The 3rd International Symposium on Radiation Emergency Medicine in Hirosaki University

Hirosaki University Graduate School of Health Sciences, Hirosaki, Japan
September 17, 2011

http://www.hs.hirosaki-u.ac.jp/kouhou/hokengaku/
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Floor Plan

Hirosaki University Graduate School of Health Sciences

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- Drink
- Registration
- Poster Presentation Room
- Lounge
- Conference Room
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5F

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- Meeting Room
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Theme

“Dose Estimation and Protection in Radiation Exposure”
Program

September 17, 2011(Sat)

9:00 – Registration

9:30 – 9:40 Opening & Welcome
Prof. Hitoshi Tsushima,
Dean of Hirosaki University Graduate School of Health Sciences

9:40 – 10:40 Symposium I
Chair: Guest Prof. Mikinori Kuwabara, Hirosaki University

9:40 – 10:20 Measures against nuclear accident at TEPCO Fukushima Daiichi Nuclear Power Plant: Activities carried out by Hirosaki University Institute of Radiation Emergency Medicine
Shinji Tokonami
Department of Radiation Physics, Hirosaki University Institute of Radiation Emergency Medicine

10:00 – 10:20 Environmental radiation in Fukushima after nuclear power station accident
Masahiro Hosoda
Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

10:20 – 10:40 Role of nurses in a nuclear disaster
Yuka Noto
Department of Disability and Health, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

10:40 – 11:00 Break

11:00 – 12:00 Symposium II
Chair: Prof. Mitsuaki Yoshida, Hirosaki University

11:00 – 11:30 Role of NIRS in response to the accident at TEPCO Fukushima Daiichi NPS — from viewpoint of radiation emergency medical preparedness —
Takako Tominaga, Takaya Gotoh, Hideo Tatsuzaki, Misao Hachiya, Nobuyuki Sugiura, Makoto Akashi
National Institute of Radiological Sciences

11:30 – 12:00 IAEA system for responding to radiological and nuclear emergencies and the IAEA response to the accident at Fukushima Daiichi
Pat Kenny
Incident and Emergency Centre,
Department of Nuclear Safety and Security,
International Atomic Energy Agency (IAEA), Austria
12:00 – 14:30  **Lunch Time**

13:30 – 14:20  **Symposium III**

1. **Change in image of radiation by nurses due to nuclear power plant accident**
   Mayumi Urushizaka¹, Yuka Noto², Noriko Ogura³, Maiko Kitajima¹, Yoshiko Nishizawa¹, Tomoko Ichinohe¹, Hideki Yamabe¹
   1) Department of Health Promotion, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
   2) Department of Disability and Health, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
   3) Department of Development and Aging, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

2. **Quality of life and acute radiation damage among breast cancer patients undergoing radiotherapy**
   Noriko Ogura¹, Mayumi Yamauchi², Yuka Noto¹, Yoshiko Nishizawa¹, Hideaki Yamabe¹, Yoichiro Hosokawa¹, Masahiko Aoki², Eriko Kudo³, Yumiko Sato², Harumi Shikanai², Makoto Akashi¹, Yoshiko Ando³, Noriko Kudo³, Yayoi Tsutsumi³, Akiko Nezato³, Yoshiko Fukushima³
   1) Hirosaki University Graduate School of Health Sciences
   2) Hirosaki University School of Medicine and Hospital
   3) Hospital at Research Center for Charged Particle Therapy National Institute of Radiological Sciences (NIRS)

3. **Risk perception of radiation among first-year students of a health sciences school — Differences before and after the March 2011 Fukushima nuclear power plant disaster —**
   Keiko Aizu¹, Chieko Itaki², Toshiko Tomisawa¹, and Ayako Ohgino²
   1) Department of Health Promotion, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
   2) Department of Disability and Health, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

4. **Exploratory study on preparation of public health nurses for a radiation disaster based on the experience of health care in areas affected by the Tokai village nuclear accident**
   Chiaki Kitamiya¹ and Ruriko Kidachi²
   1) Department of Health Promotion, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
   2) Department of Development and Aging, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

5. **Awareness of nuclear accidents among visiting nurses and home caregivers in communities neighboring nuclear plants**
   Daisuke Murakami¹, Ruriko Kidachi², Chikako Yonayama², Haruka Otsu², Yu Kitajima² and Chiaki Kitamiya³
   1) Hirosaki University of Health and Welfare
   2) Department of Development and Aging, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
   3) Department of Health Promotion, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences
6 Current status and issues of radiation emergency medicine education at school of health sciences, Hirosaki University —focus on undergraduate education—
Saichi Wakayama
Department of Development and Aging, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

7 Rehabilitation targeting disuse syndrome in the elder evacuee following radiation exposure accidents: Literature review
Hirokazu Narita, Shuhei Koeda and Hitoshi Tsushima
Department of Development and Aging, Division of Health Sciences, Hirosaki University Graduate School of Health Sciences

8 Morphological study of the thoracic and abdominal organs in X-irradiation mice
Shoji Chiba¹, Takahiro Miura², Fumiya Nakamura³, Koichi Itoh⁴, Kyoko Itoh⁵, Manabu Nakano⁴, Hiroyuki Nozaka⁴, Kohsei Kudo⁵
1) Department of Pathologic Analysis, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Hospital Hakodate Hokkaido
3) Iwate Medical University Hospital
4) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
5) Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

9 The effects of ionizing radiation on pattern recognition receptors
Hironori Yoshino and Ikuo Kashiwakura
Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

10 Characteristic analysis of megakaryocytopoiesis and thrombopoiesis by human hematopoietic stem cells exposed to ionizing radiation
Satoru Monzen¹, Toshiya Nakamura², and Ikuo Kashiwakura¹
1) Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

11 Identification of radiation-induced inflammatory gene network in human umbilical vein endothelial cells
Yuukihiro Furusawa¹, Qing-Li Zhao¹, Seiji Yamamoto², Yuichi Hattori², Takaharu Nomura³ and Takashi Kondo¹
1) Department of Radiological Sciences, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama
2) Department of Molecular and Medical Pharmacology, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama
3) Radiation Safety Research Center, Nuclear Technology Research Laboratory, Central Research Institute of Electric Power Industry
12 Effects of continuous low dose-rate $\gamma$-irradiation on diabetic nephropathy in type II diabetes mellitus model mice
Takaharu Nomura$^1$, Xiao-Han Li$^2$, Hiromitsu Ogata$^3$, Kazuo Sakai$^1$, Takashi Kondo$^4$, Yasuo Takano$^2$ and Junji Magae$^1$
1) Radiation Safety Research Center, Nuclear Technology Research Laboratory, Central Research Institute of Electric Power Industry
2) Department of Diagnostic Pathology, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama
3) National Institute of Public Health
4) Department of Radiological Sciences, Graduate School of Medicine and Pharmaceutical Sciences, University of Toyama

13 Development of immunocompetent lymphocytes in vivo by mixed umbilical cord blood transplantation in mice
Akira Nakano$^1$, Kyoko Ito$^2$ and Koichi Ito$^2$
1) Department of Cellular Biochemistry and Molecular Biology, Division of Biomedical Sciences (Doctoral Course), Hirosaki University Graduate School of Health Sciences
2) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

14 Promising potential of umbilical cord blood cell transplantation in repopulating bacterial-reactive natural IgM-producing B-1a cells
Ayumi Wakayama$^{1,2}$, Kyoko Ito$^1$ and Koichi Ito$^1$
1) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Transfusion and Cell Therapy Center, Hakodate Municipal Hospital

15 Radiation-responsive transcriptome analysis in human hematopoietic cells
Mitsuru Chiba$^1$, Satoru Monzen$^2$, Kosuke Kasai$^3$, Tomisato Miura$^3$, Ikuo Kashiwakura$^4$, Toshiya Nakamura$^1$
1) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Department of Radiological Life Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
3) Department of Pathologic Analysis, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences

16 Analysis of the profile and mechanism of radiation-induced hair loss through examination of protein derived from hair of patients with acute alopecia caused by external radiation exposure
Naoki Nanashima$^1$, Koichi Ito$^1$, Hiroyuki Nozaka$^1$, Takashi Ishikawa$^1$, Manabu Nakano$^1$, Asako Ishizawa$^2$, Honami Narita$^2$ and Toshiya Nakamura$^1$
1) Department of Biomedical Sciences, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Department of Medical Technology, Hirosaki University School of Health Sciences
Optimization of the protein phosphatase-inhibitors (okadaic acid) treatment in premature chromosome condensation (PCC)-ring method for biodosimetry of accidental high dose exposure
Akifumi Nakata¹, Yasuhiro Satoh², Kiyotaka Shibutani², Hiromi Ichikawa², Tomisato Miura¹ and Mitsuaki A. Yoshida¹
1) Department of Radiation Biology, Institute of Radiation Emergency Medicine, Hirosaki University
2) Graduate School of Health Sciences, Hirosaki University

14:30 – 16:30 Symposium IV
Chair: Prof. Takashi Kondo, Toyama University
Prof. Ikuo Kashiwakura, Hirosaki University

14:30 – 14:50 Optimization of calyculin A-induced premature chromosome condensation assay for chromosome aberration studies
Tomisato Miura¹ and William F. Blakely²
1) Department of Pathologic analysis, Division of Medical Life Sciences, Hirosaki University Graduate School of Health Sciences
2) Armed Forces Radiobiology Research Institute (AFRRI), USA

14:50 – 15:20 Radiation mitigators: Compounds, which are effective when administered immediately after exposure to radiations
Kazunori Anzai¹, Megumi Ueno², Ken-ichiro Matsumoto³, Jiro Takata⁴
1) Nihon Pharmaceutical University
2) National Institute of Radiological Sciences
3) National Institute of Radiological Sciences
4) Faculty of Pharmaceutical Sciences, Fukuoka University

15:20 – 15:30 Break

15:30 – 16:00 Biomarkers for Early-response Assessment of Radiation Exposure
Natalia I. Ossetrova, David J. Sandgren, William F. Blakely
Armed Forces Radiobiology Research Institute (AFRRI), USA

16:00 – 16:30 United States Armed Forces Radiobiology Research Institute Countermeasures Program and Related Policy Questions
Radiation Countermeasures Program, Armed Forces Radiobiology Research Institute (AFRRI), USA

16:30 – 16:35 Closing Remarks
Prof. Toshiya Nakamura,
Hirosaki University Graduate School of Health Sciences

16:35 Memorial photography

18:00 – 20:00 Banquet (Hotel New Castle)
Instruction for Speakers

Presentation
The symposiums of The 3rd International Symposium on Radiation Emergency Medicine in Hirosaki University consist of invited talks as well as contributed orals. Foreign and Domestic Invited talks are allocated 20 minutes for presentation plus 10 minutes for discussion. University lecture talks and Regular Oral presentations will be 15 minutes long followed by a 5-minutes discussion.
In a Poster session, please discuss freely.

A window-based PC will be provided in the conference room. We strongly encourage you to use this computer facility. In this case please bring with you the presentation as PowerPoint or PDF on CD or USB stick. If you prefer to use your own PC for your presentation, please inform operation staff in advice.

Proceedings
The proceedings of The 3rd International Symposium on Radiation Emergency Medicine in Hirosaki University will be published from Hirosaki University Press.
Abstract
On Friday, March 11, an earthquake and a tsunami of unprecedented magnitude led to major problems in the stabilization of nuclear power reactors in northeast Japan. Operating reactors were automatically shut down, with control rods inserting into the reactor cores. However, a 14-meter tsunami triggered by the earthquake disabled all AC power to Unit 1, 2, and 3. Fuel tanks for emergency diesel generators were carried off by the tsunami. Water injection failed in the emergency core cooling system of Units 1, 2, and 3.

Since the normal cooling system was not available, a pressure valve was opened to reduce pressure. In spite of such efforts, hydrogen explosions damaged these facilities. Eventually a large amount of radioactive material was released to the environment.

On March 13, members of Radiation Safety Council at Hirosaki University were convened and discussed a policy for this accident. Upon request of the Japanese government, the council concluded that university staff members would be dispatched to Fukushima so as to support their lives.

Main actions we took were as follows:

1. Screening tests for radioactive contamination for the general public in Fukushima Prefecture;
2. Field work of radiation measurement and sampling for restructuring their doses.

Figure 1 illustrates screening test using GM-tube survey meter. In this presentation, an overview of our activities in Fukushima will be introduced.
Symposium I  Masahiro Hosoda

Environmental radiation in Fukushima after nuclear power station accident

Masahiro Hosoda

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Hirosaki University Graduate School of Health Sciences
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A car-borne survey for dose rate in air was carried out in March, April and July 2011 along an expressway passing northwest of the Fukushima Dai-ichi Nuclear Power Station (Fukushima NPS) which released radionuclides into the environment starting after the Great East Japan Earthquake on March 11, 2011, and in an area closer to the Fukushima NPS which was known to have been strongly affected. The following results were obtained.

(1) Temporal variation of dose rate in air by car-borne survey along an expressway

The dose rates along the expressway, i.e. relatively far away from the power station were higher after than before March 11, in some parts by several orders of magnitude.

(2) In-situ gamma spectra

The counts of cesium-134 and -137 at Oshu City, Iwate Prefecture on April 25 were much higher than those on March 19. This implies that there were some additional releases from Fukushima NPS.

(3) Comparison of measured dose rates in air on pavement and uncovered surfaces

The dose rates on the unsealed surfaces were from 1.0 to 2.1 times (average: 1.3 times, n = 20) higher than those on pavement.

(4) External dose estimation within the high level contamination area

As a result of car-borne survey (Figure), the maximum dose rate in air and the cumulative external dose within the high level contamination area were 36 μGy h\(^{-1}\) and 194 mSv, respectively. The maximum cumulative external dose for evacuees who came from Namie Town to evacuation sites (e.g. Fukushima, Koriyama and Nihonmatsu Cities) was estimated to be 68 mSv. The evacuation is justified from the viewpoint of radiation protection.

Figure. Survey route and dose rate distribution map within the high level contamination area.
A severe earthquake occurred in Japan on March 11, 2011. The giant tsunami unleashed by the magnitude 9.0 earthquake devastated many coastal communities across a wide area of eastern Japan and crippled the Fukushima Daiichi nuclear power plant. The battle to bring the situation under control continues. The Japanese government decided to conduct a large-scale survey of residents affected by the nuclear and natural disasters in Fukushima prefecture and requested the dispatch of contamination survey personnel from Hirosaki University. In response to the government’s request, Hirosaki University decided to dispatch a Radiation Exposure Research Team to Fukushima prefecture. In addition, the Fukushima prefecture Temporary Home Visit Project was initiated.

This presentation describes the activities of the Radiation Exposure Research Team and the Temporary Home Visit Project. Next, based on my experience of the 2 projects described above, I would like to talk about the characteristics of the Fukushima nuclear accident and what I feel is important in terms of the care of residents affected by this nuclear disaster.

This unprecedented nuclear disaster will leave a large scar within Fukushima and throughout Japan. Our role, as medical staff in the field of radiation medicine, is to protect the health of those affected and try to reduce their anxiety. In addition, as the persons responsible for implementing the Radiation Emergency Medicine Project, it is also necessary that we educate and foster the development of medical care personnel with the appropriate knowledge and skills to carry out the measures required.
In January 2010, National Institute of Radiological Science (NIRS) has established the Radiation Emergency Medical Assistance Team (REMAT) to support primary medical care when an accident of radiation exposure or contamination with radioactive materials has occurred overseas. The team consists of physicians, nurses, radiation protection experts, and health physicists ready to respond to radiation emergencies, and the team is equipped with advanced portable radiation measuring instruments and medicines. NIRS is ready for providing international medical assistance for radiation emergencies, based on human resources with experiences in this field. However, the first REMAT activity was made for the TEPCO Fukushima Daiichi Nuclear Power Station (NPS) accident. Before NIRS established REMAT, we had a system for dispatching staffs to an accident site in Japan based on the Act on Special Measures Concerning Nuclear Emergency Preparedness. However, this system was for providing advice to local staff and the activities were limited.

The Great East Japan Earthquake attacked the Pacific coast area of eastern Japan on 11 March 2011 and this earthquake and tsunami caused enormous damage to the NPS of Tokyo Electric Power Co. (TEPCO). NIRS dispatched REMAT to the local headquarters located 5 km from the NPS almost 17 hours after the earthquake. Since then, many experts at NIRS have been sent to Fukushima. REMAT is still being involved in responses to this nuclear accident including the public issue in Fukushima. Thus, this nuclear accident requires response at an unprecedented scale and over a lengthy period.

In this accident, the response system for radiation emergency medicine did not work effectively since community lifelines such as water supply and electricity were severely damaged. Thus, hospitals lost their function including radiation emergency medicine. In our efforts for recovery from the damages, reconstruction of the medical system in the affected areas has to be hurried. Moreover, reestablishment of the system for radiation emergency medical response is also a key point since reactors have not been stabilized. From our response to this combined disaster of earthquake, tsunami, and radiation, we have learned that there is an urgent need for all-hazard approaches.
IAEA system for responding to radiological and nuclear emergencies and the IAEA response to the accident at Fukushima Daiichi

Pat Kenny

Incident and Emergency Centre, Department of Nuclear Safety and Security, International Atomic Energy Agency, Austria
Change in image of radiation by nurses due to nuclear power plant accident

Mayumi Urushizaka¹, Yuka Noto², Noriko Ogura³, Maiko Kitajima¹, Yoshiko Nishizawa¹, Tomoko Ichinohe¹, Hideki Yamabe¹

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[Purpose] Many people are concerned about radiation exposure due to the accident at the nuclear power plant caused by the Great East Japan earthquake that occurred on March 11, 2011. The aim of this study is to elucidate the change in image of radiation by nurses between before and after the disaster.

[Method] The subjects for the study were 307 nurses. A placement method using a questionnaire was conducted in February 2010 (before the disaster) and again in May through June 2011 (after the disaster). They answered questions regarding their personal details and their image scale of radiation. The scale was composed of 3 factors including 17 items: affectivity, usefulness and certainty, and each of the 17 items use the semantic differential methods with a 7-stage response scale. In this questionnaire a low score indicates a negative image and a high score indicates a positive image. We explained the aim of this study to the subjects and the anonymous and voluntary nature of their participation. The analysis conducted was a two-sample t-test and ANOVA, and the significance level was set at p < 0.05.

[Results] There were 262 responses (88 before the disaster, 174 after the disaster) and the collection rate was 85.3%. The scores for all items were significantly lower after the disaster than before it. It was determined that there was no relationship between the image score and the personal details of the nurses. Although a group who had attended in seminars on radiation before the disaster showed a significantly higher score than the group who had not in regards to the item “dangerous – safe” before the disaster, the difference diminished between both groups after the disaster.

[Conclusion] Nurses had a more negative image of radiation after the disaster than before it. In spite of attendance in seminars on radiation, the item “dangerous-safe” tended towards a more dangerous image after the disaster and this may be due to people thinking the impact of the nuclear power plant accident was so serious.
Quality of life and acute radiation damage among breast cancer patients undergoing radiotherapy

Noriko Ogura¹, Mayumi Yamauchi², Yuka Noto¹, Yoshiko Nishizawa¹, Hideaki Yamabe¹, Yoichiro Hosokawa¹, Masahiko Aoki², Eriko Kudo², Yumiko Sato², Harumi Shikanai², Makoto Akashi³, Yoshiko Ando³, Noriko Kudo³, Yayoi Tsutsumi³, Akiko Nezato³, Yoshiko Fukushima³

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3) Hospital at Research Center for Charged Particle Therapy National Institute of Radiological Sciences (NIRS)
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[Purpose] Radiotherapy is an effective method for cancer treatment. However, radiation harms normal cells and tissues, therefore patients may get distress. The purpose of this study is to clarify the patients’ QOL and symptoms of acute radiation damage in breast cancer patients undergoing radiotherapy.

[Methods] Subjects were 47 breast cancer patients treated with radiotherapy at Hirosaki University School of Medicine and Hospital. The patients were asked to record symptoms using a symptom diary from the day before the start of radiotherapy until its completion. The recorded symptoms were classified into Grade 1 to Grade 5 according to the Common Terminology Criteria for Adverse Events v3.0, Japanese translation JCOG/JSCO. QOL by the SF-8™ standard version was collected before irradiation, after the completion of irradiation, and 4–6 weeks after irradiation. Clinical data were obtained in a similar fashion as the QOL data.

[Results] The symptoms tended to appear from around the third week of irradiation. It is clear that fatigue and pain tended to appear at an early stage. After completion of irradiation, the QOL scores increased in all eight domains, but the scores 4–6 weeks later did not show major changes from the completion of irradiation. Symptom scores during irradiation were negatively correlated with QOL scores. Lymphocyte and platelet counts decreased in the third through fifth weeks, but other clinical measures remained stable.

[Conclusion] It became clear that QOL scores did not decrease with radiotherapy. However, there was a negative correlation between the symptoms during irradiation and QOL. Therefore, in order to improve QOL of patients during radiotherapy, nursing intervention is necessary to abate these symptoms.
**Risk perception of radiation among first-year students of a health sciences school-Differences before and after the March 2011 Fukushima nuclear power plant disaster**

*Keiko Aizu¹, Chieko Itaki², Toshiko Tomisawa¹, and Ayako Ohgino²*

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**[Purpose:]** It is important in risk communication that those engaged in medical treatment have the right perception about radiation. In order to educate students effectively, it is important to grasp a student's interest. The aim of this study was to clarify the differences in radiation risk perception of first-year students entered in a Hirosaki University School of Health Sciences before and after the March 2011 Fukushima nuclear power plant disaster.

**[Methods]** The subjects were first-year students enrolled in the Hirosaki University School of Health Sciences. A questionnaire survey was conducted in 196 students who entered the school in 2010 (2010 group), and 199 students who entered in 2011 (2011 group). The survey consisted of eight questions about the perception of radiation, one question about radiation risk in general, and one question about interest in radiation. Data analyses were done using the chi-square test, the Mann-Whitney test, and Spearman's rank correlation coefficient.

**[Results]** In regard to the image of radiation, the items of "contamination" and "nuclear power generation" scored significantly higher as radiation-associated terms in the 2011 group than in the 2010 group. About the risk of radiation danger, the 2011 group had a significantly lower perception on six items, for example, about receiving radiotherapy. In the 2010 group, the items of "fearfulness" and "interest" about radiation showed a significant positive correlation.

**[Conclusion]** While the first-year students in the 2011 group entered the school after the nuclear power plant accident in Fukushima and had a high perception of the fearfulness of radiation, they had a low perception of radiation risk in a medical sense. It seemed that radiation related to medicine and radiation of nuclear power plants are recognized as separate things. It is necessary to examine and tailor educational curriculums according to the current student perception of radiation.
Exploratory study on preparation of public health nurses for a radiation disaster based on the experience of health care in areas affected by the Tokai village nuclear accident

Chiaki Kitamiya\textsuperscript{1} and Ruriko Kidachi\textsuperscript{2}

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[\textbf{Purpose}] We attempted to reconstruct the experience of public health nurses who responded to a radiation disaster based on an analysis of their narrative accounts of the Tokai Village JCO nuclear accident.

[\textbf{Methods}] This study was done through half-structured interviews. It was approved by The Committee of Medical Ethics of Hirosaki University Graduate School of Medicine, Hirosaki, Japan.

[\textbf{Results}] Interviews were conducted at Higasidouri Village in Aomori on November 8, 2010 and at Tokai Village, Naka City, Hitachinaka City, Hitachi-Oomiya City, Ibaraki prefectural office, Ibaraki Hitachinaka Health Center, and Ibaraki Hitachi-Oomiya Health Center in Ibaraki on November 24 and 25, 2010. The average interview lasted 30 minutes to 1.5 hours. In this analysis, an interview of three cities surrounding Tokai village was held data. It is qualitative analysis about these three interviews. As a result, three categories were extracted from nine subcategories.

Three of these categories were (1) Activity for mitigating residents' anxiety, (2) Mental attitude toward a radiation disaster, and (3) Training of public health nurses as members of the local government.

[\textbf{Discussion}] By continuing training based on the experience gained in the JCO accident, this experience will not be wasted, and we will be able to prepare inexperienced persons who may be called on to respond to future emergencies. The data we gathered on the JCO disaster established the necessity for prioritizing mitigation of residents' anxieties.
Awareness of nuclear accidents among visiting nurses and home caregivers in communities neighboring nuclear plants

Daisuke Murakami¹, Ruriko Kidachi², Chikako Yonaiyama², Haruka Otsu², Yu Kitajima² and Chiaki Kitamiya³

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[Background] In Japan, the protocols for responding to a nuclear accident are established in the Fundamental Disaster Relief Act. At the prefecture and municipality levels, there are established regional disaster plans, and disaster drills are also conducted. However, individuals require assistance during disasters is not concretely established in any of these forms of preparation. Therefore, visiting nurses and home caregivers should play an active role in their establishments. Studies on natural disasters have reported the importance of collaboration among various institutions and individuals. Visiting nurses and home caregivers are key players in disaster response because they know the location of people who require assistance during disasters and the point of assistance required during evacuation.

[Purpose] This study was conducted to clarify whether visiting nurses and home caregivers working in prefectures with atomic facilities are aware of their responsibilities in the event of a nuclear accident and what measures they would take in identifying those who might need assistance in the event of a nuclear accident.

[Methods] Subjects were employees at visiting nurse stations and home care helper stations located near nuclear plants in prefectures with atomic facilities, to target the 10 prefectures (Hokkaido, Aomori, Niigata, Ishikawa, Ibaraki, Fukui, Ehime, Shimane, Saga, and Kagoshima) except Miyagi, Fukushima, and Shizuoka. Information was collected by a self-administered questionnaire. Requests to participate in the survey were sent to office directors, and consent was obtained along with an agreement on how many questionnaires would be completed. The requisite number of copies of the questionnaire was then sent to the director, and employees at the office were asked to complete and mail in the questionnaires. The survey began on April 22, 2011 and will continue until March 31, 2012.

[Results] Consent was obtained from 13 of 192 specialty facilities (6.78%). Ninety questionnaires were dispatched, and 20 (22.22%) of them have been returned. Three were invalid. The surveys indicate that interest in radiation is high but that there is insufficient knowledge. Learning opportunities are not sufficient. When asked that what associate from word of "disaster", the most respondents answered, "the earthquake". And when asked that about "nuclear accidents", the most respondents answered "the nuclear power plant accident".

[Study status] This study was approved by the research ethics committee of Hirosaki University. Data collection began in June 2011. Questionnaires are still being collected.
[Introduction] The Radiation Emergency Medicine (REM) education subject "Introduction to Basic Radiation" at Hirosaki University started in April 2010. In 2011, started May because of the 2011.3.11 Earthquake East Japan and nuclear disaster at Fukushima. This subject is aimed at paramedical personnel to provide them with basic knowledge related to radiation protection and an outline of radiation emergency medicine.

The purpose of this study is to clarify issues with the current state of undergraduate education in this subject.

[Methods]
The following survey items were analyzed. The numbers of attended students, test results, awareness and opinion of the course starts about radiation, opinions on this subject at the end of class test.

[Result] Numbers of students taking this subject were 161 in School of Health Sciences, and the other students were 4 in other Faculties. All students passed this subject. Last day of the test results, the average was 81.8 points, SD8.6 at test. In the beginning of the class, awareness and opinion of the radiation were “scary” 20%, “atomic bomb of Hiroshima and Nagasaki” 42%, “Nuclear power plant” 33%, “Accident of JCO” 2%, “nothing special” 4% (N=153). The opinions on this subject at the end of class test were positive for example “important of duplication of course contents”, “understanding basic radiation and REM”, “what could be interested as necessary”. Negative opinions were “duplication of course contents”, “learn more”, “using radiation measuring devices”.

[Discussion] In 2010, the undergraduate Radiation Emergency Medicine Education subject "Introduction to Basic Radiation" at Hirosaki University had started. The survey results are increasingly understanding of the classes, and we obtained a positive opinion to the contents. As issues, duplication of education, course content and study methods, and the like ready to create the text were obtained.

In the class of 2011, students will be affected by nuclear disaster at Fukushima, and expected to change the consciousness and awareness about health and radiation exposure.
Rehabilitation targeting disuse syndrome in the elder evacuee following radiation exposure accidents: Literature review

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We considered the role of rehabilitation in radiation emergency medicine, in particular the rehabilitation of patients exposed to large doses of radiation in accidents such as the Tokaimura JCO criticality accident. Following a literature review, we determined that rehabilitation focused on radiodermatitis and respiratory management is especially important in this context. We are now building an experimental system using an animal model to investigate the effects of rehabilitation on large doses radiation-exposed patients.

On the other hand, following the nuclear accident at Fukushima nuclear power plant No. 1, triggered by the Great East Japan Earthquake of March 11, 2011, many individuals were forced to live in evacuation centers. Because of long-term stay in these evacuation centers, health problems such as disuse atrophy occurred in the frail elderly. Therefore, we decided to explore the role of rehabilitation for elderly evacuees as one of radiation emergency medicine. In this study, we reviewed reports on the rehabilitation offered to evacuees in the past disaster, considered the symptoms targeted for rehabilitation, and examined its effects.

We found that the symptoms targeted for rehabilitation included disuse syndrome and deep vein thrombosis (DVT). Inactivity caused by stay at evacuation centers was the main cause of these complications. The elderly were known to be at high risk, and prevention was seen as most important. For prevention of disuse syndrome and DVT, exercise of lower limbs was especially important, and the effect for the evacuee was demonstrated. However, because in DVT-affected persons, a clot isolated during exercise might lead to pulmonary thromboembolism, we thought that rehabilitation should be undertaken with the cooperation of a doctor and others providing care to the evacuees. Additionally, in nuclear-power disasters such as those at Fukushima, we determined that conveying correct information about radiation to evacuees and reducing their uneasiness are indispensable.
Morphological study of the thoracic and abdominal organs in X-irradiation mice

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[Introduction] The present study was conducted to confirm previously documented morphological damage from X-irradiation in the thoracic and abdominal organs, especially the digestive organs.

[Materials and Methods] The whole bodies of 7-week-old ICR mice (2 males and 2 females) were exposed to 2, 4, 8, and 10 Gy of X-irradiation using a Hitachi MBR-1505R2 at a dose rate of 0.133 Gy/min. At the 10th day after X-irradiation, the mice and their organs were stored in 10% formalin. We measured the weights of the lungs, heart, liver, spleen, and urinary bladder, and obtained photographs of the digestive organs. The above-mentioned organs and digestive organs were cut into 5-μm sections, stained using HE, and observed under a light microscope.

[Results] The size of the spleen diminished in accordance with the strength of the X-irradiation dose, and the spleen weight at 8 and 10 Gy in male and female mice was decreased markedly in comparison with that of the non-exposed mice (P < 0.05). The weights of the remaining organs were similar between non-exposed and exposed mice. On macroscopic investigation, both male and female mice that received 8 and 10 Gy showed bleeding in the digestive organs. On histological examination, bleeding was found in the stomach, duodenum, jejunum, and ileum at 4, 8, and 10 Gy; of the cecum at 10 Gy; and of the sigmoid colon at 8 and 10 Gy. No differences were found in the morphological appearance of the intestinal glands and villi between the non-exposed and exposed mice. Lymphocyte number in the lymphoid nodules of the ileum and sigmoid colon decreased at 2 Gy. Lymphocyte number in the spleen decreased in proportion to the strength of X-irradiation dose; this decrease was obvious at 8 and 10 Gy. In the lungs, bleeding in the alveoli and substantial hypertrophy of the alveoli were found at 4, 8, and 10 Gy. In the urinary bladder, bleeding was found at 2, 8, and 10 Gy. In tissues of the heart, liver, and pancreas, morphological differences were not observed between the non-exposed and exposed mice.

[Discussion] X-irradiation causes a decrease in lymphocyte number in the spleen and lymphoid nodules of the small and large intestines. The size and weight of the spleen were obviously diminished in inverse proportion to the X-irradiation strength. A 10-Gy dose of radiation is a critical value; exposure at this level influences the growth of the intestinal glands and villi.
The immune system is composed of innate and adaptive immunity. Antigen presenting cells (APCs), such as dendritic cells and macrophages, serve as a link between innate and adaptive immunity. APCs express pattern recognition receptors (PRRs) which recognize pathogen-associated molecular patterns (PAMPs). Toll-like receptor (TLR) and retinoic-acid inducible gene-I (RIG-I) are well-studied PRRs that play important roles in anti-bacterial or anti-viral immunity. The activation of APCs through PRRs is required for the induction of adaptive immune responses. However, it remains unknown whether ionizing radiation affects PRRs. The effects of ionizing radiation on the expression of PRRs and the response against PAMPs were herein investigated using THP1 cells (human acute monocytic leukemia cells). THP1 expressed TLR2 and TLR4 which are receptors for peptidoglycan and lipopolysaccharide (LPS), respectively. Those expressions after X-ray irradiation (1-5 Gy) were higher in irradiated cells than in non-irradiated cells. The response against LPS was estimated by the induction of tumor necrosis factor-\(\alpha\) (TNF-\(\alpha\)). The TNF-\(\alpha\) positive cells were higher in irradiated-THP1 (2%) than in non-irradiated cells (0.5%). To investigate the effects of ionizing radiation on PRRs in detail, THP1 cells were treated with phorbol 12-myristate 13-acetate (PMA) in order to differentiate into macrophage-like cells, and then the similar experiments were performed. In contrast to the results of THP1, the expression of TLR4 of PMA-treated THP1 after irradiation was lower in irradiated cells than in non-irradiated cells. Although TNF-\(\alpha\) positive cells after LPS stimulation was higher in PMA-treated THP1 (40%), no significant difference in the response of LPS was observed between non-irradiated and irradiated conditions. We next investigated the expression of RIG-I which recognizes double-strand RNA. Although non- or PMA-treated THP1 did not express RIG-I, RIG-I was expressed in PMA-treated THP1 after LPS stimulation. No significant effects of irradiation on the RIG-I expression in PMA-treated THP1 after LPS stimulation was observed. In conclusion, this study demonstrated that ionizing radiation affects PRRs expression and the response of PRRs to PAMPs, but these effects depend on the cell types and differentiation state.
Hematopoietic processes, especially megakaryocytopoiesis and thrombopoiesis, are highly sensitive to extracellular oxidative stresses such as ionizing radiation and chemotherapeutic agents. This study examined the terminal maturation of megakaryocytes and platelet production in hematopoietic stem/progenitor cells (HSPCs) irradiated with ionizing radiation. Highly purified CD34+ cells derived from human placental/umbilical cord blood were exposed to X rays (2 Gy, 150 kVp, 20 mA; 0.5-mm aluminum and 0.3-mm copper filters), at a dose rate of approximately 1 Gy/min, and then cultured in a serum-free medium supplemented with thrombopoietin and interleukin-3. The number of cells generated from X-irradiated CD34+ cells decreased with time in the culture. However, the fraction of CD34+Tie-2+ and CD41+Tie-2+ cells among the total cells generated from X-irradiated cells increased significantly in comparison to non-irradiated controls on day 7. In addition, the CD42a+ particles, which appeared to be platelets, generated from the X-irradiated HSPCs appeared to be normal. Quantitative real-time reverse transcriptase-polymerase chain reaction analysis of the expression of various genes in cells harvested from the cultures showed that the early hematopoiesis-related genes FLI1, HOXB4, and Tie-2, the cytokine receptor genes KIT and IL-3RA, and the oxidative stress-related genes HO1 and NQO1 were up-regulated on day 7. These results suggest that normal terminal maturation of megakaryocytes and platelet production occur in residual HSPCs after ionizing radiation, despite the adverse effect of radiation on proliferation and differentiation of HSPCs. Ionizing radiation may have the potential to promote both megakaryocytopoiesis and thrombopoiesis.
Identification of radiation-induced inflammatory gene network in human umbilical vein endothelial cells

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Cardiovascular disease has been considered as a major health-risk factor after radiation exposure such as A-bomb survivors. In a trial to understand the molecular mechanisms underlying the inflammatory reaction frequently encountered in vascular system post exposure to ionizing radiation, we carried out global scale microarray and computational gene expression analyses in human umbilical endothelial cells (HUVEC). Global scale microarray gene expression analysis of irradiated HUVEC (2.5 Gy) identified 1128 genes that were up- or down-regulated by a factor 1.8 or greater at 6, 12, or 24 hr after irradiation. Hierarchical cluster analysis of the differentially expressed genes identified four clusters. The bioinformatic analysis using ingenuity pathway analysis tools revealed that the down-regulated genes in cluster I were associated with cell cycle regulation whereas the up-regulated genes in cluster IV were associated with inflammatory responses. The analysis also identified a gene network containing interferon response factor 7 (IRF7) and its transcriptional target interferon-induced transcripts (IFITs) and Mx1, which have been known to be associated with inflammation in endothelial cells. The up-regulated genes and the gene network identified here may explain the inflammatory response induced by X-irradiation. These findings uncover some of the molecular basis of the mechanism(s) of the inflammatory disorder in response to X-irradiation in human umbilical vein endothelial cells.
Effects of continuous low dose-rate $\gamma$-irradiation on diabetic nephropathy in type II diabetes mellitus model mice

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We previously showed that continuous exposure of mice to low dose-rate $\gamma$-irradiation enhances antioxidant activity. Here, we study the ameliorative effect of continuous whole-body irradiation with low dose-rate $\gamma$-rays on diabetic nephropathy. Ten-week-old, female db/db mice, an experimental model for type II diabetes, were irradiated with low dose-rate $\gamma$-rays from 10 weeks of age throughout their entire lives. Nephropathy was studied by histological observation and biochemical analysis of serum and urine. Antioxidant activities in kidney were biochemically determined. Continuous low dose-rate $\gamma$-irradiation significantly increases lifespan in db/db mice. Three of 24 irradiated mice were free of glucosuria after 80 weeks of irradiation. Histological studies of kidney suggest that low-dose irradiation increases the number of normal capillaries in glomeruli. Antioxidant activities of superoxide dismutase, catalase, and glutathione are significantly increased in kidneys of irradiated db/db mice. Continuous low dose-rate $\gamma$-irradiation ameliorates diabetic nephropathy and increases lifespan in db/db mice through the activation of renal antioxidants. These findings have noteworthy implications for radiation risk estimation of non-cancer diseases.
Development of immunocompetent lymphocytes in vivo by mixed umbilical cord blood transplantation in mice

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[Introduction:] At the 2nd International Symposium on Radiation Emergency Medicine in Hirosaki University last year, we reported that mixed umbilical cord blood cell (UCBC) transplantation has a high success rate and induces development of phenotypically mature lymphocytes in lethally X-ray-irradiated recipient mice. However, the extent to which the lymphocytes derived from the mixed UCBC transplant recover their immune function is still unclear. In this study, we evaluated the functional maturity of T and B lymphocytes reconstituted by mixed UCBC transplantation in recipient mice.

[Materials and Methods] C57BL/6 (B6) (H-2ª) female mice X-ray-irradiated with 9 Gy were transplanted with 3 different combinations of mixed UCBCs: (1) green fluorescent protein (GFP)-Tg B6 (H-2ª) and C3H (H-2ª), (2) GFP-Tg B6 (H-2ª) and BALB/c (H-2ª), and (3) C3H (H-2ª) and BALB/c (H-2ª). At 16 weeks after transplantation, all recipients received skin grafting to assess the functional maturity of the newly developed T and B lymphocytes.

[Results] Flow cytometric analysis showed that cells of all lineages were reconstituted by GFP’ hematopoietic stem cell (HSC) and B6 recipient’s own X-ray-resistant HSC in almost all recipients of combinations (1) and (2), and combination (3), respectively, suggesting predominant engraftment of major histocompatibility complex (MHC) antigen-matched HSCs. These recipients rejected third-party skin grafts from BALB/c (H-2ª) and C3H (H-2ª) mice but not from B6 mice. Furthermore, they produced antibodies only against MHC antigens on the rejected skin grafts, indicating the presence of functional T and B lymphocytes.

[Conclusion] Our observations indicate that T and B lymphocytes derived from mixed UCBCs transplants are immunologically fully competent with the ability to distinguish self from non-self using different MHC antigens. Currently, we are evaluating the selective mechanism of MHC-matched HSCs in the recipients’ bone marrow.
Promising potential of umbilical cord blood cell transplantation in repopulating bacterial-reactive natural IgM-producing B-1a cells

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[Introduction]: Hematopoietic stem cell transplantation is an effective treatment for accidental high-dose total-body radiation exposure. Although umbilical cord blood cells (UCBCs) and bone marrow cells (BMCs) essentially have a similar ability of recovering the hematopoietic system, it remains unclear whether UCBC and BMC transplantation reconstitute all immune cells. B cell population is composed mainly of 3 different subsets: B-1a, B-1b, and B-2 cells. Among these, B-1a cells spontaneously secrete natural IgM antibodies against bacteria and viruses localized primarily in the peritoneal and pleural cavities. In this study, we compared the extent of B-1a cell reconstitution between UCBC and BMC transplantation.

[Materials and Methods] RAG2−/− C57BL/6 (B6) [H-2b] recipient mice were lethally irradiated with 9 Gy and immediately given a transplant of syngeneic UCBCs or BMCs obtained from green fluorescent protein (GFP)-transgenic B6 mice. At 16 weeks after the transplantation, reconstitution of peritoneal B-1a cells in the recipients was detected by multicolor staining with anti-CD45R/B220, anti-CD5, anti-IgM, and anti-CD11b antibodies. Furthermore, production of natural IgM antibodies from B-1a cells in the recipient mice was measured by enzyme-linked immunosorbent assay before and after immunization with bacterial-derived polysaccharide (Pneumovax).

[Results] Flow cytometric analysis revealed that the number of GFP-positive peritoneal B-1a cells was relatively high in UCBC recipients and low in BMC recipients. Bacterial polysaccharide-reactive IgM antibodies were produced predominantly in UCBC recipients, indicating that IgM production is correlated with the presence of B-1a cells.

[Conclusion] Our results indicate that UCBCs have promising potential in the reconstitution of functional natural IgM-producing B-1a cells involved in front-line innate defense. This finding is clinically valuable for hematopoietic stem cell transplantation.
Symposium III  Poster presentation 15  Mitsuru Chiba

Radiation-responsive transcriptome analysis in human hematopoietic cells

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[Introduction] Ionizing radiation (IR) causes DNA injury and induces multiple signal mechanisms, including the regulation of DNA repair, cell cycles, and gene expression by the activation of p53-related pathways. Natural antisense transcripts (NATs) are recognized as important regulators of gene expression in eukaryotic cells, but the effects of IR on NAT expression is unknown.

[Materials and Methods] We investigated the gene expressions of sense transcripts (mRNA) and NATs after X-ray irradiation in a human B lymphoblast cell line (IM-9) using sense/antisense custom-microarray and strand-specific RT-qPCR. The intracellular localization of these RNAs was examined using the nuclear and cytoplasmic fraction RNAs.

[Results] Sense/antisense microarray analysis showed that the mRNA expression of 93 genes and NAT expression of 24 genes were upregulated more than 1.5-fold in irradiated samples (1, 2, and 4 Gy) as compared to non-irradiated samples (0 Gy). Among the mRNA of 93 genes, the cis-NAT expression genes, which transcribed from the opposite DNA locus of the gene, were 5 genes, including the MDM2 and CDKN1A genes. The mRNA and cis-NATs of these 5 genes were upregulated by X-ray exposure in a dose-dependent fashion. The mRNA and cis-NATs of MDM2 and CDKN1A were also found to upregulated in proportion to X-ray exposure by strand-specific RT-qPCR. Analysis of intracellular localization showed that the cis-NATs of MDM2 were localized in the cytoplasm, and cis-NATs of CDKN1A were localized in the nucleus and cytoplasm.

[Discussion] In the present study, radiation-responsive NATs were identified for the first time. MDM2 and CDKN1A were the p53-related genes involved in DNA repair and cell cycle checkpoint, and it would be expected that these cis-NATs regulate the gene expression of mRNA post-transcription. In the future, IR dose-dependent upregulation of these mRNA and cis-NATs may be used as a biomarker for evaluating DNA injury.
Analysis of the profile and mechanism of radiation-induced hair loss through examination of protein derived from hair of patients with acute alopecia caused by external radiation exposure

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[Introduction] A rapid, non-invasive screening test is needed to evaluate the effects of radiation exposure following radiation accidents, acts of nuclear terrorism, or extensive radiation disasters. Acute alopecia is a recognized consequence of radiation exposure, but the mechanism by which radiation causes acute alopecia and chemical changes in hair protein is unknown. Therefore, we analyzed the mechanism underlying acute alopecia following radiation exposure and examined whether hair protein could be used as a new biomarker for evaluating radiation exposure.

[Materials and Methods] Six-week-old, male C57/BL6 mice were exposed to 6 Gy of X-irradiation, and their hair and skin were collected as they were shed. Hair protein was extracted from hairs of mouse with 8 M urea. SDS-PAGE liquid chromatography/electro-ionization mass spectrometry (LC/ESI-MS) was used to analyze the altered biochemical character of the hair protein. Protein localization was detected by immunohistochemistry.

[Results] The hair of irradiated mice turned white and were shed. To examine whether hair protein profiles are different between irradiated mice and controls, proteins extracted from the hair of each mouse were subjected to SDS-PAGE. Staining with Coomassie Brilliant Blue dye revealed 2 major bands at 51 kDa and 40 kDa; expression of these proteins was not different between control and irradiated mice. To identify these bands, samples recovered from SDS gels were subjected to LC/EMS-MS analysis. There was no change in the composition of major hair keratins, such as Krt81, Krt83, and Krt86, but the expression of Krt15, which is a marker of hair stem cells, was detected in control mice but not the mice with alopecia. Krt1, Krt5, Krt10, Krt14, and Krt78 were detected in the hair of mice with alopecia. Real-time quantitative PCR showed that the expression of the CD34 gene, which is a marker of hair follicle stem cells, decreased to 50% of its previous level. Hair follicles of control mice showed a positive reaction to anti-CD34 antibody, but hair follicles of mice with alopecia had a negative response.

[Discussion] Expression of Krt15 and CD34 was absent in mice with alopecia; this suggests that hair follicle stem cells were damaged, causing changes in the keratin profile. Therefore, cytokeratins such as Krt1, Krt10, and Krt15 might be used as new biomarkers of exposure to external radiation.
Optimization of the protein phosphatase-inhibitors (okadaic acid) treatment in premature chromosome condensation (PCC)-ring method for biodosimetry of accidental high dose exposure

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The dicentric chromosome assay (DCA) is very important for dose estimation in acutely irradiated person. However, this method is not available for the case of high dose exposure to ionizing radiation over the lethal dose. Because, the mitotic delay and apoptosis resulting in poor mitotic index are induced in lymphocytes of such highly irradiated cases. In order to estimate accurately the radiation dose in high dose exposure, the premature chromosome condensation (PCC) method which induces the DNA condensation in the interphase cell has been developed and a ring chromosome is recommended as chromosome aberration used for biodosimetry in the cases of high dose radiation exposure. Okadaic acid and calyculin A which are protein phosphatase inhibitors can induce PCC in any phase of the cell cycle. However, the mechanism of PCC and the influence of these reagents on the chromosome condensation have not been elucidated sufficiently yet. The aim in the present study is to optimize the okadaic acid treatment which induces PCC for biodosimetry in the case of accidental high dose exposure. We investigated the frequency of PCC and the degree of chromosome condensation in both non-irradiated and X-ray irradiated lymphocytes by various treatment time and concentration of okadaic acid. It was found that the frequency of PCC in the cell irradiated by 10 Gy of X rays was lower than that in non-irradiated cells and the optimized condition was one hour treatment with 1000 nM of okadaic acid. Furthermore, the highly condensed chromosomes were frequently identified according to the increase of the concentration and treatment time of protein phosphatase inhibitors. These results in the present study indicate that the protein phosphatase inhibitors have some influences on the chromosome morphogenesis.
Optimization of calyculin A-induced premature chromosome condensation assay for chromosome aberration studies

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Calyculin A (CA)-induced premature chromosome condensation (PCC) assay is a simple and useful method to assess structural chromosome aberrations in interphase cells. PCC assay is the recommended biodosimetry method, vice the gold-standard lymphocyte mitogen-stimulated metaphase-spread dicentric and ring assay (DCA), for use in cases of high-dose (>5 Gy) and partial-body exposures. Treatment with CA (50 nM) for 60 min, which is typically used in these studies, however, results in the induction of fuzzy and shortened chromosomes. In this study, the effect of the CA exposure on chromosome morphology and PCC frequency was investigated in a human peripheral blood ex vivo irradiation (60Co-γ rays; ~0.6 Gy/min) model. Treatment with CA for 60 min increased the frequency of G2/M-PCC cells with shortened chromosomes by 71.7 ± 21.5-fold at dose range of 0 – 3 Gy compared with 15- or 30-min treatment. The frequency of G2/M-PCC cells with fuzzy chromosomes in 60-min treated group was also increased by 3.0 ± 0.5-fold and 1.9 ± 0.2-fold of the value obtained from the 15- and 30-min treated group, respectively. The G2/M-PCC scoring index in 60-min treated group was decreased by 44.3 ± 5.6% and 42.7 ± 6.0% of the values in 15- and 30-min treated groups, respectively. The G2/M-PCC efficiency of 30-min treated group was highest in the three conditions of CA exposure to obtain scorable PCCs, those without fuzzy or shortened chromosomes. We conclude that CA treatment for 30 minutes before harvesting is optimum for chemical induced-PCC assay in PHA-stimulated PBL ex vivo radiation model.

Funded by AFRRI, grant number: RAB4AM
Radiation mitigators: Compounds, which are effective when administered immediately after exposure to radiations

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We can classify the compounds that modulate radiation injuries to three categories with respect to the timing of administration: the compounds for prophylaxis, mitigation, and treatment of radiation injuries. Most of radiation protectors so far reported are for prophylaxis and should be present in the appropriate position in the living body before exposures. In contrast, for countermeasure to accidental overexposure, it is necessary to use the compounds that are effective when administered after exposure. These compounds are called mitigator and relatively few agents have been reported so far. Here we will show several mitigators we found. One is heat-treated mineral yeast, especially Zn-overloaded one called Zn-yeast. When mice were administered i.p with Zn-yeast immediately after whole-body exposure to 7.5 Gy X-rays, the 30-day survival rate was increased from 0% (control group) to more than 90%. The dose reduction factor of Zn-yeast (100 mg/kg) was about 1.2. Another type of radiation mitigator is a water-soluble vitamin E analog, tocopherol-mono-glucoside (TMG). When TMG (650 mg/kg) was administered intraperitoneally (i.p.) to C3H mice immediately after whole-body exposure to 7 Gy radiation, the 30-day survival was significantly higher than that of the control mice. The i.p. administration of TMG at 4 h after irradiation significantly improved survival compared to that of the controls, but administration 8 h after irradiation did not have a significant effect. Recently we have found that another water-soluble vitamin E analog, γ-TDMG, is a more potent radiation mitigator. Several results on γ-TDMG will be presented, too.
Biomarkers for Early-response Assessment of Radiation Exposure

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Acts of nuclear or radiological terrorism could expose large numbers of people to ionizing radiation. Early treatment of populations exposed to ionizing radiation requires accurate and rapid biodosimetry with a precision as high as possible to determine an individual’s risk for life-threatening exposures. Early-phase and tissue-specific (i.e., parotid glands, skin, bone marrow, liver, small bowel, etc.) protein biomarkers detected in peripheral blood can provide rapid estimation of an individual’s exposure dose as well as diagnostic information of organ-specific radiation injury. Their combination with hematological and blood chemistry changes has potential triage biodosimetry applications to help in early-phase medical management of radiation accidents. Development and validation of radiation injury biomarkers are critical for triage as well as to assess efficacy of treatment and follow-up of irradiated individuals. These biomarkers need to be validated in suitable radiation model systems. We established animal (i.e., Mus musculus, Rhesus macaque) radiation models to transition this research platform for application to humans. We recently reported results from a murine total-body irradiation (TBI) model demonstrating for the first time that a protein expression profile measured in samples collected 1 and 2 d after exposure can predict mice exposed to radiation and distinguish the level of photon-radiation exposure, ranging from 1 to 7 Gy. Combination of protein and hematological biomarkers demonstrated an enhanced separation of 1-Gy irradiated animals from controls and an improvement of the threshold for exposure detection compared to the selected protein profile only. We also reported results from several nonhuman primate (NHP) TBI studies. Dose- and time-dependent hematological and blood chemistry changes were demonstrated to be associated with early-phase and organ-specific plasma protein biomarkers in rhesus macaques irradiated to a broad dose range up to 13 Gy with $^{60}$Co $\gamma$-rays and 6MV LINAC x-rays. Herein, we present preliminary summary results for two animal irradiation models demonstrating proof-of-concept that proteomics shows promise as a complimentary approach to conventional biodosimetry for early assessment of radiation exposure as well as prognostic indicators of acute radiation sickness outcome. Research supported by Armed Forces Radiobiology Research Institute (AFRRI), Biomedical Advanced Research and Development Authority (BARDA), and Defense Threat Reduction Agency (DTRA).
One of the most serious disasters that could befall a city is the detonation of a nuclear device. Radiation exposure would be from mixed neutrons and gamma rays initially, and gamma rays from fallout. At doses where countermeasures would be useful, mortality from the Acute Radiation Syndrome (ARS) would be due to failure to produce blood elements, and gastrointestinal injury. Since much radiation injury is due to free radical production, a major theme of countermeasure development has been free radical scavengers. A second mechanism explored extensively is stimulation of blood cell progenitors using growth factors or inducers of growth factors. Other avenues are blocking cell death, and slowing cell division to enhance radioresistance. One important need is additional large animal models for advanced drug development. Four agents have US Food and Drug Administration (FDA) Investigational New Drug (IND) status, meaning they can be tested for safety in humans. These are 5-androstenediol (5-AED, Neumune), genistein (BIO300), Ex-Rad® and CBLB502. One, granulocyte colony-stimulating factor (G-CSF, Neupogen®), has Emergency Use IND status. AFRRI has been involved in the development of all five of these agents. G-CSF, 5-AED and genistein were conceived and initially developed as countermeasures at AFRRI. Ex-Rad® also was conceived at AFRRI, where most early development took place. CBLB502 was initiated at a company; AFRRI collaborated at an early stage. AFRRI is involved in evaluating hematopoietic progenitor cells as a radiation countermeasure. This therapy could be given days after an incident. Other promising AFRRI agents are tocols (Vitamin E), and dual use drugs that address both the ARS and late effects such as cancer. Policy questions revolve around planning for particular disaster scenarios, timing and routes of drug administration, priorities of various classes of agents, addressing specific ARS subsyndromes, and funding allocations.
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