

VERY HIGH BACKGROUND RADIATION AREAS OF RAMSAR, IRAN: PRELIMINARY BIOLOGICAL STUDIES

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Abstract—People in some areas of Ramsar, a city in northern Iran, receive an annual radiation absorbed dose from background radiation that is up to 260 mSv y⁻¹, substantially higher than the 20 mSv y⁻¹ that is permitted for radiation workers. Inhabitants of Ramsar have lived for many generations in these high background areas. Cytogenetic studies show no significant differences between people in the high background compared to people in normal background areas. An *in vitro* challenge dose of 1.5 Gy of gamma rays was administered to the lymphocytes, which showed significantly reduced frequency for chromosome aberrations of people living in high background compared to those in normal background areas in and near Ramsar. Specifically, inhabitants of high background radiation areas had about 56% the average number of induced chromosomal abnormalities of normal background radiation area inhabitants following this exposure. This suggests that adaptive response might be induced by chronic exposure to natural background radiation as opposed to acute exposure to higher (tens of mGy) levels of radiation in the laboratory. There were no differences in laboratory tests of the immune systems, and no noted differences in hematological alterations between these two groups of people.

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Key words: health effects; naturally occurring radionuclides; radiation, background; exposure, population

INTRODUCTION

LIFE EVOLVED in an environment with higher radiation levels than exist today, and background radiation levels today are lower than at any time in the history of life on

Earth. Since life first evolved, background radiation levels have decreased by a factor of about 10, although there has been a negligible reduction since the evolution of humans (Karam and Leslie 1999). At present, natural background radiation levels on Earth vary by at least two orders of magnitude today, so humans and other organisms are subject to a wide range of background radiation levels. The annual background doses in some areas of the world are given in Table 1. These do not include contributions from radon progeny in the lungs, which are estimated to be even greater than the absorbed doses shown if the radiation weighting factor of alpha particles is taken into account. Areas with unusually high background (high background radiation areas, or HBRAs) are found in Yangjiang, China; Kerala, India; Guarapari, Brazil; and Ramsar, Iran. Some areas of Ramsar, a city in northern Iran, have among the highest known background radiation levels in the world. For the purposes of this paper, “dose” will be used to mean absorbed beta/gamma radiation dose because the contribution of alpha emitters is not considered.

The high background radiation in the “hot” areas of Ramsar is primarily due to the presence of very high amounts of ²²⁶Ra and its decay products, which are brought to the Earth’s surface by hot springs. Groundwater is heated by subsurface geologic activity and passes through relatively young and uraniumiferous igneous rock. Radium is dissolved from the rocks by hot ground water. Uranium is not dissolved because the groundwater is anoxic and uranium is insoluble in anoxic waters (Langmuir 1978; Grandstaff 1976). When the groundwater reaches the surface at hot spring locations, travertine, a calcium carbonate mineral, precipitates out of solution with dissolved radium substituting for calcium in the mineral. A secondary cause of high local radiation levels is travertine deposits with a high thorium concentration (Sohrabi 1990). The radioactivity in local soils and the food grown in them are also high because soils are derived from the weathering of local bedrock. Table 2 details the range of radioactivity levels measured in some local rocks and soil samples.

There are at least nine known hot springs with various concentrations of radioactivity around Ramsar. Residents and visitors use these springs as health spas. Residents of these “hot” areas have also used the residue of the hot springs as building materials to construct

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Table 1. Mean and maximum annual background absorbed doses (mGy y⁻¹) to the inhabitants of some areas of the world from geologic and cosmic radiation. The areas in Iran, India, Brazil, and China are all due to local geologic and geochemical effects, giving rise to very high and very localized radiation exposure. The other elevated radiation areas noted stem largely from the presence of large-scale geologic formations such as granitic mountain ranges, widespread geothermal activity, and so forth. (1) Radiation Research Foundation, Kyoto, Japan (used with permission of Radiation Research Foundation); (2) UNSCEAR (2000); (3) UNSCEAR (2000) reports maximum dose rates of 789 mGy y⁻¹ on beaches of monazite sands. The levels reported here are those found in the city itself; (4) Only mean exposure information is provided in the reference noted.

Area, Country	Mean dose (mGy y ⁻¹)	Maximum dose (mGy y ⁻¹)	Minimum dose (mGy y ⁻¹)
Ramsar, Iran		260 (1)	0.61
		149 (2)	
Kerala, India		35	1.75
Guarapari, Brazil (3)		1.49	0.79
Yangjiang, China (2, 4)	3.24		
Ireland (2)	0.37	1.58	0.01
Austria (2)	0.38	1.31	0.18
USA (2)	0.41	1.03	0.12
Germany (2)	0.44	3.07	0.04
Denmark (2)	0.46	0.61	0.31
Japan (2)	0.46	0.67	0.18
India (2)	0.49	9.64	0.18
China (2)	0.50	0.76	0.15
France (2)	0.60	2.19	0.09
Iran (2)	0.62	1.14	0.32
Norway (2)	0.64	10.52	0.18
Italy (2)	0.65	2.00	0.03
Hong Kong (2)	0.76	1.05	0.45
World Average (2)	0.50	0.82	0.16

Table 2. Analytical results from Ramsar plants and geologic media. All results are in Bq g⁻¹ with the 95% confidence intervals in parentheses. All analytical results are from Esmaili and Asgharnejad (2002). It is particularly interesting to note the difference in thorium and radium radioactivity concentrations. This is due to the different solubility of these two elements in water; radium (particularly in the form of RaCl) is very soluble while thorium is relatively insoluble.

Location	No. of samples	²²⁶ Ra	²³² Th	⁴⁰ K
Lamtarmaballeh	5	53.9 (8.2)	17.2 (2.0)	385.5 (18.5)
Ramak	3	5700.0 (93.4)	120.0 (20.0)	418.0 (62.0)
Gharmirh	5	110.5 (88.1)	32.2 (8.9)	423.0 (132.0)
Sadalmahelleh	4	127.0 (2.0)	28.3 (1.6)	296.0 (41.0)
Sefid Tamcahk	6	20.9 (3.2)	21.2 (2.2)	265.0 (81.0)
Lapasar	5	25.6 (2.0)	26.7 (11.7)	425.7 (92.0)
Chaparsar	11	114.1 (7.3)	15.7 (3.2)	634.0 (38.6)
Khake'safied	6	38,000 (10,000)	25.2 (1.8)	235.0 (82.2)
Talesh Mahalleh	15	19,000 (11,000)	43.5 (16.8)	436.6 (134.5)
Local stone (normal)		461.0 (11.5)	20.1 (2.8)	365.0 (51.0)
Travertine stone		421,000 (3000)	22.8 (4.1)	138.1 (22.0)

houses. The indoor and outdoor gamma radiation absorbed dose rates in some parts of Ramsar range from 50 to 900 μGy h⁻¹, although other areas in the city have absorbed dose rates that are much lower. The annual dose to monitored individuals ranges up to 132 mGy, and we have calculated maximum credible annual radiation exposures of up to 260 mGy. The recommended dose limit for workers in Iran is 20 mSv y⁻¹, so some residents in the Ramsar area receive a much higher annual radiation exposure than is permitted for radiation workers. Figs. 1 and 2 show the location of Ramsar and the highest background radiation areas with respect to populated areas.

The people who live in high radiation areas of the world are of considerable interest because they and their

ancestors have been exposed to abnormally high radiation levels over many generations. If an annual radiation exposure of a few hundred mSv is detrimental to health, causes genetic abnormalities, or an increased risk of cancer, it should be evident in these people, given a large enough population to study. A description of our study methods and results follows.

METHODS

Following is a brief description of the methods employed for testing accomplished to date.

Potential participants were identified from residents in areas of normal (14 persons) and elevated (21 persons) background radiation levels of Ramsar. After description of



Fig. 1. A map of Iran showing the location of Ramsar. The city lies between the Elburz Mountains and the Caspian Sea at an average elevation near sea level (the Caspian Sea is about 30 m lower than sea level). As noted in the text, this region is geologically young with an abundance of subsurface geothermal activity that is directly responsible for the elevated radiation levels present.

the study and its objectives to the participants, they were asked to complete questionnaires, participate in interviews, allow radiation measurements of their homes, and to submit blood samples. Every attempt was made to match data from the HBRA participants with corresponding ones from the normal background radiation area (NBRA) participants in as many aspects as possible, although this was not always feasible. Most families in this part of Iran have lived in the same geographic area for many generations, often remaining in the same house their entire lives. We are in the process of collecting detailed information from study participants on their family histories regarding moving to and

within Ramsar; this information will be included with our final results.

Cell and chromosome studies

Venous blood samples were drawn from healthy donors of both sexes who lived in high background areas (HBRA) or a neighboring normal background radiation area (NBRA) into heparinized vacutainers. The blood was cultured in culture flasks at 37°C. Fifty-two hours after phytohemagglutinin (PHA) stimulation, colcemid was added to arrest the dividing lymphocytes in mitosis. At 54 h, the cells were harvested. Metaphases with 46

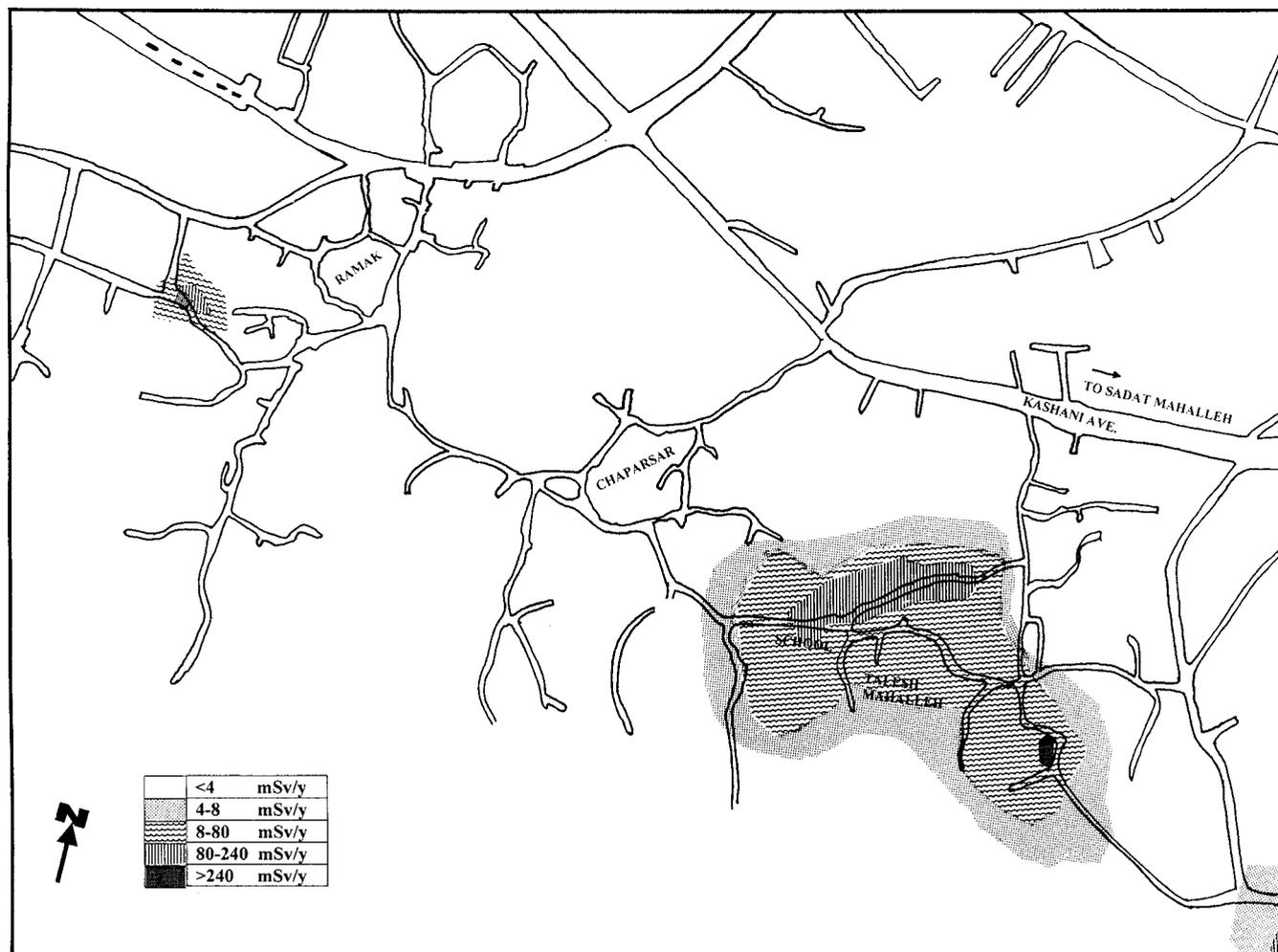


Fig. 2. A map of the HBRAs in Ramsar. The areas of highest background are found in discrete locations, corresponding to specific geologic features. Other areas of elevated background radiation levels are found elsewhere in the city; these are the highest levels found in residential areas. Although the locations of individual houses are not noted on this map, the entire area is residential, and 1–2 family houses are spaced about 20–30 m apart along each of the roads shown. On this map, one inch is equal to a distance of about 100 m.

chromosomes were scored and the frequency of chromosome aberrations determined.

Hematological studies

Healthy blood donors from HBRA of Ramsar as well as a neighboring NBRA were selected. Blood cell counts were performed by a Data Cell 20 machine.

Immunological studies

Healthy blood donors from HBRA and a neighboring NBRA were selected. Serum concentrations of different immunoglobulin classes were determined by Single Radial Immuno Diffusion (SRID) method.

Adaptive response studies

At least 200 lymphocytes from each blood sample were analyzed for chromosomal aberrations according to the procedure noted above, after which the blood samples

were then exposed only to a challenge dose of 1.5 Gy (i.e., no conditioning dose was given). The cells were exposed to the challenge dose of 1.5 Gy of gamma rays 48 h after PHA stimulation. The background level of chromosomal aberrations (i.e., before the conditioning dose was administered) in all residents was similar. Following exposure to the challenge dose, 200 lymphocytes from each participant were analyzed for the number of chromosomal aberrations and these results were averaged to determine the mean number of chromosomal aberrations per cell (reported as MCAPC).

RESULTS

Cancer incidence and life expectancy

Although there is not yet solid epidemiological information, most local physicians in Ramsar report anecdotally there is no increase in the incidence rates of

cancer or leukemia in their area (Mortazavi 2002). The life span of HBRA residents also appears no different than that in residents of nearby NBRAs, although this information is, again, anecdotal at this time. Such findings, if confirmed, are in keeping with the results of previous studies of HBRA residents and radiation workers (for example, Ikushima 1999; Chen and Wei 1991; Smith and Doll 1981).

Adaptation to radiation stress

It is known that cellular damage from a small amount of ionizing radiation and a variety of DNA damaging agents, such as UV, alkylating or oxidizing agents, and heat, can induce repair responses (Ikushima et al. 1996; Ikushima and Mortazavi 2002). The results of many studies indicate that when cells are exposed to low doses of stressing agents, they often become less sensitive to the harmful effects of a subsequent higher dose (Ikushima et al. 1996). This type of induced repair is called an adaptive response.

Our studies showed 56% fewer MCAPC resulting from the challenge dose among high background residents compared to lymphocytes from residents of neighboring low background areas ($p < 0.001$). These results are in keeping with those reported by Mitchell and Boreham (2000) in laboratory studies. We feel these results are intriguing because no overt conditioning dose was given to Ramsar residents; in this case, the “conditioning” dose was exposure to the natural background radiation in the Ramsar HBRA. The results of these studies are shown in Figs. 3, 4, and 5.

Hematological studies

Beginning in 1999, the National Radiation Protection Department of the Iranian Nuclear Regulatory Authority performed an integrated multi-disciplinary study on the health effects of relatively high levels of natural radiation in some areas of Ramsar. The main purpose was to look for hematological changes due to prolonged

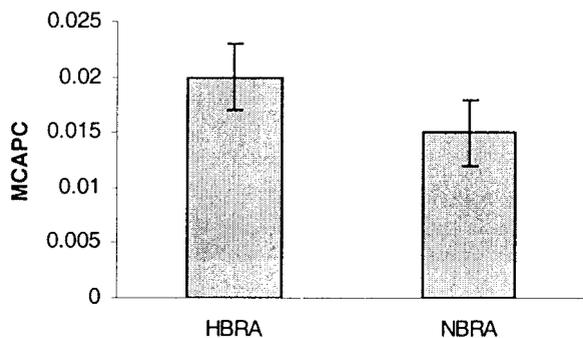


Fig. 3. Mean chromosome aberrations per cell (MCAPC) among 21 inhabitants of high background radiation areas (HBRA) and 14 living in normal background radiation areas (NBRA). Note that the 95% confidence intervals for these two populations overlap, indicating there is no statistically significant difference in the level of background chromosomal abnormalities in these two populations.

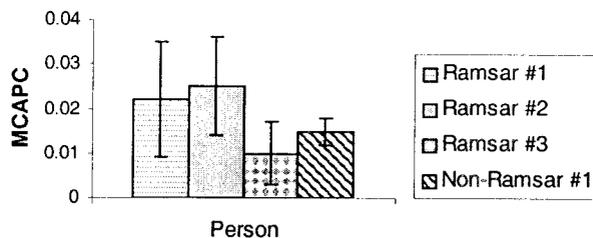


Fig. 4. MCAPC among individuals who live in the Ramsar HBRA and the average of 14 from a nearby control area. Persons 1 and 2 are married and live in a house with radiation levels of about 20 $\mu\text{Gy h}^{-1}$ in the bedroom, giving an average annual radiation dose of 58.4 mGy y^{-1} simply from sleeping. Person 3 has a hot spring in her house with radiation levels of up to 50 $\mu\text{Gy h}^{-1}$. Each data point is based on analysis of 200 cells per person obtained during two separate sampling events. Note that, as in Fig. 1, the 95% confidence intervals all overlap, suggesting that the number of background chromosome abnormalities in these people are statistically similar.

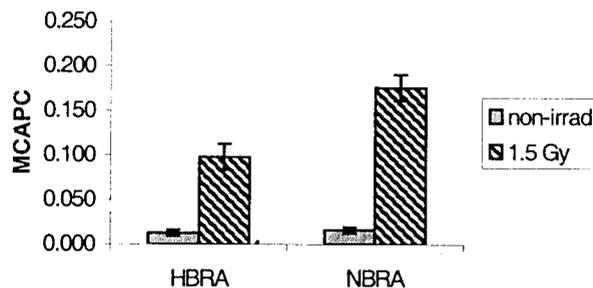


Fig. 5. MCAPC in irradiated and non-irradiated cells from inhabitants of HBRA and NBRA. Samples from inhabitants of both areas were examined for chromosomal abnormalities before and after irradiation with 1.5 Gy. Although there is no statistically significant difference in chromosomal abnormalities in both populations before irradiation, there is a statistically significant difference in post-irradiation abnormalities. In this case, cells from inhabitants of the HBRA have fewer chromosomal abnormalities than those of inhabitants of NBRA. Adaptive response has previously been noted in organisms exposed to acute conditioning doses at relatively high exposure rates; these results suggest that chronic exposure to lower exposure rates may stimulate adaptive response as well.

high background radiation. Healthy volunteers of both sexes from high background and neighboring normal background areas in Ramsar were examined for changes in the following hematological parameters: counts of white blood cells (leukocytes), lymphocytes, monocytes, granulocytes, and erythrocytes (red blood cells); measurements of hemoglobin, hematocrit, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH), mean corpuscular hemoglobin concentration (MCHC), red cell distribution width (RDW), platelet, and mean platelet volume (MPV). As noted above, our results indicate there are no statistically significant differences between the two groups of participants in all these hematological parameters.

Immunological studies

It is well known that high doses of ionizing radiation can suppress the immune system (Anderson et al. 1980; Celer 1990). On the other hand, some have suggested low doses can stimulate the immune system (e.g., Feinendegen et al. 1998; Mine et al. 1990). Studies were done to assess the effects of high background radiation on immune parameters. Blood samples were obtained from healthy individuals living in both high background and neighboring normal background areas. The following studies were done: concentration of serum immunoglobulins: immunoglobulin A (IgA), immunoglobulin G (IgG), immunoglobulin M (IgM), and complement components 3 and 4 (C3 and C4). Our findings showed a slight increase in IgA and IgG levels of residents from high, compared to normal, background areas. IgM, C3 and C4 complements were in the normal range for both groups. Although increases in IgA and IgG were not sufficient to show enhanced immune function, it is clear that high doses of natural radiation are not immunosuppressive. More research is needed to clarify the immunological alterations induced by different levels of natural radiation.

PRELIMINARY OBSERVATIONS

Although the long-term epidemiological studies of Ramsar residents are currently in progress, we report here some of the preliminary results of the cellular studies. Figs. 3, 4, and 5 show these preliminary results, which are based on the analysis of blood samples from 21 people living in the Ramsar HBRA and 14 controls from a nearby NBRA. On the basis of our preliminary data, we believe the following remarks are suitable:

1. These tests show no statistically significant difference in chromosome abnormalities at the 95% confidence level between these two groups (Fig. 3).
2. Blood samples from three individuals from an extraordinary HBRA (external gamma dose was up to $155 \mu\text{Gy h}^{-1}$) and 14 persons from a nearby NBRA also show no difference in the number of mean chromosome aberrations per cell (MCAPC). These results are based on examining 200 cells per person obtained during two separate sampling events (Fig. 4).
3. Some blood samples from both HBRA and NBRA were exposed to a challenge dose of 1.5 Gy. The purpose of the challenge dose is to induce radiogenic chromosomal abnormalities; cells demonstrating adaptive response will have fewer chromosomal abnormalities than those that do not have an adaptive response. In this case, the confidence intervals do not overlap and there is a statistically significant *reduction* in chromosomal abnormalities following this challenge dose among the residents of the HBRA (Fig. 5). Specifically, HBRA residents' cells had an average of 0.098 chromosomal aberrations per cell following this exposure compared to 0.176 aberrations noted in cells from residents of NBRA.

4. Previous studies of adaptive response have concentrated on stimulating effects with acute, relatively high doses of radiation (UNSCEAR 1994). The results shown in Fig. 5 suggest that chronic exposure to relatively low levels of natural radiation may also induce adaptive response. Further, it is possible that continual radiation exposure may help to overcome the relatively short adaptive response "half-life" that has been reported.

DISCUSSION AND CONCLUSIONS

Cytogenetic analysis of chromosome aberrations in peripheral blood lymphocytes is widely used to quantify dose from radiation and other clastogens (Hoffmann and Schmitz-Feuerhake 1999). Our preliminary cytogenetic studies indicate *no* statistically significant difference in chromosomal abnormalities between residents of the HBRA and people in control areas. It is worth noting that our preliminary results do not agree with that reported by Fazeli (1990) for residents in high background areas. This needs further investigation. However, on the basis of these preliminary results, we argue that the risk from the high levels of natural background radiation found in Ramsar may be less than the predictions of linear no-threshold or supra-linear models of radiation dose-response. We would also like to note that the annual absorbed radiation dose to some residents of Ramsar are significantly higher than the recommended limit for the general public under ICRP recommendations and Iranian regulations. In some cases, these levels are even higher than the ICRP-recommended annual limits for radiation workers (ICRP 1990).

In addition, we note that the chromosomal aberration studies reported here, *if borne out by research currently underway*, suggest that radioadaptive response may be initiated by long-term exposure to relatively low levels of ionizing radiation. Previous experiments in adaptive response had looked primarily at the effects of acute exposure to relatively high levels of radiation, and the effects decreased over a period of days, as reported by the United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR 1994). The stimulation of adaptive response by elevated levels of natural background radiation suggests a plausible evolutionary origin for this effect.

Please note that adaptive response (AR) refers to the observation that exposing cells or some organisms to relatively low "conditioning" doses of radiation prior to exposure to a much higher "challenge" dose results in a reduction in observed radiation effects (e.g., chromosomal aberrations, death, etc.). By comparison, hormesis is the induction of beneficial effects (e.g., reduction in cancer rates, increase in life span) resulting from exposure to a small amount of an agent. Our preliminary studies seem to indicate the presence of adaptive response in the cells of some Ramsar residents, but we do not claim to have seen hormetic effects in any of those studied.

Given the apparent lack of ill effects among observed populations of these high dose rate areas, these data suggest that current dose limits may be overly conservative. However, the available data do not yet seem sufficient to cause national or international advisory bodies to change their current conservative radiation protection recommendations; for this to happen more definitive data are needed (Roth et al. 1996).

Radio-epidemiological studies of the residents of high background areas, such as Ramsar, will provide useful supporting data. The population in the high background areas of Ramsar is estimated to be about 2,000 persons. To obtain statistically reliable results, a long-term study will be needed to provide sufficient person-years of observation. The life span of residents of the high background areas of Ramsar should be a part of future long-term studies. In time, we plan to extend our study to include residents of Iran living in other areas that, while not as high-dose as Ramsar, are still well above the global average radiation dose. We feel that these factors will make it possible to examine a wider portion of the dose-response curve than is possible in any other location on Earth.

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