HOW RELEVANT TO RADIATION PROTECTION IS THE ADAPTIVE RESPONSE MECHANISM?

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ABSTRACT

There is evidence that the phenomenon of adaptive response (AR) which results from a low dose exposure could modify the risk of a subsequent radiation exposure, and conceivably could even provide a net benefit rather than the putative radiation detriment at low doses. The AR has been widely observed in human and other mammalian cells exposed to low doses and low-dose rates. The phenomenon has been demonstrated at the level of one track per cell, the lowest insult a cell can receive. The AR to radiation has been shown to: (i) protect against the DNA damaging effects of radiation and many chemical carcinogens; (ii) increase the probability that improperly repaired cells will die by apoptosis, thereby reducing risk to the whole organism; (iii) suppress both spontaneous- and radiation-induced neoplastic transformation *in vitro*; and (iv) reduce life-shortening in mice that develop myeloid leukemia as a result of a radiation exposure. It remains unclear, however, if the AR will be relevant to either risk assessment or radiation protection. There is currently no evidence of AR's influence on the incidence of radiogenic cancer *in vivo* although recent data indicate that adapting doses could lead to reduced risk in animal or human populations. Currently the existing dose control and dose management programs attempt to limit or eliminate even very low exposures, without evidence that such an approach has economic and societal benefits. Indeed, if adaptation from exposure to low doses provides the same responses *in vivo* as have been shown *in vitro*, then the current approach to protection against low doses may be counterproductive. However, the demonstrated principles of the adaptive response to radiation *in vitro* will not likely influence the long held current formulation of radiation protection practices until the biological action of accumulated low doses of radiation *in vivo* and its impact on the modulation of radiation carcinogenesis are better understood.

INTRODUCTION

Current radiation risk estimates and all radiation-protection standards and practices are based on the so-called "Linear No-Threshold (LNT)" model. The LNT model assumes that risk is linearly proportional to dose, without a threshold, and thus allows the radiation protection practitioners to establish a number of assumptions about the dose-effect relationship: (a) every dose, no matter how low, carries with it some risk; (b) risk per unit dose is constant; (c) risk is additive; (d) risk can only increase with dose; and (d) biological variables are insignificant compared to dose. The radiation protection community has historically accepted the LNT model as the basis for a conservative approach to radiation protection practice. Demonstration of the adaptive response *in vivo* in animals and *in vitro* in human cells (Mitchel et al., 1997) exposed to doses between 1 and 100 mGy, however, challenges the validity of the LNT model, and has led to speculation that chronic or low doses of ionizing radiation might have a net protective effect on cancer induction (Mitchel and Trivedi, 1993).

CELLULAR RESPONSE TO RADIATION AND LEVEL OF RISK

If we consider the potential biological outcomes of a radiation exposure to a cell, there are three general possibilities, as shown in Figure 1. When DNA damage is created as a result of one or more tracks of radiation through a cell, the cell will attempt to repair that damage. If the cellular repair is successful and the DNA is restored to its original state, i.e., an error-free repair, then the cell is also restored to normal. In this case, there is no resulting consequence to the cell and hence no resulting risk.

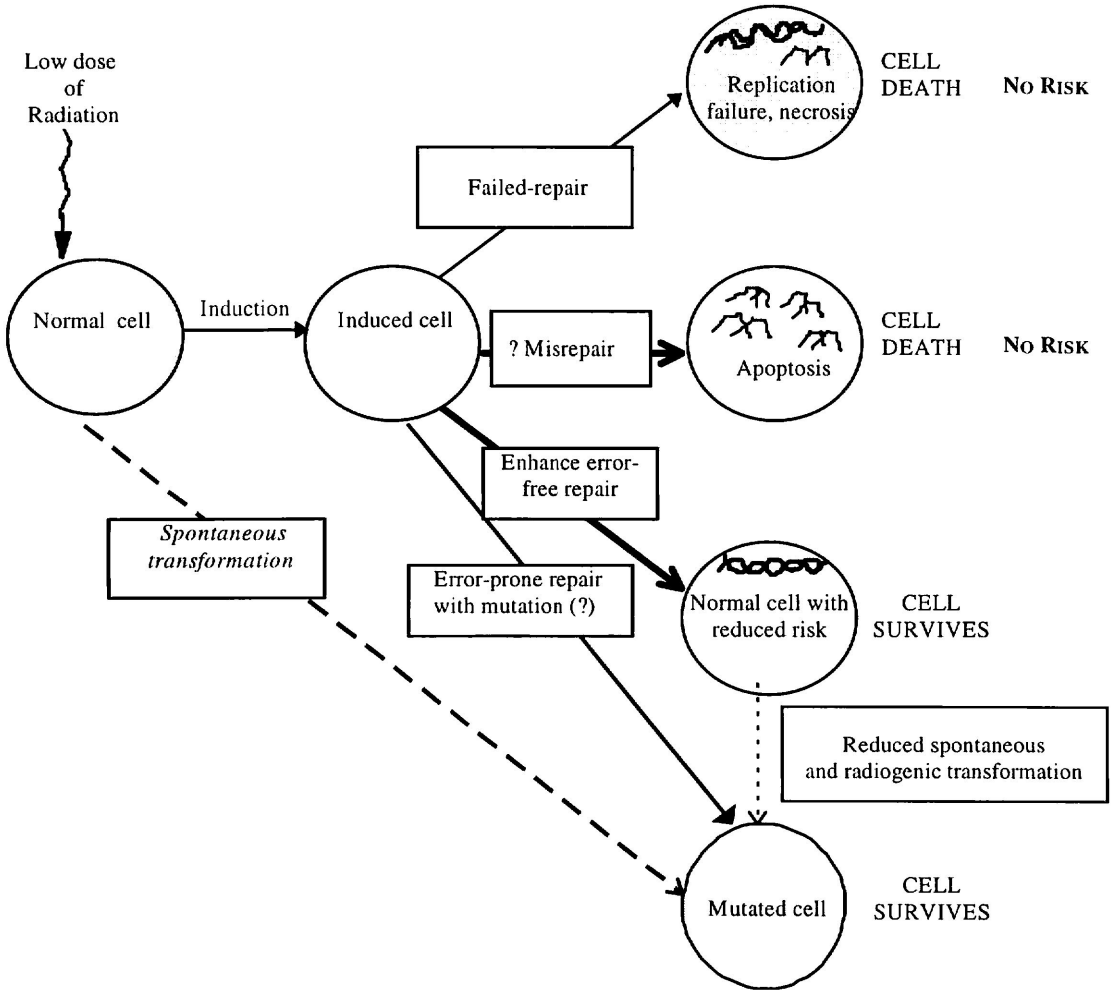

Figure 1. Potential outcomes of radiation exposure in a normal cell.

A second possibility is that the cell has DNA damage and recognizes that it cannot properly repair the damage, and as a consequence activates its genetically encoded programmed cell death process, called apoptosis. Again, in this case, no risk of carcinogenesis results since dead cells do not produce cancer. The third possible outcome of the DNA damage is repair which avoids cell death but which is either error-prone or error-free. The error-free repair system often restores DNA damage to its original state with stimulated repair capability, while the error-prone mechanism can result in mistakes that creates a mutation. At this point, the adapted cell

may still activate its apoptotic cell death program but could also simply resume dividing. While the vast majority of mutations do not create the potential for cancer, there are some that do and it is these mutations that represent the risk. Of the three possible outcomes, therefore, only one creates a risk of carcinogenesis (see Fig. 1).

It is useful to remember that the LNT model predicts that risk is influenced only by dose, and hence predicts that the relative proportions of the biological possibilities must be constant. If they were not constant, then risk would vary with their relative proportions, i.e., not as a function of dose. This, however, is precisely the situation that occurs when cells are exposed to low doses and they respond by altering the relative probabilities of the three possible outcomes described above.

Low doses of radiation also induce radioresistance in neighboring cells, where the exposed cell appears to involve in gap-junctional intercellular communication, and thus permitting the normal cells to become more radioresistant (not shown in Fig. 1) (Ishii and Watanabe, 1996). Low dose may also stimulate the immune system toward improved surveillance of malignant cells in tissues (Shu-Zheng, 1994; Xu et al., 1996). These cellular responses have the additional potential of protecting against damage to DNA in adapted cells. Since prevention of DNA damage reduces the incidence of malignant tumors, that radiation-induced protection against DNA damage from the adaptive dose may affect both spontaneous and radiogenic cancer cells from subsequent exposures.

It follows that certain probabilities need to be considered in assessing the risk of detriment in an irradiated cells or organism. At the cellular level, the probabilities of: (a) the cell being hit by an energy deposition event; (b) the hit cell responding by correct repair of radiation-induced DNA damage; (c) the hit cell failing to properly repair its DNA and experiencing, for example a malignant transformation; and (d) the hit cell being benefited by prevention of a subsequent spontaneous or radiation induced detriment, will all influence the outcome. At the organ level through intercellular communication, the probability of (a) cells comprising the organism repairing or compensating for radiation-induced structural and/or functional failure in the tissue; and (b) the multitude of hit cells in the organism influencing tissue responses that result in either detriment or benefit to organism, will further modify the net result. The interplay of these probabilities will change with the level of absorbed dose. At high doses the net probability clearly favors detriment. However, it is possible that—because of the AR—a low dose of radiation has a higher probability of preventing a cancer than causing one.

BALANCE OF EVIDENCE

Decisions about the validity of a scientific hypothesis are normally based on the balance of evidence. We consider here the experimental evidence for the AR, and examine them for and against the validity of the LNT model as it applies at low doses and dose rates.

The scientific data which indicates harm from a low dose of ionizing radiation is easy to summarize: there is no evidence. Human epidemiological studies and *in vivo* animal studies are often described as "consistent with the LNT model" yet no study actually shows statistically significant data indicating a risk of cancer in animals or persons exposed to doses less than about 100 mSv, particularly if the exposure was at a low dose-rate (Pierce et al., 1996; ADS, 1997). Most studies indicate no risk while a few indicate a risk lower than the control while ignoring the effect of ionizing radiation on the DNA damage-control system. In such studies, emphasis is placed on the relative difficulty of repairing infrequent double standard breaks (0.4/cell/cGy;

Ward, 1995), while ignoring the daily removal and control of very large numbers of other environmental and spontaneous DNA damage by the adaptive response, necrosis, apoptosis and immune system (Abelson, 1994).

Contrary to the lack of scientific valid data for increased risk associated with low dose radiation, there is a plethora of experimental evidence to support the presence of radiation-induced adaptive response in living beings (Mitchel et al., 1997). Such responses are well known in lower eukaryotes, are relatively well characterized, and appear to be evolutionarily conserved (Boreham et al., 1991). Adaptation by exposure to low radiation doses in mammalian cells and tissue, and stimulation of repair of DNA damage has been accepted by the international societies (HPS, 1996; ADS, 1997) and committees (ICRP, 1990; UNSCEAR, 1994).

Cellular responses to low doses of radiation have already been shown to reduce the spontaneous occurrence of chromosomal aberrations in human lymphocytes (Feinendegen et al., 1996), and to temporarily enhance the elimination of DNA damage incurred from higher doses of radiation and other toxic agents (Mitchel and Trivedi, 1993). Here, we have evaluated a few selected studies to address following questions (1) How probable is AR activation, as a function of dose and dose-rate? (2) What degree of protection is provided by the AR in adapted cells against potentially carcinogenic event? (3) Is the AR versatile and provide same level of protection among individuals? and (4) How long does the AR persist in a cell without further activation?

Experiments with rodent cells, using low LET radiation, showed that the lowest dose and dose rate possible (1 mGy/track/cell*) produced the same level of adaption as much higher doses given at low dose rates (Azzam et. al 1996). In human fibroblasts a low dose of 50 cGy produced a higher level of adaption when given at low dose rate (2 mGy/min) than at high dose rate (2 Gy/min), but adaption was still evident after the high dose rate exposure (Mitchel et al. 1997). These results show that there are likely to be differences in the dose and dose rate responses between different tissues.

Azzam et al. (1996) observed a 75-80% reduction in spontaneous transformation frequency in C3H 10T1/2 cells following irradiation with 1 mGy of low LET radiation, and doses of 100 mGy given at low dose rate produced the same result. Similar large reductions are also seen for human cells. These results indicate therefore that the lowest possible dose to a single cell (1 mGy) is sufficient to trigger the maximum possible AR and that further radiation dose does not influence this maximum response.

The AR at low doses has been shown to provide protection in many cellular systems, including mammalian (Tempel and Schleifer, 1995) and human (Ishii and Watanabe, 1996) embryonic cells *in vitro*. The AR has also been shown to occur *in vivo* in rabbits (Liu et al., 1992) using adapting doses between 0.3 and 1.5 Gy delivered at very low dose rates (0.1 mGy/min) and in mice (Wojcik and Tuschl, 1990) exposed to 50 mGy/day for four days at 1.25 mGy/min. The AR, resulting from a 30 cGy exposure of pregnant mice, has also been shown to protect the developing mouse embryos against the lethal and teratogenic effects of a high radiation dose subsequently delivered to the pregnant mice (Wang et al., 1998). In humans, the AR appears to present in children (Tedeshi et al., 1996) and adults (Barquinero et al., 1995); but varies among individuals (Hain et al., 1992). The human result is based only on lymphocyte

*A dose of 1 mGy represents only about one radiation track per cell nucleus for low LET radiation. A single alpha particle track (high LET) will deposit 20-30 cGy per cell nucleus.

data, and usually does not account for radiation-induced apoptosis. The maximum protective effect has generally been observed for 24-72 h after the first low-dose exposure, depending on cell type and perhaps other factors, although there are scattered evidence that the AR could be sustained over a long period (months) (Boreham et al., 1997).

Given its existence *in vitro* in human cells and *in vivo* in animals, it is reasonable to postulate that the AR occurs in humans, although the reasons for it and its overall significance are not well understood. Radiogenic cancers in humans are clearly observed only at doses ≥100 mSv and most epidemiological evidence is consistent with both no effect and a protective effect at doses below this (Little and Muirhead, 1996; Heidenreich et al., 1997). Although the cellular responses following the adapting dose are transient, the AR may provide temporary or uncertain advantages in an environment where the level and occurrences of exposures are unpredictable. Even a temporary protection against the effects of a possible subsequent exposure or spontaneous event has a benefit. If low dose exposure provides a protective effect against radiation or other carcinogens in the workplace, and additionally a net protective effect against an individual's inherent risk of spontaneous cancer, then there is a benefit from AR in an operational radiation protection program. Cucinotta et al. (1998), using mutlistage carcinogenesis models and low doses data for animals, have predicted that the adaptive response mechanism in cells could lead to a 20-30% reduction for carcinogenic risk.

Arguably, better scientific understanding of the protective effects of the AR to low-level radiation would result in a realistic assessment of the risk of low doses. There is substantial optimism that research on the molecular and cellular mechanisms of the AR phenomena can lead to an improved understanding of the influence of radiation on the process and probability of cancer. To provide more definite conclusions about the significance of the AR in the practice of radiation protection, we suggest to: (1) better determine the triggering event(s) required to activate the AR; (2) investigate the probability of activation under varying dose and dose-rate conditions; (3) examine the extent to which the protective effect can be transmitted from a cell which has gathered radiation-induced DNA damage to surrounding cells; (3) establish the effectiveness of the AR in multi-step carcinogenesis; (4) understand the transient mechanism of the response in order to prolong the decay time; and foremost (5) elucidate the functioning of the AR under *in vivo* conditions.

CONCLUSION

A lack of supporting evidence, coupled with substantial information about adaptive responses, carcinogenesis, and human radiobiology at low doses calls into question the validity of the LNT model assumptions at these doses. Experiments in cells at low doses have shown that each of the four assumptions of the LNT hypothesis, listed in Introduction, are incorrect at low doses. We have shown that (a) not every dose creates a risk and a dose of one track can reduce the risk of neoplastic transformation; (b) the risk per unit dose is not constant and a low dose exposure reduces the risk from a subsequent dose; (c) risk is not additive since the biological changes induced by one dose can influence the risk associated with a second dose; and (d) biological variables are not insignificant since they can change the outcome of an exposure by factors of two or more.

However, the demonstrated principles of the adaptive response to radiation in animals and *in vitro* human cells will not likely influence the long held current formulation of radiation protection practices until the biological action of accumulated low doses of radiation *in vivo* and its impact on the modulation of radiation carcinogenesis are better understood. More research is

needed before any benefits from low dose exposure might be included in applied radiation protection concepts.

In the interim, an alternative linear-threshold model can be used as a practical radiation protection guideline to assess risk and make decisions about radiation protection standards. While this linear-threshold model is not consistent with the observed protective effects which result from the adaptive response mechanism at low doses, the model at least removes the concept of risk at those low doses. The model offers a way to provide practical guidance at doses about 100 mSv where radiation is detrimental.

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