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Short-term exposure to high levels of natural external gamma radiation does not induce survival adaptive response

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Background: In some areas of Ramsar, a city in northern Iran, residents receive a much higher annual radiation exposure than is permitted for radiation workers. Induction of adaptive response in residents of Ramsar has been reported previously. In this study induction of such a response in short term exposure to high background levels of gamma radiation is investigated. ***Materials and Methods:*** Fifty male NMRI mice were randomly divided into four groups of 10-17 animals and 53 Wistar rats were randomly divided into five groups of 10-12 animals were studied. Animals in the 1st group were kept for 7 days in an outdoor area with normal background radiation while the 2nd, 3rd, 4th and 5th (in case of rats) groups were kept in 3 different outdoor areas with naturally elevated levels of gamma radiation. Animals were then exposed to a lethal dose of 8 Gy gamma radiation. ***Results:*** For mice, 30 days after exposure to lethal dose, the survival fraction for the control group was 40% while the 2nd, 3rd, and 4th groups had survival rates of 20%, 33.30%, and 35.20%, respectively. For rats, 30 days after exposure to the lethal dose, the survival fraction for the control group was 40% while the 2nd, 3rd, 4th and 5th groups had survival rates of 20%, 41.6%, 60.0% and 35.7%, respectively. ***Conclusion:*** Results indicate that short-term exposure to extremely high levels of natural gamma radiation (up to 196 times higher than the normal background) do not lead to induction of survival adaptive response. **Iran. J. Radiat. Res., 2012; 10(3-4): 165-170**

Keywords: *Ramsar, natural background radiation, survival adaptive response, short term exposure.*

INTRODUCTION

Ramsar a city in northern Iran is among the world's well-known inhabited areas with highest levels of natural radiation. In areas with elevated levels of natural radiation annual exposure rates are up to 260 mSv y^{-1} and average dose rates are about 10 mGy y^{-1} for a population of about 2000 residents (1-3). Due to the local geology, which includes high levels of radium in rocks, soils, and groundwater, Ramsar residents are also exposed to high levels of alpha particles in the form of ingested radium and its decay progenies as well as very high radon levels in their dwellings (over 3000 Bq m^{-3} in some cases). It has been reported that the inhabitants of these areas receive doses much higher than the current ICRP-60 occupational dose limit of 20 mSv y^{-1} (4).

Elevated levels of low dose rate natural radiation of Ramsar provide a unique opportunity to study epidemiological effects for the inhabitants. However to obtain reasonable and consistent data for 2000 residents, we need to observe them for long time to acquire considerable number of person-years for reliable statistical data. One of the low dose radiation effects is radioadaptation which has important implications for human health. Human and laboratory animal studies are indicative of occurrence of this phenomenon *in vivo*. The induction of the adaptive response in the

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lymphocytes of occupationally exposed hospital workers to X- and gamma rays [5-8] and human lymphocytes exposed to the fallout from the Chernobyl accident [9] are some sorts of these studies. *In vivo* experiments have also shown that low doses of radiation help protect against radiation-induced myeloid leukemia [10] and spontaneous cancer in mice [11].

Radioadaptive responses of the residents who lived in high background radiation areas of Ramsar has been reported previously [1]. Radioadaptaion induced by natural background radiation has been shown to be more effective than occupational exposure [12]. It has also been shown that short-term exposure to elevated levels of radon could induce an adaptive response in laboratory animals [13].

Recently Mortazavi *et al.* have shown that short-term exposure to artificially elevated levels of radon may induce an adaptive response in an animal model [13]. In this study we investigate the possible induction of oxidative stress after short-term exposure to extremely high levels of natural radiation. In addition, since there is no report on the induction of adaptive response in short term exposures to high background levels of gamma radiation, the main aim of this study is to verify if exposure of laboratory animals to extremely elevated levels of natural external gamma in high background radiation areas of Ramsar can lead to induction of survival adaptive response.

MATERIALS AND METHODS

Animals

Fifty male NMRI mice (age 3 weeks, 25-30 g) and 53 male Wistar rats (age 9 weeks, 200-250 g) were purchased from Pasteur and Razi Institute in Amol, Iran, respectively. Mice were randomly divided into four groups of 10-17 animals and rats were randomly divided into five groups of 10-12 animals. Animals were kept under a 12-hour light and 12-hour dark cycle at ambient temperature (21 ± 1 °C) with free access to food and water. All guidelines of Shiraz University of Medical Sciences (SUMS) for ethical treatment of animals were observed.

Exposure to naturally elevated levels of radiation

In the first phase of this study, animals in the 1st group were kept for 7 days in an outdoor area with normal background radiation (0.10 μSv/h) while the 2nd, 3rd, and 4th groups were kept for 7 days in Talesh Mahalleh, Ramsar with 3 different naturally elevated levels of gamma radiation of 0.64, 2.09 and 3.73 μSv/h, respectively. A calibrated RDS-110 survey meter (RADOS Technology, Finland), mounted on a tripod approximately 1 meter above the ground, was used to measure exposure rate at each location. In the second phase of the study, higher dose rates of natural radiation were investigated (in the 1st phase the highest dose rate was only 37 times greater than the normal background). Therefore, animals in the 1st group were kept for 7 days in an outdoor area with normal background radiation (0.18 μSv/h) while the 2nd, 3rd, 4th and 5th groups were kept in 4 different outdoor areas with naturally elevated levels of gamma radiation in Ramsar; i.e. 3.92, 8.47, 16.43 and 35.28 μSv/h, respectively. Thus we were able to examine the effects of dose rates as high as 196 times the background level.

Exposure to lethal dose

On day 8, all animals were exposed to 8 Gy gamma radiations emitted by a Theratron Phoenix (Theratronics, Canada) Cobalt-60 therapeutic source (dose rate 55 cGy/min) at Radiotherapy Department of Razi Hospital in Rasht, Iran.

Survival study

After irradiation rats were moved back to their cages and their survival was carefully monitored three times a day for 30 days. Kaplan-Meier survival analysis (SPSS 17.0) was used to evaluate the statistical significance of the differences of survival rates among different groups.

RESULTS

As shown in table 1, in the first phase of the study with NMRI mice, survival rate in the control group (only exposed to lethal dose) was 90% on day 24, while the survival rates for those animals pre-irradiated with low, moderate and high dose rates of natural radiation before exposure to lethal dose were 40%, 75% and 64.7%, respectively. Thirty days after exposure to lethal dose of gamma radiation, the survival fraction for the control group was 40% while the 2nd, 3rd, and 4th groups had survival rates of 20%, 33.30%, and 35.20%, respectively.

As shown in table 2, in the second phase of the study with rats, survival rate in the control group (those exposed to normal background radiation of 0.18 µSv/h) was 40% on day 25, while the survival rates for those animals exposed to higher levels of natural radiation before irradiation with lethal dose; i.e. 3.92, 8.47, 16.43 and 35.28 µSv/h, were 20%, 41.6% and 60% and 35.7%, respectively. Kaplan-Meier survival analysis could not reveal any statistical significance differences in survival rates among different groups for the 1st phase of the study (P=0.717) and the 2nd phase (P=0.362). Pair-wise comparisons also could not show any significance differences in survival rates between each two groups (i.e. survival rate of the animals kept in an area with a dose rate of 0.18 µSv/hr as compared to that of 3.93, 8.48, 16.43 or 35.28 µSv/hr). Figures 1 and 2 show the survival rates of the animals in 30 days for the 1st phase and in 25 days for the 2nd phase of the study, respectively.

DISCUSSION

In our previous cytogenetic studies, after *in vitro* irradiation with a challenge dose of gamma rays, a significant cytogenetic adaptive response was observed in the

Table 1. The pattern of exposure to high levels of natural radiation in different groups.

Group	Dose Rate ($\mu Sv\ h^{-1}$)	Survival Rate 30 Days After Exposure to lethal Dose
G1 (Normal Background)	0.10	40%
G2 (HBRA-1)	0.64	20%
G3 (HBRA-1)	2.09	33.3%
G4 (HBRA-1)	3.73	35.2%

G- Group
HBRA – High Background Radiation Area

Table 2. The pattern of exaposure to high levels of natural radiation in different groups.

Group	Dose Rate ($\mu Sv\ h^{-1}$)	Survival Rate 30 Days after Exposure to Lethal Dose
G1 (Normal Background)	0.18 µSv/hr	40%
G2 (HBRA-1)	3.93 µSv/hr	20%
G3 (HBRA-2)	8.48 µSv/hr	41.6%
G4 (HBRA-3)	16.43 µSv/hr	60%
G5 (HBRA-4)	35.28 µSv/hr	35.7%

G- Group
HBRA – High Background Radiation Area

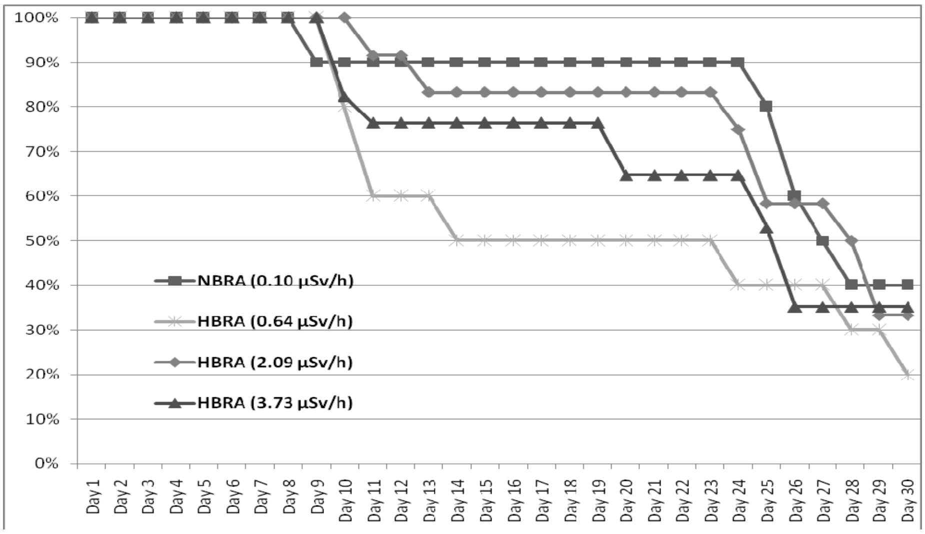

Figure 1. Survival plots of different groups within 30 days after irradiation of mice pretreated with various low doses of NHLR for 7 days.

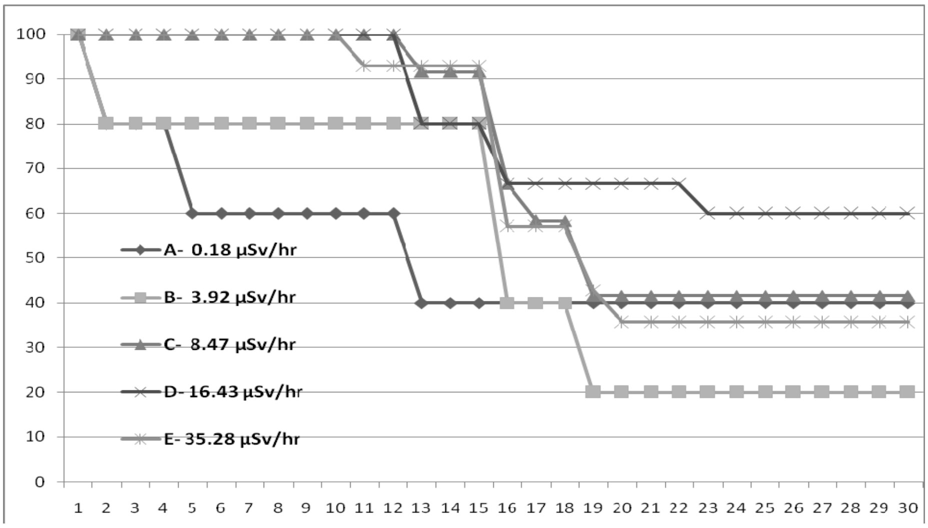

Figure 2. Survival plots of different groups within 30 days after irradiation of mice pretreated with various low doses of NHLR for 7 days.

lymphocytes of residents of the very high background radiation areas of Ramsar [1]. However, in spite of methodological differences, results obtained in both phases of this study show that high levels of natural radiation cannot induce survival adaptive response. Moreover, current findings are not consistent with the results obtained in our previous study on the induction of adaptive response in laboratory animals after short-term exposure to elevated levels of radon [13]. The discrepancies in these findings might reside on cellular response to low levels of ionizing radiation. Similar to all biological responses of cells to IR induced or exogenous stress [14], inducible cellular processes occurring in cells following exposure to low doses of ionizing radiation is mediated through DNA damage. These evidences show that radiation has a stimulating effect on a number of biological processes and can induce resistance against higher doses of ionizing radiation.

In early studies, Feinendegen *et al.* proposed that adaptive response could be induced by reactive oxygen species (ROS) [15, 16]. The ROS refers to a group of molecules including peroxides and free radicals which are derived from oxygen and are highly reactive toward biomolecules [17]. ROS react with critical biomolecules such as DNA and induce oxidative stress (imbalance of prooxidants versus antioxidants) and damage in these macromolecules, multiple localized lesions such as base damage, single strand breaks (SSBs) and double strand breaks (DSBs), DNA-DNA cross links and DNA-protein cross links [18-21]. As indicated by some investigators [22-24], we believe that induction of adaptive response by pre-exposure to ionizing radiation needs a minimum level of damage that triggers this phenomenon and increases the resistance of living organisms (*in vivo*) or cells (*in vitro*) to higher levels of the same or of other sources of stress.

Findings of this study show that in contrary with the results obtained in short term exposure to radon, exposure of animals to naturally elevated levels of gamma radiation does not lead to the induction of survival adaptive response. Consistent with our previously reported data [2, 25-34], radiobiological studies on the health effects of the chronic exposure to elevated levels of natural radiation in residents of areas such as Ramsar may lead to the identification of the cellular and molecular mechanisms by which susceptibility to genetic damage and cancer is decreased by chronic radiation exposure. These findings might have implications in areas such as radiation therapy, radiation protection and even selection of appropriate candidates for long term manned space missions [35].

Conflict of interest:
None Declared

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REFERENCES

1. Ghiassi-nejad M, Mortazavi SMJ, Cameron JR, Niroomand-rad A, Karam PA (2002) Very high background radiation areas of Ramsar, Iran: Preliminary biological studies. *Health Physics, **82:** 87-93.*
2. Mortazavi SMJ, Ghiassi-Nejad M, Rezaiean M. Cancer risk due to exposure to high levels of natural radon in the inhabitants of Ramsar, Iran. In: Sugahara T, Morishima H, Sohrabi M, Sasaki Y, Hayata I, Akiba S, editors.2005. pp: 436-7.
3. Mortazavi SMJ, Ghiassi-Nejad M, Karam PA, Ikushima T, Niroomand-Rad A, Cameron JR (2006) Cancer incidence in areas with elevated levels of natural radiation. *International Journal of Low Radiation, **2:** 20-7.*
4. ICRP (1990) Recommendations of the International Commission on Radiation Protection. Publication 60 ed. Tarrytown, NY: Elsevier Science.
5 . Barquinero JF, Barrios L, Caballin MR, Miro R, Ribas M (1995) Occupational exposure to radiation induces an adaptive response in human lymphocytes. *Int J Radiat Biol, **67:** 187–191.*
6. Barquinero JF, Barrios L, Caballin MR, Miro R, Ribas M, Subias A, Egozcue J (1996) Decreased sensitivity to the cytogenetic effects of bleomycin in individuals occupationally exposed to ionizing radiation. *Mutation Res, **354:** 81-86.*
7. Gourabi H and Mozdarani H (1998) A cytokinesis blocked micronucleus study of the radiadaptive response of lymphocytes of individuals occupationally exposed to chronic doses of radiation. *Mutagenesis, **13:** 475-480.*
8 . Mozdarani H and Gourabi H (2001) Chromosomal aberrations and micronuclei in lymphocytes of medical radiation workers exposed to ionizing radiation below dose limit. *Yakhteh Medical Journal, **3:** 153-158.*
9. Tedeschi B (1996) Do human lymphocytes exposed to fall out of Chernobyl accident exhibit an adaptive response. *Mutation Res, **354:** 77-80.*
10. Mitchel REJ, Jackson JS, McCann RA, Boreham DR (1999) Adaptive response modification of latency for radiation-induced myeloid leukemia in CBA/H mice. *Radiation Research, **152:** 273-279.*
11. Mitchel REJ, Jackson JS, Morrison DP, Carlisle SM (2003) Low doses of radiation increase the latency of spontaneous lymphomas and spinal osteosarcomas in cancer-prone, radiation-sensitive Trp53 heterozygous mice. *Radiation Research, **159:** 320-327.*
12. Monfared AS, Mozdarani H, Amiri M (2003) Natural background radiation induces cytogenetic radioadaptive response more effectively than occupational exposure in human peripheral blood lymphocytes. *Czechoslovak Journal of Physics, **53** Suppl **A:** 791-795.*
13. Mortazavi SMJ, Mosleh-Shirazi MA, Mehdizadeh S, Rouintan MS, Ebrahimi J, Tamaddon M, *et al.* (2010) Short-term radon inhalation induces significant survival adaptive response in Balb/c mice. *International Journal of Low Radiation,**7:** 98-109.*

14. McClintock B (1984) The significance of responses of the genome to challenge. *Science, 226: 792-801*.
15. Feinendegen LE, Bond VP, Sondhaus CA, Muehlensiepen H (1996) Radiation effects induced by low doses in complex tissue and their relation to cellular adaptive responses. *Mutation Res, 358: 199-205*.
16. Feinendegen LE, Bond VP, Sondhaus CA, Altman KI (1999) Cellular signal adaptation with damage control at low doses versus the predominance of DNA damage at high doses. *CR Acad Sci III, 322: 245-51.*
17. Maynard S, Schurman SH, Harboe C, de Souza-Pinto NC, Bohr VA (2009) Base excision repair of oxidative DNA damage and association with cancer and aging. *Carcinogenesis, 30: 2-10*.
18. Goldberg Z and Lehnert BE (2002) Radiation-induced effects in unirradiated cells: a review and implications in cancer. *Int J Oncol, 21: 337-49*.
19. Marnett LJ, Riggins JN, West JD (2003) Endogenous generation of reactive oxidants and electrophiles and their reactions with DNA and protein. *J Clin Invest, 111: 583-93.*
20. Cejas P, Casado E, Belda-Iniesta C, De Castro J, Espinosa E, Redondo A, *et al*. (2004) Implications of oxidative stress and cell membrane lipid peroxidation in human cancer (Spain). *Cancer Causes Control, 15: 707-19*.
21. Murray D, Allalunis-Turner M, Weinfeld M (2005) VIIIth International Workshop on Radiation Damage to DNA. *Int J Radiat Biol, 81: 327-37*.
22.Dimova EG, Bryant PE, Chankova SG (2008) Adaptive response: some underlying mechanisms and open questions. *Genet Mol Biol, 31: 396-408*.
23. Bose Girigoswami K and Ghosh R (2005) Response to gamma-irradiation in V79 cells conditioned by repeated treatment with low doses of hydrogen peroxide. *Radiat Environ Biophys, 44: 131-7*.
24. Yan G, Hua Z, Du G, Chen J (2006) Adaptive response of Bacillus sp. F26 to hydrogen peroxide and menadione. *Curr Microbiol, 52: 238-42.*
25. Ghiassi-nejad M, Mortazavi SM, Cameron JR, Niroomand-rad A, Karam PA (2002) Very high background radiation areas of Ramsar, Iran: preliminary biological studies. *Health Phys, 82: 87-93*.
26. Karam PA, Mortazavi SMJ, Ghiassi-Nejad M, Ikushima T, Cameron JR, Niroomand-rad A (2002) ICRP evolutionary recommendations and the reluctance of the members of the public to carry out remedial work against radon in some high-level natural radiation areas. *Radiation and Homeostasis, Proceedings, 1236: 35-7.*
27. Mortazavi SMJ, Ghiassi-Nejad M, Ikushima T (2002) Do the findings on the health effects of prolonged exposure to very high levels of natural radiation contradict current ultra-conservative radiation protection regulations? *Radiation and Homeostasis Proceedings, 1236: 19-21*.
28. Mortazavi SM, Cameron JR, Niroomand-rad A (2003) Adaptive response studies may help choose astronauts for long-term space travel. *Adv Space Res, 31: 1543-51.*
29. Mortazavi SMJ, Abbasi A, Asadi R, Hemmati A. The need for considering social, economic, and psychological factors in warning the general public from the possible risks due to residing in HLNRAs. In: Sugahara T, Morishima H, Sohrabi M, Sasaki Y, Hayata I, Akiba S, (Eds), 2005. pp: 440-1.
30. Mortazavi SMJ, Cameron JR, Niroomand-Rad A (2005) The life saving role of radioadaptive responses in long-term interplanetary space journeys. *International Congress Series, 1276: 266-7*.
31. Mortazavi SMJ and Karam PA (2005) Apparent lack of radiation susceptibility among residents of the high background radiation area in Ramsar, Iran: can we relax our standards? *Natural Radiation Environment, 7: 1141-7.*
32. Mortazavi SMJ, Shabestani-Monfared A, Ghiassi-Nejad M, Mozdarani H. Radioadaptive responses induced in lymphocytes of the inhabitants in Ramsar, Iran. In: Sugahara T, Morishima H, Sohrabi M, Sasaki Y, Hayata I, Akiba S, (Eds). 2005, pp: 201-3.
33. Mortazavi SMJ and Ikushima T (2006) Open questions regarding implications of radioadaptive response in the estimation of the risks of low-level exposures in nuclear workers. I*nternational Journal of Low Radiation, 2: 88-96.*
34. Mortazavi SMJ and Mozdarani H (2006) The search for a possible optimum adapting dose under the optimum irradiation time scheme in cultured human lymphocytes. *International Journal of Low Radiation, 3: 74-82*.
35. Mortazavi SMJ, Cameron JR, Niroomand-rad A (2003) Adaptive response studies may help choose astronauts for long-term space travel. *Advances in Space Research, 31: 1543-51.*