# ID: 0831 Radiation/Nuclear Medical Countermeasures Development Program

## Bert Maidment<sup>\*1</sup>, Jessica Benjamin, David Cassatt, Andrea DiCarlo, Joseph Kaminski, Narayani Ramakrishnan, and Richard Hatchett

<sup>1</sup> Division of Allergy, Immunology, and Transplantation; National Institute of Allergy and Infectious Diseases; National Institutes of Health, Bethesda, Maryland 20892, USA

**Abstract.** The threat of nuclear or radiological attack has grown in recent years, with increased activity of global terrorist organizations and a rise in illicit trafficking of radioactive materials. Very few medical products exist to mitigate and/or treat acute and long-term injuries that can result from a nuclear or radiological accident or attack. The National Institute of Allergy and Infectious Diseases, National Institutes of Health was given the responsibility by the Department of Health and Human Services to identify, characterize and develop new medical countermeasures against radiological and nuclear attacks. NIAID is supporting a number of efforts to ensure the availability of safe and effective medical countermeasures for the Strategic National Stockpile. NIAID provides funding for basic/translational research programs and product development support services through grant and contract mechanisms. The research and development programs include development of new medical products to mitigate and/or treat acute and long-term radiation injury, facilitate the elimination of internal radionuclide contamination, and determine levels of individual radiation exposure. The priority areas of research and product development interests are:

- Medical products and regimens that mitigate and/or treat radiation injury post-exposure (i.e., administered at least 24 hours after radiation exposure), with emphasis on broad activity, ease of administration in an emergency scenario, safety, and long shelf-life;
- Radionuclide decorporation agents that facilitate elimination of various radionuclides from the body;
- New medical product formulations that can be easily administered to civilian populations including special populations (e.g. children, elderly, immunocompromised); and
- Minimally invasive biodosimetry methods or devices useful for emergency triage that can rapidly and accurately distinguish individuals who need treatment from those who do not, and that can identify and measure internal and/or external exposure.

#### KEYWORDS: Radiation injury mechanisms, radionuclides, medical countermeasures, biodosimetry

#### **1.0 Introduction**

The threat of nuclear or radiological attack has grown in recent years, with increased activity of global terrorist organizations and a rise in illicit trafficking of radioactive materials. Very few medical products exist to mitigate and/or treat acute and long-term injuries that can result from a nuclear or radiological accident or attack. The National Institute of Allergy and Infectious Diseases (NIAID), National Institutes of Health (NIH) was given the responsibility by the Department of Health and Human Services (HHS) to identify, characterize and develop new medical countermeasures against radiological and nuclear attacks. NIAID's Radiation/Nuclear Medical Countermeasure Development Program is supporting a number of efforts to ensure the availability of safe and effective medical countermeasures for the Strategic National Stockpile. NIAID provides funding for basic/translational research programs and product development support services through grant and contract mechanisms. This paper provides an overview of NIAID's comprehensive radiation medical countermeasure research and development program.

<sup>\*</sup> Presenting author, E-mail: <u>maidmentb@niaid.nih.gov</u>

### 2.0 Radiation/Nuclear Medical Countermeasure Basic and Translation Research Grant Programs in Chronological Order

#### 2.1 Centers for Medical Countermeasures against Radiation (CMCR)

In FY 2005, the NIAID established eight Centers for Medical Countermeasures against Radiation to support a range of basic and applied radiobiology research spanning biodosimetry, immune reconstitution, therapeutics and training. The eight Centers for Medical Countermeasures against Radiation and the respective Principal Investigators (PIs) are;

- Columbia University Medical Center, New York, NY PI: David Brenner
  Center for High Throughput, Minimally Invasive, Radiation Biodosimetry
- Dana-Farber Cancer Institute, Boston, MA PI: Alan D'Andrea Dana-Farber/Harvard Center for Medical Countermeasures Against Radiation
- Duke University Medical Center, Durham, NC PI: Nelson Chao Radiation Countermeasures Centers of Research Excellence (RadCCORE)
- Fred Hutchinson Cancer Research Center, Seattle, WA PI: George Georges Radiation Dose-Dependent Interventions
- Medical College of Wisconsin, Milwaukee, WI PI: John Moulder
  Post-Irradiation Intervention to Mitigate and Treat Non-Hematological Injuries
- University of California David Geffen School of Medicine, Los Angeles, CA PI: William McBride UCLA Center for Biological Radioprotectors
- University of Pittsburgh, Pittsburgh, PA PI: Joel Greenberger Mitochondrial Targeting Against Radiation Damage
- University of Rochester Medical Center, Rochester, NY PI: Paul Okunieff
  Center for Biophysical Assessment and Risk Management Following Irradiation

The Centers for Medical Countermeasures against Radiation (CMCRs) carry out critical research and development of radio-protectants, -mitigators and -therapeutics to treat the short and long-term medical consequences of radiation exposure. Ongoing research in the CMCRs also includes discovery of radiation biodosimetric markers and the development of devices and protocols to assess radiation dose received by an individual for prompt triage and treatment decisions after any radiological event. Also, this program supports innovative and novel research through pilot projects, and a number of research studies are underway to better understand the mechanism of radiation damage to the cell, and identify key points in radiation cellular and organ response pathways where damage can be prevented.

#### 2.2 Protecting the Immune System against Radiation

NIAID funded four grants in FY05 to support research projects focused on practical methods for preexposure protection of the immune system against damage by radiological or nuclear terrorist attacks, and/or practical methods to reconstitute hematopoietic stem cells, their progeny, or mature cells of the immune system following exposure to immunosuppressive radiation. Researchers are identifying cellular therapies (including use of mesenchymal stem cells with allogeneic transplants), and compounds (e.g. combination cytokine therapy, growth factor treatments, and novel TLR signaling molecules) for treating acute radiation syndrome. Animal models in which this research is being performed include non-human primates, dogs and rodents.

#### 2.3 Radionuclide Decorporation Agents for Radiation/Nuclear Emergencies

NIAID awarded five grants in FY06 to support the development of novel radionuclide decorporation agents that are more effective, safe, and bind a broader range of radionuclides. Also, these grants will develop oral decorporation agent formulations that will have advantages of ease of distribution and administration in response to a large terrorist event. Five different approaches are being explored:

### 2.3.1 Siderophore analogs for actinide decorporation (desferrithiocin and hydroxypyridinonone analogs)

Dr. Raymond Bergeron at the University of Florida and Dr. Kenneth Raymond at the Lawrence Berkeley National Laboratories are exploring other agents for decorporating Uranium (U) and transuranic metals, due to the limitations of diethylenetriaminepentaactate (DTPA). Dr. Bergeron is evaluating orally effective forms of desferrithiocin and its derivatives. Since desferrithiocin has unacceptable renal toxicity, he is searching for less-toxic analogues for U decorporation. His search has resulted in the identification of several orally available analogues with improved decorporation and toxicity profiles. In rat models, the reduced toxicity analogues led to a significant increase in fecal excretion of U and resulted in a net reduction of U in the body, especially the kidneys. In contrast, DTPA was ineffective at decorporating U. Dr. Raymond is developing analogs of hydroxypyridinonate for Pu, Am, Thorium (Th) and U. He has identified two lead hydroxypyridinonate compounds, an octadentate and a tetradentate HOPO analog. Orally administered HOPO analogs are more potent actinide decorporating agents than DTPA. Results have shown that HOPO analog are up to 30 times more effective than DTPA in Pu decorporation and can be orally administered. Recent evaluation in rodents has demonstrated a low toxicity profile.

#### 2.3.2 Amphipathic oral chelators

Dr. Scott Miller at the University of Utah is developing amphipathic polyaminocarboxylic acid chelators based upon triethylenetetramine-hexaacetic acid (TT). TT analogs are structurally similar to DTPA, however, they are orally bioavailable and can be modified for preferential uptake into specific tissues and for excretion through the biliary (more lipophilic) or urinary (more hydrophobic) system. These compounds have one more binding moiety compared to DTPA and can chelate a broader range of radionuclides. When administered to rodents in their food, the  $C_{22}TT$  analog was shown to decorporate Am, U, and Cobalt (Co).

#### 2.3.3 Biomaterials: chitosan

Dr. Tatiana Levitskaia at the Pacific Northwest National Laboratory is developing and evaluating chitin derivatives as potential decorporating agents for Strontium (Sr), Radium (Ra), Co, Am, Pu, and U. Chitin is an abundant natural biopolymer which is non-toxic and can be administered orally. Chitosan can readily be chemically modified to make it more suitable as a decorporating agent for other metals such as Cesium (Cs) and Iridium (Ir).

#### 2.3.4 Nano-engineered sorbents

Dr. Charles Timchalk at the Pacific Northwest Laboratory is developing a new class of nanostructured sorbents - self-assembled monolayer on mesoporous supports (SAMMS) materials. SAMMS are hybrid materials of mesoporous silica (SiO<sub>2</sub>) that are covalently linked to selective chelators based on desired radionuclide chelating properties. SAMMS were developed to facilitate radioactive complex waste cleanup at Department of Energy sites. SAMMS have been shown to effectively chelate U, Pu, Am, I, Co, Cs, Th, and other metals. Dr. Timchalk is developing SAMMS for gastrointestinal (GI) chelation of radionuclides. SAMMS insolubilize the radionuclide in the gut and prevent systemic absorption or re-absorption. He is also developing and evaluating extracorporeal devices incorporating SAMMS for rapid removal of radionuclides from blood.

Successful development of novel decorporation agents will expand the range of radionuclides that can be safely and effectively removed from the body, provide more effective agents to remove radionuclides deposited in various tissues, and provide oral forms that have operational advantages in the event of a terrorist incident.

#### 2.4 Medical Countermeasures to Restore Gastrointestinal Function after Radiation Exposure: Project Bioshield

In August, 2007, ten grants were awarded for the development of medical countermeasures to restore GI function after radiation exposure. The goal of these awards is to accelerate the development of safe and effective medical products to prevent, mitigate and treat the GI injury and to restore GI function after radiation exposure from radiological and nuclear terrorist attacks. Under this program several potential countermeasures to mitigate and treat radiation-induced GI injury are being tested in different animal models.

### **2.5** Medical Countermeasures to Enhance Platelet Regeneration and Increase Survival Following Radiation Exposure

Radiation exposures from radiological and nuclear terrorist attacks can lead to hematopoietic acute radiation syndrome (ARS), which can result in the depletion of hematopoietic stem cells and progenitors, leading to severe neutropenia and thrombocytopenia. Platelets play an essential role in hemostasis and thrombosis. As the level of circulating platelets drops, the risk of catastrophic hemorrhage increases markedly. Severe thrombocytopenia is a contributor to mortality following radiation exposure. Currently there are no approved post-exposure drugs for radiation-induced thrombocytopenia. The only therapies currently available to patients are platelet concentrates or fresh whole blood transfusions. In small accidents, supportive transfusion is the standard of care. The logistical requirements for providing these transfusions to large numbers of victims after a mass casualty event are challenging. This year, NIAID will support up at least four grants to accelerate the development of safe and effective medical products to mitigate and treat thrombocytopenia and to enhance platelet regeneration thereby increasing survival after radiation exposure. An effective pharmacologic therapy that mitigates or treats radiation-induced thrombocytopenia would also offer operational advantages over platelet or blood transfusions in a mass casualty scenario.

#### **2.6** BARDA/NIAID Medical Countermeasures to Mitigate and/or Treat Ionizing Radiation-Induced Pulmonary Injury: Project Bioshield

This year, NIAID and HHS/Biomedical Advanced Research and Development Authority (BARDA) will collaboratively fund up at least four grants to development medical countermeasures for radiationinduced pulmonary injury. The objectives of this grants program are to develop products for the mitigation and/or treatment of radiation induced pulmonary damage. Successful medical countermeasure candidates are expected to eventually obtain Food and Drug Administration (FDA) licensure through additional support from industry or other government-funded programs. This product development program was established since exposure to ionizing radiation in a terrorist event has the potential to cause life-threatening pulmonary injuries (pneumonitis and/or fibrosis), whether through external or internal exposure. Radiation-induced injuries of the hematopoietic, cutaneous, and gastrointestinal systems are thought to be the main causes of mortality following acute radiation exposure. As new medical countermeasures are developed and supportive care protocols improved, exposed individuals may succumb to radiation-induced pneumonitis and/or pulmonary fibrosis. There is a need to develop countermeasures against the lethality caused by radiation-induced pulmonary damage. The availability of new products that can mitigate and/or treat pulmonary injury from radiation exposure will increase the medical management options and potentially reduce the numbers of casualties in a radiation exposure event.

#### 2.7 BARDA/NIAID Medical Countermeasures to Mitigate and/or Treat Ionizing Radiation-Induced Cutaneous Injury: Project Bioshield

NIAID and HHS/Biomedical Advanced Research and Development Authority (BARDA) will collaboratively fund at least four grants to development medical countermeasures for radiation-induced cutaneous injury. The goals of this program are to develop medical countermeasures for mitigation and/or treatment of radiation (or combined) induced cutaneous injury. Successful medical countermeasure candidates that prove efficacious are anticipated to advance towards FDA licensure. This medical countermeasure development program was established because the level of cutaneous radiation injury is one of the major determinants for early mortality and long-term morbidity.

### **2.8** Radiation Combined Injury: Radiation Exposure in Combination with Burn, Wound, Trauma or Infection

In many terrorist radiation exposure scenarios, injury will likely involve radiation exposure and additional, complicating trauma. A majority of the people injured in a nuclear event involving either a dirty bomb or nuclear detonation would also be expected to receive other injuries in addition to those caused by radiation exposure. Such combined injuries, including wounds, blunt trauma, and/or concomitant infection could lead to decreased overall survival. Studies of radiation combined injury in animal models show increased susceptibility to infection, delayed wound healing and decreased survival. There are currently medical countermeasures available for the management of the different aspects of radiation combined injury (e.g. burn care, wound closure and treatment, trauma minimization, and infection control); however, there has been only limited testing on the efficacy of these compounds when radiation exposure is a factor. Therefore, there is an urgent need to understand the mechanisms behind the synergistic lethality observed with radiation combined injury, to define appropriate animal models to study this complex form of radiation injury, to determine efficacy of treatments against damage resulting from radiation combined injury, and to identify appropriate targets for the development of novel countermeasures. This year NIAID will fund ten research grants using the R21/R33 NIH mechanism, which allows for the gathering of preliminary data during the 2-year, milestone-based R21 phase, and full exploration of the implications of the science during a 3-year R33 phase.

### **2.9** Mechanisms, Diagnosis and Treatment of Radiation Injury from a Nuclear Accident or Terrorist Attack

This year NIAID will fund at least ten R01 grants to expand the options available to identify, mitigate and/or treat radiation-induced injury, and thereby help minimize the terrorist threat. NIAID has established a basic and translational research program by soliciting proposals using a Request for Applications (RFA) mechanism. Currently, investigator-initiated applications span a broad range of topics, ranging from investigations into the mechanisms of radiation-induced cellular and organ injury to development of new methods of rapid dose assessment or new mitigators/therapeutics to address the short- and long-term effects of radiation exposure. Some of the studies to be funded support research to advance diagnostic devices and/or medical products already in early developmental stages, as well as supporting efforts to identify and develop new candidate products through basic and translational research.

#### **3.0 Contract Programs**

### **4.1** Medical Countermeasures against Radiological Threats: Product Development Support Services

Key to NIAID's ability to develop safe and effective radiation and nuclear medical countermeasures was the establishment of the broad range of product development expertise, capabilities, facilities, and services to support FDA licensure. To achieve these goals, NIAID awarded a product development support services contract in FY05 to the University of Maryland School of Medicine (UMSOM). Under the leadership of the PI, Dr. Thomas MacVittie, UMSOM and a number of subcontractors established the Medical Countermeasures against Radiological Threats (MCART) group. MCART includes all the elements of drug development, including GLP research and animal safety testing facilities, manufacturing facilities, regulatory support and clinical study support and allows for NIAID to perform efficacy studies and to fill in other data gaps for medical countermeasures. A GLP compliant data collection and management system has been established to facilitate communication, evaluation and storage of data to be generated by MCART. Screening and evaluation facilities and protocols were developed to study the efficacy candidate medical countermeasures to mitigate or treat acute radiation syndromes (ARS) in animal models of hematological ARS and gastrointestinal (GI) ARS. Included in the GLP network are the University of Maryland and the University of Illinois in Chicago which provide support for non-human primate studies. Indiana University which provides support for screening and rodent animal model development and testing, and EpiStem in Manchester, UK which provides support for GI studies.

In addition, specialized facilities to screen and evaluate candidate countermeasures ability to rapidly eliminate radionuclide contamination from the body are being renovated at the Lovelace Biomedical and Environmental Research Institute and should be operational shortly. With the establishment of MCART, the University of Maryland with all their subcontractors, NIAID has developed a virtual pharmaceutical company capable of developing medical countermeasures for radiation exposure from discovery to FDA licensure.

#### 4.2 Development of Improved DTPA for Radionuclide Chelation

Ca DTPA and Zn DTPA are approved for decorporation of transuranic (Pu, Am, and Curium (Cm)) radionuclides deposited internally in the body. However, DTPA is poorly absorbed when administered orally and must be administered intravenously or by inhalation using a nebuliser. These routes of administration are difficult to use in a mass casualty situation. There is a need to develop alternative formulations or forms of DTPA that are suitable for a mass casualty event. NIAID funded a program for the development of improved forms of DTPA in FY05. The awardees were the University of Kentucky, SRI International, and Nanotherapeutics, Inc. The goal of the program is to develop a formulation of DTPA that can be given orally with improved bioavailability.

At the University of Kentucky, the PI, Dr. Michael Jay, and his colleagues have been using a pro-drug approach to enhance the oral absorption of DTPA. Their approach is to link lipophilic moieties to the carboxylic acid groups of DTPA through an ester linkage, with the resulting pro-drug being metabolized by esterases to yield the active DTPA. This approach has resulted in a pro-drug form of DTPA with demonstrated high oral bioavailability and effective decorporation of Am.

At SRI International, the PI, Dr. Gita Shankar, and her group are formulating DTPA with additives (GI absorption enhancers) that enhance intestinal transport of associated compounds. Tests on a variety of additives and combinations were first performed *in vitro*, with the most promising candidates tested *in vivo*. This approach has also yielded a formulation that demonstrated increased bioavailability of DTPA and showed decorporation of Am.

At Nanotherapeutics, Inc., the PI, Dr. James Talton, and his colleagues have incorporated DTPA into formulated Nanoparticles. The nanoparticle formulation has shown increased oral bioavailability *in vivo*.

#### **5.0 Other Programs**

#### 5.1 Interagency Agreement with NIH, National Cancer Institute

In FY05, NIAID entered into an Interagency Agreement (IAA) with the National Cancer Institute (NCI). The goals of this IAA were to allow for cooperation between the NCI and NIAID on specific research and development projects within the following areas: 1) development of safe and effective medical countermeasures to prevent, mitigate, and treat the immediate and long-term medical effects of ionizing radiation; 2) improvement of the basic understanding of radiation-related health risks associated with types and levels of radiation exposure (epidemiology & dosimetry), mechanisms of radiation injury, and host responses; and, 3) development of biology-based diagnostic assays or biomarkers to assess cellular and tissue damage following exposure to ionizing radiation. Studies are being funded at NCI include: 1) DNA double-strand break formation in human skin models after radiation exposure (Dr. William Bonner), 2) radiation-induced metabolomics changes (Dr. Frank Gonzalez), 3) research program on radiological and nuclear threats (Drs. Martha Linet and Steve Simon), 4) development of nitroxide radiation protectors (Dr. James Mitchell), and 5) novel, protein-based biodosimetry methods (Dr. Kevin Camphausen).

#### 5.2 Interagency Agreement with Armed Force Radiobiology Research Institute

In FY 2004, the NIAID established an IAA with the Armed Forces Radiobiology Research Institute (AFRRI) to develop and evaluate the safety and efficacy of medical countermeasures to prevent and treat the effects of ionizing radiation; and also to automate a cytogenetic approach to biodosimetry. Under this program, AFRRI is evaluating the toxicity, survival efficacy and dose reduction factor of several compounds in rodents following exposure to ionizing radiation. AFRRI has developed automated laboratory methods to process approximately 500 cytogenetic samples weekly. Results of the cytogenetic assay could be used for triage and dose assessment. In addition, AFRRI along with 4 other cytogenetic laboratories in US, Canada, Europe and Asia conducted the first inter-laboratory comparison of the dicentric cytogenetic assay since 1988. In addition, procedures have been developed and optimization of the methodologies has been initiated.

#### 5.3 Memorandum of Understanding with NSBRI

In FY07, the NIAID established a Memorandum of Understanding (MOU) with the National Space Biomedical Research Institute (NSBRI) to promote collaboration and transparency in research and development between agencies to benefit the nation's ability to respond to a radiological or nuclear incident, to advance the space exploration program, and to promote the educational missions of both institutes. Both parties have agreed to exchange scientific information by communicating openly and, where feasible, seeking to coordinate research initiatives in areas of mutual interest, such as 1) prodromal syndrome mechanisms and chronic metabolic/oxidative stress resulting from acute exposure to ionizing radiation, 2) effects of radiation exposure on the skin, 3) development of animal models, which closely resemble human responses, for testing countermeasures, 4) development of biodosimetric techniques and devices, 5) study of late-effects resulting from ionizing radiation that are triggered by acute exposure effects; 6) radiation effects on the central nervous system and development of behavioral test models, and 7) joint and/or coordinated education and outreach.

#### **6.0** Conclusions

NIAID's radiation/nuclear medical countermeasure research and development programs include development of new medical products to mitigate and/or treat acute and long-term radiation injury, facilitate the elimination of internal radionuclide contamination, and determine levels of individual radiation exposure. The priority areas of research and product development interests are:

- Medical products and regimens that mitigate and/or treat a broad range of radiation injuries post-exposure (i.e., administered at least 24 hours after radiation exposure), with emphasis on ease of administration in an emergency scenario, safety, and long shelf-life;
- Radionuclide decorporation agents that facilitate elimination of various radionuclides from the body;
- New medical product formulations that can be easily administered to civilian populations including special populations (e.g. children, elderly, immunocompromised); and
- Minimally invasive biodosimetry methods or devices useful for emergency triage that can rapidly and accurately distinguish individuals who need treatment from those who do not, and that can identify and measure internal or external exposure.

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