FY2010 Highlights in Research Progress

Clinical Studies

Association of radiation and hepatitis virus infection on risk of hepatocellular carcinoma: A nested case-control study using stored sera

We conducted a nested case-control study using sera stored from A-bomb survivors who had participated in the Adult Health Study between 1970 and 2002. The study included 224 hepatocellular carcinoma (HCC) cases with prediagnostic stored sera available and 644 controls that were matched to the cases on gender, age, city, and time and method of serum storage, and countermatched on radiation dose.

The relative risks (RRs) (95% confidence interval [CI]) of HCC for radiation at 1 Gy, hepatitis B virus (HBV) infection alone, and hepatitis C virus (HCV) infection alone, with adjustment for alcohol consumption, body mass index (BMI), and smoking, were 1.67 (1.22–2.35), 63 (20–241), and 83 (36–231), respectively. Furthermore, when radiation and hepatitis virus infection were fit simultaneously, the RRs (95% CI) of HCC for radiation at 1 Gy, HBV infection alone, and HCV infection alone were 1.82 (1.09–3.34), 50 (16–184), and 87 (37–251), respectively. The RR (95% CI) of non-B, non-C HCC at 1 Gy was 2.74 (1.26–7.04) with adjustment for alcohol consumption, BMI, and smoking.

Radiation exposure was associated with increased risk of HCC, independently of HBV and HCV infections. In particular, radiation exposure was a significant risk factor for non-B, non-C HCC with no confounding by alcohol consumption, BMI, and smoking.

Long-term effects of blood pressure at midlife on stroke incidence

We assessed the lifetime risk of stroke and the impacts on that risk for various blood pressure-based groups based on the Adult Health Study. This study calculated the risk with consideration of competing risk of non-stroke death, which could cause bias for risk calculation in the long-term observation study.

The lifetime risks of stroke of 7,487 subjects were 20.5% in men, 22.2% in women. Hypertension (systolic/diastolic blood pressure ≥140/90 mmHg) was a significant factor in determining risk of stroke for men and women, with distributions of cumulative risk for stroke significantly different across blood pressure groups at baseline age of 55 years. The lifetime risks of all-stroke for normotension (<120/80) were 13.8% in men and 16.0% in women, whereas the lifetime risks after stage 2 hypertension (≥160/100) were 25.8% in men and 30.5% in women. The estimates did not differ significantly (p = 0.16) between normotensive and prehypertensive (120–139/80–89) subjects. Hypertension at age 55 predicted a similar degree of future increased risk for both ischemic and hemorrhagic stroke.

Our estimates clarified the long-term effects of blood pressure at midlife on stroke incidence.

Radiobiology/Molecular Epidemiology

An increase in the percentage of CD25+/CD127− regulatory CD4 T cells in the peripheral blood of A-bomb survivors

Our previous observations on long-term effects of A-bomb radiation on the T-cell system include impaired mitogen-dependent proliferation and IL-2 production, decreases in naïve T-cell populations, and increased proportions of anergic and functionally-weak memory CD4 T-cell subsets. We recently found a radiation-dose-dependent increase in the percentage of CD25+/CD127− regulatory T cells in the CD4 T-cell population of A-bomb survivors. Collectively, these effects of radiation resemble effects of aging on the immune system, suggesting that ionizing irradiation might direct the T-cell system toward a compromised phenotype and thereby might contribute to an enhanced immunosenescence.

Molecular and morphological characteristics of papillary thyroid cancer with *ALK* gene rearrangements

A new type of rearrangement, i.e., rearranged *ALK* gene has been found in papillary thyroid cancer (PTC) cases among A-bomb survivors who were exposed to relatively high doses. We recently identified a partner gene of rearranged *ALK* in six of these ten PTC cases, that is, *ALK* exon 20 was rearranged with exon 13 and 20 of *EML4*, in five cases and one case, respectively. Furthermore, trabecular/solid-like architectures were observed in six of these ten PTC cases with *ALK* rearrangements, while such morphology was found in only two of 15 PTC cases without *ALK* rearrangement. Similar structures have frequently been observed in Chernobyl PTC cases. These results suggest that *ALK* rearrangement plays an important role in radiation-associated adult-onset thyroid carcinogenesis.

Epidemiology

Risk on health in atomic-bomb survivors and those exposed *in utero*

High radiation doses lead to early natural menopause; above a threshold dose of 0.4 Gy an approximately linear dose-response curve was seen, with a risk at age 50 from 1 Gy of 34 excess cases per 1,000 women. The median age at which they experienced natural menopause was earlier by 0.3 year in those exposed to radiation at 1 Gy than in non-exposed. As the risk of breast cancer is associated with female reproductive histories in the general population, the interaction of those histories and radiation exposure was examined and no significant difference in risks was observed among the three female groups of pre-menarche, menarche to first birth, and after first birth at their experience of the atomic bomb. The increased risk of breast cancer was significant with an excess of around 100% to 150% at 1 Gy for all subjects (collaboration with the University of Washington, U.S.).

Survivors who were exposed to high-dose radiation *in utero* tended to have a low birth weight (around 200 g reduction at 1 Gy) and an increase in the risk of noncancer death in the early period, within five years since birth (around 30% increase at 1 Gy). The latter was rather strongly associated with low birth weight than radiation.

New mail survey

A new mail survey on the survivors (Life Span Study subjects) was completed for the 24,000 people who responded to the previous mail surveys, including recruitment to the Adult Health Study. Around 14,000 people responded (75% of those who received the mail).

FY2010 Highlights in Research Progress

Genetics

Electron spin resonance analysis of tooth enamel provides no evidence for exposures to large radiation doses in distally-exposed A-bomb survivors

We measured individual doses of 56 molars from 49 A-bomb survivors in Hiroshima who were present at distances of about 3 km or greater from the hypocenter of the bomb, using electron spin resonance (ESR) technique. The results provided estimated doses which vary from –200 mGy to 500 mGy, and the median dose was 17 mGy (25% and 75% quartiles are –54 mGy and 137 mGy, respectively) for the buccal parts and 13 mGy (25% and 75% quartiles: –49 mGy and 87 mGy, respectively) for the lingual parts of the molars. Three molars had ESR-estimated doses of 300 to 400 mGy for both, the buccal and lingual portions, which indicate possible exposures to excess doses of penetrating radiation, although the origin of such radiation remains to be determined. The results provided no evidence to support claims that a large fraction of distally-exposed survivors received large doses (e.g., 1 Gy) of external penetrating radiation resulting from indirect residual exposures.

A large-scale genetic study will be launched

For the past half-century, scientists in the Department of Genetics have searched for inherited differences between the children of A-bomb survivors and their parents that may be due to the A-bomb radiation exposure, i.e., whether germline mutation rates have increased in A-bomb survivors. However, due to the extremely low frequency of such germline mutations, it has been difficult to detect a sufficient number of mutations for a reliable estimation of genetic risk from radiation. Using the microarray technique, which has undergone remarkable development recently, it appears possible to test the whole genome in detail and determine the presence or absence of genetic effects. We have improved our microarray experimental methods and have started evaluation of genetic effects of radiation using animal models (a mouse model for paternal exposure and a rat model for maternal exposure). The improved reproducibility and reduced costs have made possible the initiation of a large-scale study in FY2011 of Hiroshima and Nagasaki A-bomb survivors and their children. The estimation of the risk of genetic effects from radiation exposure might be possible at some time in the near future.

Statistics

Radiation risk assessment and dosimetry

In the area of risk assessment, Statistics researchers began to investigate the use of multiple imputation for handling missing data on smoking in collaboration with researchers at the U.S. National Cancer Institute and presented related material to the Scientific Council, completed a manuscript on mechanistic vs. descriptive modeling of solid cancer and multi-model inference in collaboration with investigators from the Helmholtz Centre Munich, Germany, and continued other collaborative work on mechanistic model-based risk assessment. A new statistical research protocol (RP) was approved on specialized methods of intermediate risk factor analysis for stratified (counter-matched) nested case-control sampling designs used in RERF studies. Members of the Statistics Department also contributed as statisticians to a number of papers on risk assessment using established statistical methods.

In the area of dosimetry, Statistics organized and hosted an international workshop (in March 2011) on ways to improve and extend DS02 organ dosimetry by using DS02's existing core calculations. Members of Statistics continued to support the Dosimetry Committee's efforts, particularly in improvement of survivor location estimates, which included the acquisition of aerial photographic maps of the cities and the initiation of work on using them to precisely locate survivor's shielding history neighborhood diagrams. Members continued collaborative work with external investigators on dose error, biodosimetry, and neutron dose weighting factors.

Statistical methodology for other RERF studies

Members of Statistics prepared a manuscript on power and sample size determination in genetic-disease association studies, performed research on choice of time scales in Cox regression of epidemiologic follow-up data and presented methods and recommendations in an RERF colloquium, produced an S-plus (statistical software) program to perform a specialized algorithm for stratified case-cohort data, and organized and led two in-house workshops on bioinformatics. They also continued collaborative research with external investigators on a method of extrapolating survival estimates. Members of Statistics assisted other scientists in the use of specialized statistical methods in analyses for numerous manuscripts and provided statistical consultation on designs and power calculations for several new RPs.

Information Technology

The dynamic data backup system that was developed last year using the internet has started full-scale operations. Previously, we performed backup by saving data onto multiple magnetic tapes, sending one copy to the Nagasaki Laboratory on a periodic basis and keeping the information at both the Hiroshima and Nagasaki Laboratories, eventually securely storing such magnetic tapes in an underground safe deposit box at Hiroshima Bank. We decided to adopt a new backup method of transferring such data directly to a backup server installed at the Nagasaki Laboratory over the internet, and started construction on the system last fiscal year. Through this backup system, important data at RERF are accumulated in a backup server at the Hiroshima Laboratory every night, with all such data transferred to the Nagasaki Laboratory once a week. At the same time, data are separately stored on magnetic tapes, which are stored in an underground safe at Hiroshima Bank, as was the case previously. Because duplicate processing of files for remote backup takes time, it is still difficult to transfer backup files on a nightly basis although telecommunication lines have been enhanced.

Virtualization of servers, which will be useful in next-generation environments, has made further progress. This will help reduce the physical number of servers, but its main purpose is to construct a server environment that will not be affected by changes or types of operating systems. At present, 14 servers are running in the virtual environment. Various OSs, such as Windows NT, Windows Server 2003, and Linux, are operated in the virtual server environment as if they were independent servers. Therefore, even if one server goes down, the backup system is secure due to its clustering structure.

Cardiovascular Disease Study

Whether A-bomb radiation exposure causes cardiovascular diseases has attracted much attention from those concerned, and many of RERF's research results were detailed in Attachment B of a 2006 report by the United Nations Scientific Committee on the Effects of Atomic Radiation (UNSCEAR, 2008).

To address this issue comprehensively based on research in epidemiology as well as clinical and basic medicine, RERF established the Cardiovascular Disease Study Working Group, consisting of the Chief of Research, chief scientists, department chiefs, and research scientists. This group has been reviewing the issue as a project team since 2008. Starting in FY2008, the team summarized the results of studies conducted at RERF and spent considerable time in a review process aimed at streamlining hypotheses to be verified. In FY2009, the team initiated review of new studies to be conducted in the future, examined details of a study on arteriosclerosis to be started by the Department of Clinical Studies, and engaged in debate on the possibility of animal experiments. In addition, research plans on stroke, chronic kidney diseases, arteriosclerosis markers, valvular diseases, and related biomarkers and immunological functions were reviewed and given high marks by the Scientific Council that met at the end of the fiscal year. In FY2010, an animal experiment using spontaneous hypertensive rats and a clinical study to investigate involvement of cytokines in more detail were initiated.

F_1 Clinical Study

A clinical study of the F_1 offspring of A-bomb survivors was conducted from 2002 through 2006 to examine from an epidemiological perspective the presence or absence of effects of parental radiation exposure on adulthood development of multifactorial diseases among F_1 subjects. The results of this study were published in a 2008 report. In the study, however, the average age of the F_1 subjects who underwent health examinations was young, at 49 years, just at the beginning of the age range in which diseases frequently occur. At the same time, the possible presence of cross-sectional study bias could not be ruled out. Thus, the Scientific and Ethics Committees for the Health Effects Study of the children of A-bomb Survivors, the Scientific Council, and the Senior Review Panel recommended that a longitudinal study be conducted. Based on these recommendations, RERF's F_1 Clinical Study Working Group, consisting of the Chief of Research, a permanent director, chief scientists, department chiefs, and research scientists, had discussions to develop a new research plan, with Research Scientist Waka Ohishi (Department of Clinical Studies, Hiroshima) as the responsible investigator, which was completed in FY2009. This research plan was reviewed and approved by the Scientific and Ethics Committee for the Clinical Study of the F_1 Offspring of A-bomb Survivors (Chairman: Dr. Tadao Shimao, Vice Chairman: Dr. Hiraku Takebe) in July 2010, on the condition that the explanatory note in the consent form be partially revised. Upon successful completion of the preliminary work for implementation of the study in collaboration with the Department of Epidemiology, Department of Information Technology, and other departments, the longitudinal study commenced in November 2010.

Dosimetry

Maps created by the U.S. Army immediately after the war were used to identify hypocenter coordinates and survivor locations, but the issue of distortion in these maps has been raised for many years. Additionally, doses for about 7,000 people in the Life Span Study population were treated as "unknown" because DS02 could not be applied to their shielding conditions. To solve such problems in dose estimation, the Dosimetry Committee was established.

This fiscal year, aerial photographs taken immediately before the A-bombings were modified based on their exposure angle, height, and lens aberration, as well as altitude of the photographed images to prepare one flat orthogonal aerial photograph consolidating each of the entire areas in question, with which locations in the standard coordinate system were identified. Based on reference points held in common between these maps and the U.S. Army maps, we formulated equations to convert coordinates on the U.S. Army maps to those on the aerial photographs covering both of the cities in their entirety. Further, with respect to proximally exposed survivors for whom neighborhood drawings showing their shielding histories were available, we established methods to reconfirm their exposure locations on the orthogonal aerial photographs, using GPS software, and also have started the work of individual identification.