultured for 7 h in the presence of colchicine. The cells were then fixed and cytogenetically analyzed. Other females were sacrificed on day 19 of gestation. Pre- and post-implantation losses were recorded, amniotic fluid surrounding the fetuses was collected for cytokine analysis and the living fetuses were weighed and examined under the stereomicroscope for the presence of congenital anomalies. When needed (+/+ x +/- matings), the tails of the fetuses were collected for genotype analysis. So far, and although our results have still to be completed, the P53 mutation did not seem to result into the development of a chromosomal instability and/or to higher levels of congenital anomalies in irradiated embryos. (Partially funded by the research contract n° CO-90 06 2024.00 between SCK-CEN and the Federal Agency for Nuclear Control).