RESEARCH ARTICLE

Low-dose Radiation Induces Antitumor Effects and Erythrocyte System Hormesis

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Abstract

Objective: Low dose radiation may stimulate the growth and development of animals, increase life span, enhance fertility, and downgrade the incidence of tumor occurrence.The aim of this study was to investigate the antitumor effect and hormesis in an erythrocyte system induced by low-dose radiation. Methods: Kunming strain male mice were subcutaneously implanted with S180 sarcoma cells in the right inguen as an experimental in situ animal model. Six hours before implantation, the mice were given 75mGy whole body X-ray radiation. Tumor growth was observed 5 days later, and the tumor volume was calculated every other day. Fifteen days later, all mice were killed to measure the tumor weight, and to observe necrotic areas and tumor-infiltration-lymphoreticular cells (TILs). At the same time, erythrocyte immune function and the level of 2,3-diphosphoglyceric acid (2,3-DPG) were determined. Immunohistochemical staining was used to detect the expression of EPO and VEGFR of tumor tissues. Results: The mice pre-exposed to low dose radiation had a lower tumor formation rate than those without low dose radiation ($P < 0.05$). The tumor growth slowed down significantly in mice pre-exposed to low dose radiation; the average tumor weight in mice pre-exposed to low dose radiation was lighter too ($P < 0.05$). The tumor necrosis areas were larger and TILs were more in the radiation group than those of the group without radiation. The erythrocyte immune function, the level of 2,3-DPG in the low dose radiation group were higher than those of the group without radiation ($P < 0.05$). After irradiation the expression of EPO of tumor tissues in LDR group decreased with time. LDR-24h, LDR-48h and LDR-72h groups were all statistically significantly different from sham-irradiation group. The expression of VEGFR also decreased, and LDR-24h group was the lowest ($P < 0.05$). Conclusion: Low dose radiation could markedly increase the anti-tumor ability of the organism and improve the erythrocyte immune function and the ability of carrying O_2. Low-dose total body irradiation, within a certain period of time, can decrease the expression of hypoxia factor EPO and VEGFR, which may improve the situation of tumor hypoxia and radiosensitivity of tumor itself.

Keywords: Low dose radiation - S180 sarcoma - TIL - erythrocyte Immune function - EPO - VEGFR - 2,3-DPG

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Introduction

Since the hypothesis of hormesis was reported in 1982 by Luckey, people have had some new understand on this problem (Day et al., 2007; Ramola et al., 2010, 2012; Walsh & Kaiser, 2011; Rautela et al., 2012). The epidemiology showed that the incidence of tumor was not increased in those people who were workers in A-test place, victims receiving low dose radiation in explosion of atom-bomb and atomic accidents, medical staff in radiology department and patients receiving radiation treatment. On the contrary, it decreased (Ramola et al., 2010, 2012; Rautela et al., 2012).

Study showed immune function in these people was up-regulation, so tumor growth was inhibited (Little, 2009, 2010; Walsh & Kaiser, 2011 Little et al., 2012). The immune hormesis induced by low dose radiation has been proved in lymphocyte, macrophage and natural killer (NK) cells (Day et al., 2006, 2007; Halliday & Rana, 2008; Meng et al., 2012).

The effect of low dose radiation on erythrocyte immune function, the ability of carrying O_2 is not reported. This study explored the effect of low dose radiation on tumor growth and erythrocyte immune function, the ability of carrying O_2, and illuminates further the mechanism of hormesis of low dose radiation.

Radiotherapy and chemotherapy are now the main methods to treat malignant tumors. However, both of the treatment can injury the human body inevitably by inhibiting the immune function, which influences the clinical treatment. Some patients with malignant tumors died of the complications related to the inhibition of immune function. How to improve these patients' immune function has become a baffling question in clinical work. There have been some reports that low dose radiation had exciting effects on lymphocytic immune function (Day

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et al., 2006, 2007; Halliday & Rana, 2008; Meng et al., 2012). But there have been few reports about the effects of low dose radiation on erythrocyte immune function. This study explored the effects of low dose radiation on tumor growth and changes of erythrocyte immune function and the level of 2,3-diphosphoglyceric acid (2,3-DPG) in tumor-bearing mice. The study may provide theoretical evidence for the clinical use of low dose radiation.

Low-dose radiation (LDR), proven in many studies, can enhance immunity and anti-tumor effect in vivo through various means. Total body irradiation with 75mGy has the most significant effect.

However, the tumor-inhibitory mechanism of low-dose radiation is currently no unified opinion. By studying the effect of LDR on EPO, VEGFR of tumor tissues in mice bearing S180 sarcoma, we explore the molecular mechanism of tumor-inhibition, hypoxic improvement induced by LDR.

Materials and Methods

Animals and grouping

Kunming strain male mice were randomly divided into five groups: (1) normal control group (2) pre-LDR group (3) sham- pre-LDR group. (4) sham-irradiation (S) group (5) low-dose radiation (LDR) group: The mice in LDR group were randomly divided into 5 groups, executed them at 6h (LDR-6h group), 12h (LDR-12h group), 24h (LDR-24h group), 48h (LDR-48h group) and 72h (LDR-72h group) after irradiation. The mice in S group were executed at 6h after exposure.

All these mice were raised routinely with un-limited water and food. This study was carried out in strict accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the National Institutes of Health. The animal use protocol has been reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) of the Affiliated Hospital of Medical College, Qingdao University.

Radiation conditions

BeiJing F34-1 deep X-ray machine (BeiJing, China), voltage 200kv, electric current 10mA, filter 0.5mm Cu+ 1.0mmAl was used. The mice in the LDR group and pre-LDR group were packed in self-made paper box, and given whole body radiation, 75 mGy (total dosage), 12.5 mGy/min, source skin distance (SSD) was 172 cm.

S180 sarcoma cell implantations

S180 sarcoma cells brought from Chinese Medical Science Institue.S180 sarcoma cells were generated for two generations in the abdomen of the mice. The cell solution in logarithmic growth phase was made (1×10^7 cells/ml). The mice except those in the normal control group were implanted with S180 sarcoma cells with 0.1 ml (total 1×10^6 cells) in right inguen subcutaneously.

Tumor growth observation and biopsy examination

Six hours before implantation, the mice in the pre-LDR group were given 75 mGy whole body X-ray radiation. 5 days after implantation of S180 sarcoma cells, the mice's tumor growth was observed every other day by measuring the maximum horizontal diameter (a) and the maximum vertical diameter (b) twice with slide gauge and then the average values were calculated. The tumor's volume (V) was calculated with the formula: $V=1/2ab^2$ (Yu et al., 2007). Then the tumor's growth graph was drawn. 15 days after implantation, the mice were sacrificed and the tumor was separated from the body of the mice. The subcuteneous fats were got rid of, and the tumor part was weighed. The tumor was fixed by 10% formalin for several days, sliced, and HE dyed. The tumor's necrosis areas and the lymphocytes invasion were observed.

Erythrocyte immune function, the level of 2,3-DPG determination

The materials of erythrocyte immune were provided by professor GuoFeng in Affilitated CangHai Hospital of the second military Medical University. 15 days after the implantation, blood was taken from the venous sinus of the right eye of the mice, anticoagulated by heparin, and then detected the erythyocyte immune function, the level of 2,3-DPG. Erythyocyte immune function was detected by the method of GuoFeng (Mauriello et al., 2013). The red blood cell's surface's C3b receptor rate (RBC-C3bRR) and red blood cell's immune compound rate (RBC- IcR) were determined. 2,3-DPG in red blood cell was determined by spectrophotometric method, and operated according to the specification strictly, presented by umol/gHb.

EPO and VEGFR determination

Completely dissected subcutaneous tumor nodules, generally observed the tumor specimens with unaided eyes, weighed the tumor specimen in the balance with accuracy of 1mg, then rapidly fixed them in 10% formaldehyde solution.PowerVisionTM two steps method immunohistochemical staining was used to detect the expression of EPO and VEGFR. Replace Primary antibody by PBS as negative control, the known positive biopsy as a positive control. Randomly prepared each sample into 5 slices, each of which was observed 10 high-power fields (400X) under microscope to count the expression rates of EPO and VEGFR (expression rate=positive cells/all counting cell×100%).

Statistical analysis

Measurement data were all expressed by mean±standard deviation (x±s). T-test was used to compare two groups. Student-Newman-Keuls test was used to analyze multi-group data. Numeration data was analyzed by X^2-test.

Results

The inhibition effects on tumor occurrence

The mice pre-exposed to low dose radiation had a lower rate tumor occurrence than those without low dose radiation ($P < 0.05$). The former was 75.00% (30/40), while the latter was 97.29% (36/37).

The inhibition effects on tumor growth

From the 5th day after implantation of S180 sarcoma cells, the tumor's size was calculated every other day,

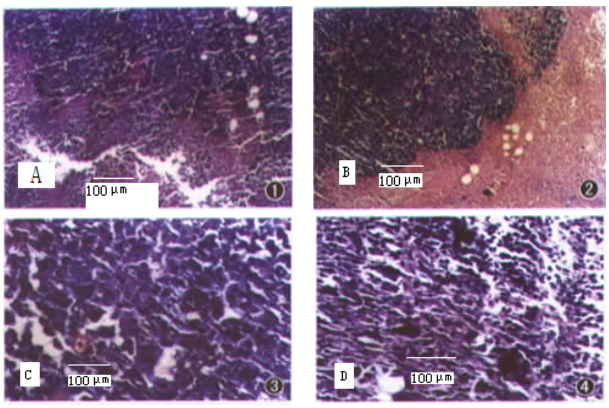

Figure 1. A) The necrosis areas of the group radiation. B) The necrosis areas of the low dose radiation group. C) Tumor-infiltrating lymphocytes (TIL) of the non-radiation. D) TILs of the low dose radiation group

which is showed in Table 1. The tumor sizes of mice of the pre-exposed to low dose radiation group were lower than those of the group without low dose radiation from the 7th day, especially from the 9th day to 15th day ($p < 0.01$).

Tumor's biopsy examination

15 days after the implantation, the mice were sacrificed, separated tumor and weighed the tumor, The tumor's average weight of the mice in the low dose radiation group on the 15th day was (0.743±0.210) g, which was lower than those of the group without low dose radiation ,which was (1.330±0.298) g, the two groups were significantly different ($p < 0.05$). The histological feature of maximum cross-section under microscope was that the mice in the low dose radiation group had more necrosis areas and significantly more lymphocytes compared with those in the group without low dose radiation (Figure 1).

Effects of low dose radiation on erythrocyte immune function and level of 2,3-DPG

The mice pre-exposed to low dose radiation had higher RBC-C3bRR, RBC- IcR, intracellular concentration of 2,3-DPG than those without radiation ($p < 0.05$), but there is no significant difference compared with normal control group ($p > 0.05$) (Table 1).

LDR induced impact on expression of EPO in tumor tissues

Analysis of EPO-positive index showed there were significant differences in statistics compared LDR-24h group with S group ($p<0.05$), LDR-48h and LDR-72h with S group ($p<0.01$), LDR-24h with LDR-48h ($p<0.05$, Table 2).

Table 1. Effects of Low Dose Radiation on the Tumor Growth (cm^3)

Days after implantation	Group without radiation	Group with radiation
5	0.202±0.096	0.167±0.091
7	0.369±0.228	0.290±0.122*
9	0.748±0.485	0.393±0.199**
11	1.331±0.927	0.598±0.375**
13	1.407±0.672	0.645±0.483**
15	1.318±0.526	0.679±0.378**

*$P<0.05$; **$P<0.01$

LDR induced impact on expression of VEGFR in tumor tissues

Semi-quantitative analysis of VEGFR expression showed there were significant differences in statistics compared LDR-12h group with S group ($p<0.05$, Table 2).

Discussion

The hormesis of low dose radiation manifest in many facet, for example, we have proved that low dose radiation may stimulate the growth and development of animal, increase in life span, enhance the fertility, degrade the incidence of tumor occurrence (Gur'eva et al., 2007; Tsetlin et al., 2008; Doss, 2012). Epidemiology research showed the mortality of the inhabitant in high radiation background involved a downward direction. Epidemiology research in 48 states in America showed there was negative correlation between the level of natural radiation and the standard mortality of carcinoma (Correa et al., 2013). The result of research in high radiation background in Indian was the same with that of America (Van Bocxlaer et al., 2009). The incidence of leucocythemia in the survivors of A-bomb in Japan who had received 6.0-9.0cGy radiation was lower than those in the control groups, the former was 0.014×10^{-5}, while the latter was 0.064×10^{-5} (Ozasa et al., 2012; Sakata et al., 2012). The mortality of inhabitants in natural high radiation background in GuangDong province was also lower than those in the control groups, the mortality of carcinoma was 44.66×10^{-5} and 51.00×10^{-5} separately (Raabe, 2010).

Low dose radiation can decrease and inhibit the occurrence of tumors by activating immune system. Our study showed that the mice pre-exposed to low dose radiation (75 mGy X-ray for whole body) may lead to stronger inhibition on tumors than those without low dose radiation, which is in accordance with the results of other reports (Zeeb et al., 2012).

Studys showed that low dose radiation could inhibit the growth and transfer of tumor under some conditions.

Table 2. Effects of Low Dose Radiation on Erythrocyte Immune Function, Level of 2,3-DPG and Activity of SOD

Groups	n	Erythrocyte immune function			2,3-DPG (umol/gHb)
		C3bRR (%)	IcR (%)		
Normal control	40	17.1±2.8	3.2±1.8	39.6±20.1	20131±552.2
Group without radiation	37	9.6±3.7*	7.3±1.7*	25.1±12.3*	14903±631.0*
Group with radiation	30	16.8±4.0	4.8±1.9	32.8±14.5	19047±715.2

compared with normal control groups, *$p<0.05$

The inhibition effects of low dose radiation on tumor growth were gained by the enhancement of immune function that was through the whole regulation of every system of organism, not through directly killing tumor cells by radiation. As reported by Liu et al. (2010), low dose radiation could accelerate the production, maturation and renewal of immune cells and then enhance immune function of organism. Another possible mechanism was the relieving inhibition of immune system through down-reguation of hypothalamus-pituitary-adrenal axis after low dose radiation for whole body (Reissfelder et al., 2011). The tumor interstitial cell invasion is thought a local immune reaction, which indirectly reacts to body's immune function. Horst's study showed that tumor's invasion lymphocytes (TIL) had 50 to 100 times more defensive effects than lymphokine-activated killer (LAK) cells, so TIL can be as an evaluation index of body's immune function (Jiang et al., 2013). Our research showed that low dose radiation could decrease the tumor occurrence and slow down the tumor growth, which was in accordance with other reports. In addition, we observed through the pathological section of tumor that low dose radiation could enlarge tumor's necrosis areas and more TILs could be found in tumor tissue under the microscope after low dose radiation. All these indicated that low dose radiation could activate mice's defensive system and improve anti-tumor ability, as a result, implanted tumor cells were inhibited and tumor growth was slowed down.

For long time, people thought that lymphocytes played very important roles in body's immune system, while red blood cells could only transmit gas such as oxygen and carbon dioxide. Since the concept of erythrocyte immune was advanced, the new domain of immune system of organism was opened up (Cremel et al., 2013). Studys showed red blood cell can adhere to tumor cells by the surface's C3b receptor (C3bR), formed immunocomplex, which could be easily eaten by macrocytes. In the peripheral blood, red blood cells (RBCs) are about 103 times more than white blood cells (WBCs), and easier to contact with circular immunocomplex, which then can be eaten by macrocytes. RBC's C3bR may be blocked with the inhibition of body's immune function and the increase of circular immunocomplexes. Thus, large amount of circular immunocomplexes may stay at tissues and organs, which may lead to immune injury. Some studies showed that the immune function of erythrocytes is positively correlated with that of the lymphocyte (Kwak et al., 2012). The erythrocyte immune function can be determined with only a small sample of blood, so it's clinically easy and useful and can substitute lymphocyte immune function examination.Red blood cells could also produce NKEF (Natural Kill Cell Enhancement Factor), enhanced the function of natural killer (NK) cells and the lethal effect on tumor. Erythrocyte immune function in tumor patients was lower than normal people, so it was perhaps linked to tumor occurrence (de Carvalho Lins et al., 2004). The effect of low dose radiation on erythrocyte immune function has not been reported. Our study showed that mice's erythrocyte immune function was inhibited after tumor cells implantation and both RBC- C3bR's activity and RBC- C3bRR decreased while circular immunocomplex and RBC-IcR increased. After being given low dose radiation, mice with implantation tumor had the same erythrocyte immune function as normal mice, which suggested that low dose radiation has hormesis on erythrocyte immune function. Tumor inhibition effects induced by low dose radiation are closely related to the improvement of erythrocyte immune function. But its mechanism is still not very clear, which needs further investigation.

Maturated red blood cells have no cellular nucleus and mitochondrion, and obtain energy through anaerobicglycolysis. 2,3-DPG is the important intermediate product in anaerobic glycolysis and plays an important role in regulating hemoglobin carrying oxygen. Oxygen dissociation curve moves right when 2,3-DPG content increase in red blood cells, as a result, oxygen and hemoglobin dissociate, so 2,3-DPG can represent the function of the ability of carrying oxygen of red cells. Our study showed low dose radiation could speed up the anaerobic glucolysis in red blood cells, increased the content of 2,3-DPG and then enhanced the ability of carrying oxygen.The exciting effect of low dose radiation on 2,3-DPG of erythrocyte might be one of the mechanisms of the enhancement of erythrocyte immune function.

The optimum dose for hormesis of low dose radiation is not reported. Some researcher thought it was suitable for mice to receive whole body 5.0 or 10.0 cGy one time (Vaiserman, 2010). The time length of hormesis of low dose radiation has not reached agreement. Some researchers (Pandey & Rizvi, 2011) reported that the enhancement of immune function could persist for several weeks under single or chronic irradiation. Other researches (Jolly & Meyer, 2009) showed that single fraction irradiation could enhance immune function, this reaction would last for 7 days, but not more than 14 days. Our study showed erythrocyte immune function was still excited 15 days after whole body low dose radiation by 7.5cGy single fraction irradiation. However, the optimum irradiation dose and time length of erythrocyte immune system is to be further observation and investigation.

Many experimental studies and clinical data showed that when malignant tumor was hypoxic erythropoietin (EPO) was in high expression, the hypoxic status could activate the vascular endothelial growth factor receptor (VEGFR) which promote the metastasis and recurrence of tumor, reduce the efficacy of radiotherapy and chemotherapy (Huang &Chen,2008). This study found that after low-dose total body irradiation on mice bearing S180 sarcoma, the EPO in tumor tissues was reduced, which indirectly reflect the hypoxic state in tumor tissues was improved.

Vascular endothelial growth factor receptor (VEGFR) is an important part of regulatory mechanism of growth and proliferation in normal human cells. VEGFR is widely expressed in human epithelial cells and stroma-cells, high expressed in many kinds of human malignant tumors. It is related to tumor resistance, proliferation angiogenesis invasion and metastasis. High expression of VEGFR could lead to curative effect of lowering and clinical treatment failure, and ultimately promote tumor

development (Huang &Chen, 2008). The study found that after low-dose total body irradiation the expression of EGFR increased after a reduction. The reduction reached the minimum at 12h after irradiation, which is statistical significantly different from the S group (p<0.05). Studies had demonstrated that the expression of VEGFR in tumor tissue is positive correlated with its hypoxic state. When the tumor tissue is hypoxia, VEGFR high expressed. That Low-dose total body irradiation mobilizes immune system would reduce the expression of VEGFR in some way, and indirectly reflect the hypoxia situation in the tumor tissue, at the same time oppose the growth of tumors and reduce tumor invasion and metastasis within a certain period of time.

Studies have shown that at the beginning of the formation of solid tumors the nutrition for tumor cells relied on diffusion, but when the solid tumor grew to a diameter more than 1-2mm, diffusion would not meet the needs of the tumor to survive or grow, the ingrowth of new blood vessels is needed. Once the new vessels grow into the tumor, it would grow rapidly in turn. Folkman (Elbarghati et al., 2008) called this phenomenon "the balance for the angiogenesis switch". In this process tumor cells were hypoxic, and the presence of hypoxia factors contributed to the high expression of HIF-1. The current study found that HIF-1 could promote high expression of vascular endothelial growth factor (VEGF), erythropoietin (EPO) by hypoxia response element. So that tumor cells can adapt to the microenvironment of hypoxia (Semenza., 2000).

This experiment found that after low-dose total body irradiation to mice bearing S180 sarcoma, the expressions of EPO and VEGF were decreased. This on the one hand restrained tumor growth, metastasis and recurrence, on the other hand when tumor cells were hypoxic the activity of HIF-1 increased, and also improved the expressions of EPO and VEGF. The reduction of EPO and VEGF could reflect the activity of HIF-1 may decrease, which means low-dose total body irradiation could improve the hypoxia situation in the tumor tissue.

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References

Correa JH, Castro LS, Kesley R, et al (2013). Salvage abdominoperineal resection for anal cancer following chemoradiation: a proposed scoring system for predicting postoperative survival. *J Surg Oncol*, **107**, 486-92.

Cremel M, Guérin N, Horand F, Banz A, Godfrin Y (2013). Red blood cells as innovative antigen carrier to induce specific immune tolerance. *Int J Pharm*, **443**, 39-49.

Day TK, Hooker AM, Zeng G, Sykes PJ (2007). Low dose X-radiation adaptive response in spleen and prostate of Atm knockout heterozygous mice. *Int J Radiat Biol*, **83**, 523-34.

Day TK, Zeng G, Hooker AM, et al (2006). Extremely low priming doses of X radiation induce an adaptive response for chromosomal inversions in pKZ1 mouse prostate. *Radiat Res*, **166**, 757-66.

Day TK, Zeng G, Hooker AM, et al (2007). Adaptive response for chromosomal inversions in pKZ1 mouse prostate induced by low doses of X radiation delivered after a high dose. *Radiat Res*, **167**, 682-92.

de Carvalho Lins CE, Pereira Crott LS, Teixeira JE, Barbosa JE (2004). Reduced erythrocyte complement receptor type 1 in systemic lupus erythematosus is related to a disease activity index and not to the presence or severity of renal disease. *Lupus*, **13**, 517-21.

Doss M (2012). Evidence supporting radiation hormesis in atomic bomb survivor cancer mortality data. *Dose Response*, **10**, 584-92.

Elbarghati L, Murdoch C, Lewis CE(2008). Effects of hypoxia on transcription factor expression in human monocytes and macrophages. *Immunobiology*, **213**, 899-908

Gur'eva TS, Dadasheva OA, Tsetlin VV, Mednikova EI, Lebedeva ZN (2007). Effect of chronic exposure to low doses of ionized radiation on embryonal development of the Japanese quail. *Aviakosm Ekolog Med*, 41, 20-4.

Halliday GM, Rana S (2008). Waveband and dose dependency of sunlight-induced immunomodulation and cellular changes. *Photochem Photobiol*, 84, 35-46.

Huang G, Chen L (2008). Tumor vasculature and microenvironment normalization: a possible mechanism of antiangiogenesis therapy. *Cancer Biother Radiopharm*, **23**, 661-7. Review

Jiang J, Wu C, Lu B (2013). Cytokine-induced killer cells promote antitumor immunity. *J Transl Med*, **11**, 83.

Jolly D, Meyer J (2009). A brief review of radiation hormesis. *Australas Phys Eng Sci Med*, **32**, 180-7.

Kwak JH, Baek SH, Woo Y, et al (2012). Beneficial immunostimulatory effect of short-term Chlorella supplementation: enhancement of natural killer cell activity and early inflammatory response (randomized, double-blinded, placebo-controlled trial). *Nutr J*, **11**, 53.

Little MP (2009). Cancer and non-cancer effects in Japanese atomic bomb survivors. *J Radiol Prot*, **29**, A43-59.

Little MP (2010). Do non-targeted effects increase or decrease low dose risk in relation to the linear-non-threshold (LNT) model? *Mutat Res*, **687**, 17-27.

Little MP, Azizova TV, Bazyka D, et al (2012). Systematic review and meta-analysis of circulatory disease from exposure to low-level ionizing radiation and estimates of potential population mortality risks. *Environ Health Perspect*, **120**, 1503-11.

Liu R, Xiong S, Zhang L, Chu Y (2010). Enhancement of antitumor immunity by low-dose total body irradiationis associated with selectively decreasing the proportion and number of T regulatory cells. *Cell Mol Immunol*, **7**, 157-62.

Mauriello CT, Pallera HK, Sharp JA, et al (2013). A novel peptide inhibitor of classical and lectin complement activation including ABO incompatibility. *Mol Immunol*, **53**, 132-9.

Meng X, Zhang Y, Zhao Y, Lou IC, Gao J (2012). Review of chinese environmental risk assessment regulations and case studies. *Dose Response*, **10**, 274-96.

Ozasa K, Shimizu Y, Suyama A, et al (2012). Studies of the mortality of atomic bomb survivors. Report 14.1950-2003: an overview of cancer and nocancer disease. *Radiat Res*, **177**, 229-43.

Pandey KB, Rizvi SI (2011). Biomarkers of oxidative stress in red blood cells. *Biomed Pap Med Fac Univ Palacky Olomouc Czech Repub*, **155**, 131-6.

Raabe OG (2010). Concerning the health effects of internally deposited radionuclides. *Health Phys*, **98**, 515-36.

Ramola RC, Gusain GS, Rautela BS, et al (2012). Levels of thoron and progeny in high background radiation area of southeastern coast of Odisha, India. *Radiat Prot Dosimetry*, **152**, 62-5.

Ramola RC, Prasad G, Gusain GS, et al (2010). Preliminary indoor thoron measurements in high radiation background area of southeastern coastal Orissa, India. Radiat Prot Dosimetry, **141**, 379-82.

Rautela BS, Yadav M, Bourai AA, et al (2012). Study of natural radionuclide and absorbed gamma dose in Ukhimath area of Garhwal Himalaya, India. Radiat Prot Dosimetry, **152**, 58-61.

Reissfelder C, Timke C, Schmitz-Winnenthal H, et al (2011). A randomized controlled trial to investigate the influence of low dose radiotherapy on immune stimulatory effects in liver metastases of colorectal cancer. *BMC Cancer,* **11**, 419.

Sakata R, Grant EJ, Ozasa K (2012). Long-term follow-up of atomic bomb survivors. *Maturitas*, **72**, 99-103.

Semenza GL (2000). HIF-1:mediator of physiological and pathophysiological responses to hypoxia [J]. *J Apple physiol*, **88**, 1474-80

Tsetlin VV, Levinskikh MA, Nefedova EL, Derendiaeva TA, Fedotova IV (2008). Effects of low doses of ionizing radiation on substrate and germination of higher plants seeds. *Aviakosm Ekolog Med*, **42**, 18-22.

Vaiserman AM (2010). Radiation hormesis: historical perspective and implications for low-dose cancer risk assessment. *Dose Response*, **8**, 172-91.

Van Bocxlaer I, Biju SD, Loader SP, Bossuyt F (2009). Toad radiation reveals into-India dispersal as a source of endemism in the Western Ghats-Sri Lanka biodiversity hotspot. *BMC Evol Biol*, **9**, 131.

Walsh L, Kaiser JC (2011). Multi-model inference of adult and childhood leukaemia excess relative risks based on the Japanese A-bomb survivors mortality data (1950-2000). *Radiat Environ Biophys*, **50**, 21-35.

Yu HS, Xue HW, Guo CB, et al (2007). Low dose radiation increased the therapeutic efficacy of cyclophosphamide on S(180) sarcoma bearing mice. *J Radiat Res*, **48**, 281-8.

Zeeb H, Hammer GP, Blettner M (2012). Epidemiological investigations of aircrew: an occupational group with low-level cosmic radiation exposure. *J Radiol Prot*, **32**, N15-9.