RC-12. Biological Dosimetry

Early Biodosimetry Response: Recommendations for Mass-Casualty Radiation Accidents and Terrorism

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EARLY BIODOSIMETRY RESPONSE: RECOMMENDATIONS FOR MASS-CASUALTY RADIATION ACCIDENTS AND TERRORISM

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Abstract

The accepted generic multiparameter and early-response approach includes measuring radioactivity and monitoring the exposed individual; observing and recording prodromal signs/symptoms and erythema; obtaining complete blood counts with white-blood-cell differential; sampling blood for the chromosome-aberration cytogenetic bioassay using the "gold standard" dicentric assay (translocation assay for long times after exposure) for dose assessment; bioassay sampling, if appropriate, to determine radioactive contamination; and using other available dosimetry approaches. In the event of a radiological mass-casualty incident, local, national and international resources need to be integrated to provide suitable dose assessment and continuing clinical triage and diagnoses. This capability should be broadly based and include i) training and equipping local responders with tools and knowledge to provide early radiological triage, ii) establishing radiological teams capable to rapidly deploy and provide specialized dose assessment capabilities (i.e., radiation screening and radiobioassay sampling, hematology, etc.), and iii) access to reach-back expert reference laboratories (i.e., cytogenetic biodosimetry-, radiation bioassay-, electron paramagnetic resonance-based dose assessment). This multifaceted capability needs to be integrated into a biodosimetry "concept of operations" for use in a mass-casualty radiological emergency. On-going research efforts to identify and validate candidate screening and triage assays should ultimately contribute towards approved, regulated biodosimetry devices or diagnostic tests integrated into local, national, and international radioprotection programs.

1. Introduction

The effective medical management of a suspected acute-radiation overexposure incident necessitates recording dynamic medical data, measuring appropriate radiation bioassays, and estimating dose from dosimeters and radioactivity assessments in order to provide diagnostic information to the treating physician and a dose assessment for personnel radiation protection records. The accepted generic multiparameter and early-response approach includes measuring radioactivity and monitoring the exposed individual; observing and recording prodromal signs/symptoms and erythema; obtaining complete blood counts with white blood cell differential; sampling blood for the chromosome-aberration cytogenetic bioassay using the "gold standard" dicentric assay for dose assessment; bioassay sampling, if appropriate, to determine radioactive contamination; and using other available dosimetry approaches (e.g., dose assessment by measurement of free radicals in solid matrix materials using electron paramagnetic resonance, EPR) [1-2]. The practice of radiation medicine dictates the establishment of response capability for rapid medical diagnosis and management of individuals overexposed. For example, many nations have established reference expert cytogenetic biodosimetry laboratories.

There are hundreds of instances in which one or more persons were accidently overexposed to ionizing radiation [3]. A subset of these incidents involves mass-casualty scenarios [2, 4]. A radiological or nuclear attack is also a possibility [5]. Because of recent terrorist activities and intelligence information, there is strong sentiment that it is not a question of if, but when, a radiological or nuclear terrorist attack will occur [6].

The clinical medical decision needs associated with potential mass-casualty events prompted Lloyd and colleagues to advocate the diagnostic role of cytogenetics in early triage of radiation casualties [7]. Reference expert cytogenetic laboratories have recently established regional (e.g., reference cytogenetic laboratories among the nations of the United Kingdom, Germany, and France) and national [8-10] networks to enhance their capabilities. In cases of urgent need for assessment in radiological exposures, individual nations often rely on international cooperation facilitated by United Nation (UN) agencies (i.e., World Health Organization or WHO, International Atomic Energy Agency or IAEA). In the event of a radiological mass-casualty incident, current national and international resources need to be enhanced to provide suitable dose assessment and medical triage and diagnoses.

A coordinated approach involving preplanning, stockpiling of reagents and equipment, establishment and exercise of specialized response teams, and a consensus “concept of operations” for biodosimetry applications in a mass-casualty radiological incident is required. Proper equipment for identifying radiation and radioactive contamination needs to be available to trained first responders. Specialist in radiological protection must also be available to provide expert advice and assistance to implement critical operational biodosimetry functions [11]. Preplanning and stockpiling of suitable reagents and equipment are essential [12]. Consensus concept-of-operations for biodosimetry application during a mass-casualty radiological emergency, tailored for specific radiological scenarios (i.e., radiation dispersal device, radiation emitting device, and improvised nuclear device), are also needed. The United States Office of Science and Technology Policy and the Homeland Security Council established an interagency working group that prioritized research areas for radiological nuclear threat countermeasures including efforts to automate biodosimetric assays and develop biomarkers for biodosimetry [13]. These biodosimetric research efforts are focused to identify, optimize, and validate novel biodosimetric assays to support triage, clinical, and definitive radiation dose and injury.

2. Biodosimetry preplanning

2.1 Radiation exposure assessment methods

Table I illustrates a list of radiation exposure assessment methods applicable for early-phase acute radiation based on international consensus of experts [2]. Protocols for use of these established and

Table I. Acute-phase patient assessment methods.*

Assessment Method	Parameters for considering assessment method for use in early (<5 d) triage screening			Applicable for scoring ARS severity	Dose (Gy) or ARS response category level to select for priority cytogenetic triage analysis	
	Time for analysis	Estimate cost per sample, US Dollars			Triage dose, Gy	Response category levels
Direct Recording of Location History	< 2 min	-			3-7	
Direct Observation of Clinical Signs and Symptoms	< 5 min	-		Yes	3-7	1-4
Personal Monitoring (Direct, non invasive)						
- *in vivo* EPR	Unknown	Unknown			3-7	
- portable hand held meters (triage/screening)	< 5 min	-			-	
- portal monitors (triage/screening)	< 2 min	-			-	
- whole-body counting	> 25 min	-			-	
Personal Monitoring (Indirect, invasive)		**Detection limit,#**	**Estimate cost per sample, US Dollars#**			
- blood chemistry (*i.e.*, amylase activity)	< 3 min		<$2		3-7	
- CBC and differential/lymphocyte count	< 2 min		<$1	Yes	3-7	1-4
- *in vitro* EPR (i.e., nails)	<15 min		Unknown		3-7	
- nasal swab	> 1 d	50 pCi/swab	$70		-	
- stool sample	> 1 d	5 pCi/g	$80		-	
- urine sample (spot; 24-hr)	< 1 d; > 1 d	30 pCi/vial	$90		-	
- cytogenetics (i.e., 20-50 metaphase triage; 1000 metaphase analysis)	>3 days	1 Gy; 0.2 Gy	Unknown; $500-3,000		-	
Area Monitoring						
- dosimetry results (*e.g.* TLDs, aerial measurements) combined with personal location information	Unknown	-			3-7	

*The Table was modified a version reported by Alexander and colleagues [2].
Note that the personal and area monitoring methods are listed in alphabetical order and, therefore, their location in the table does not infer priority or preference.
Radiobioassay detection limits and costs are based on ^{137}Cs isotope and 1 min gamma-ray spectrometry analysis with high priority count (costs 3-times routine) with no automatic sample changers used. Detection limits for cytogenetic analysis are presented in acute photon equivalent dose in units of Gy.

provisional methods were also described in the Appendix section of this report. Table I also shows several features associated with these assessment methods for considering their use for early triage screening, applicability for scoring acute radiation syndrome (ARS) severity, and criteria for their use to prioritize suspected exposed individuals dose assessment by cytogenetic biodosimetry. Depending on the radiation scenario and available resources, appropriate radiation assessment methods should be implemented in a mass-casualty radiological terrorism or radiation accident incident.

2.2 Radiation/radiological response teams and networks

Specialist in radiation protection supporting early-response to radiation emergencies are typically organized into teams with discrete functions as illustrated in Table II. For example, Remick and colleagues [14] described U.S. national resources of response teams for radiological incidents. In certain nations, components of these radiological resources are organized into teams that address assessment and medical response for nuclear, biological, and chemical threats [15].

Table II. Selected List of Radiological Response Teams

Initial Assessment	Nuclear, Chemical, and Biological
Radiation Source Search	Medical Recording and Registry
Radiation Survey and Bioassay Sampling	Haematology and Cytogenetic Biodosimetry Sampling

Specialized radiation teams are accessible through United Nation agencies. In 2000 IAEA established the "Response Assistance Network" or RANET [16], previously called Emergency Response Network (ERNET), of teams suitably qualified to respond rapidly and, in principal, on a regional basis, to nuclear or radiological emergencies. RANET's areas of assistance include: i) advisory, ii) assessment and evaluation, iii) monitoring, and iv) recovery. WHO's Radiation Emergency Medical Preparedness and Assistance Network (REMPAN) [17] consists of biodosimetry laboratories with expertise in: cytogenetic, EPR, bioassays, and molecular biology methodology. Recent efforts by WHO are focused to implement and coordinate a global network of reference biodosimetry laboratories (Figure 1).

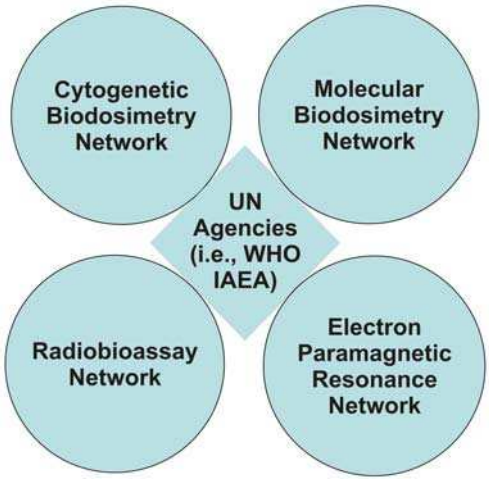

FIG. 1. Illustration of networks of expert reference laboratories specializing in dose assessment by cytogenetic biodosimetry, electron paramagnetic resonance (EPR), molecular biodosimetry, and radioactivity measurements from biological samples or radiation bioassay.

Koscheyev and colleagues [18] described that response teams responding to disasters can provide considerable benefits to both medical and psychological public-health problems. They also recommend use of a mobile diagnostic and a continuous operating pre-hospital triage system for rapid health screening of large populations after a large-scale disaster.

3. Biodosimetry - concept of operations

The primary purpose for early-response biodosimetry following suspected radiation overexposures is to rapidly provide first-responders and medical providers scientifically sound diagnostic radiation injury and dose assessment to support medical management treatment decisions. The measurement of clinical signs and symptoms associated with the severity of organ (i.e., hematological, gastrointestinal, neurovascular, and cutaneous) specific ARS, as developed and advocated by Prof. Fliedner (Ulm, Germany), is essential for triage of victims [19-20]. The risk of death from life-threatening radiation exposures is dependent on the level of medical care available (FIG. 2A). The U.S. Strategic National Stockpile (SNS) Radiation Working Group [12] recommended a treatment approach using both the organ specific clinical signs and symptoms based on the **Me**dical **Tre**atment **P**roto**col**s for Radiation Accident Victims (METREPOL) diagnostic system along with biological dosimetry (i.e., time to onset of nausea and vomiting, decline in absolute lymphocyte counts over several hours to days after exposure, and appearance of chromosome aberrations (i.e., dicentrics and rings). In the case of a mass casualty radiation emergency, this working group recommended cytokine, antibiotic, and stem-cell transplant therapies, as illustrated in the dose windows shown in Figure 2B. The SNS Radiation Working Group also encouraged cytokine therapy to be initiated 24 h after radiation exposure, based on the preclinical studies by MacVittie and colleagues [21]. This will likely necessitate an initial reliance on diagnostic information based on early bioindicators of radiation dose, which will then be replaced by bioindicators of the severity of ARS response as the clinical case evolves.

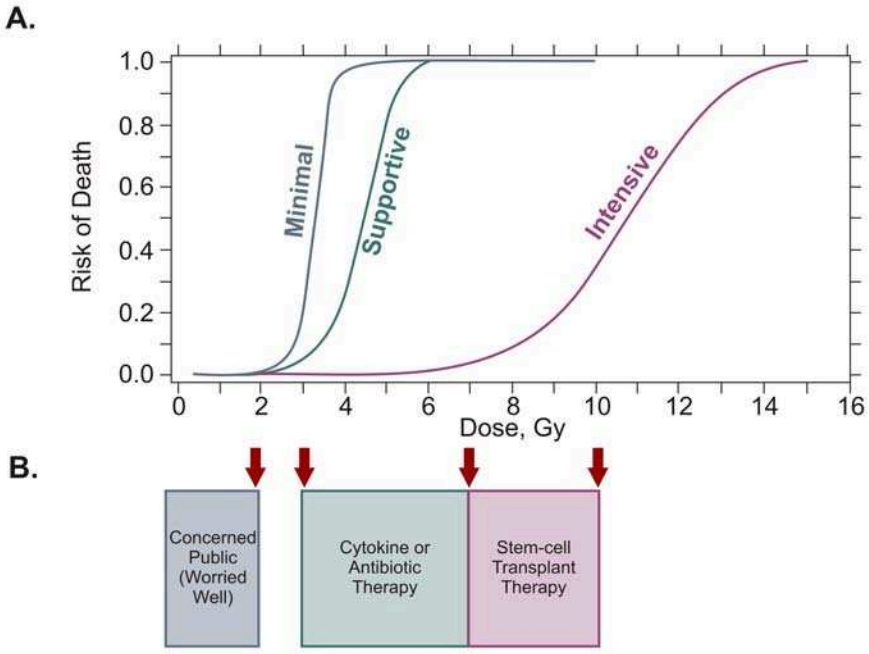

FIG. 2. Risk of death for various medical support conditions and dose windows for recommended treatments based on the U.S. Strategic National Stockpile Radiation Working Group [20].

The implementation of a multiparameter biodosimetry assessment approach is a significant confounder in a mass casualty radiological emergency. The U.S. Radiation Emergency Assistance Center/Training Site (REAC/TS) has developed a "Radiation Patient Treatment" treatment management approach that has incorporated early-response and multiple parameter biodosimetry when responding to a radiation incident with trauma or illness. The Armed Forces Radiobiology Research (AFRRI) has modified this approach, with permission from REAC/TS and incorporated it into AFRRI Pocket Guide – Emergency Radiation Medicine Response (released July 2008); see website: www.afrri.usuhs.mil. Figure 3 illustrates the components of the REAC/TS and AFRRI treatment strategy along with the concept of operations for use of multiparameter biodosimetry.

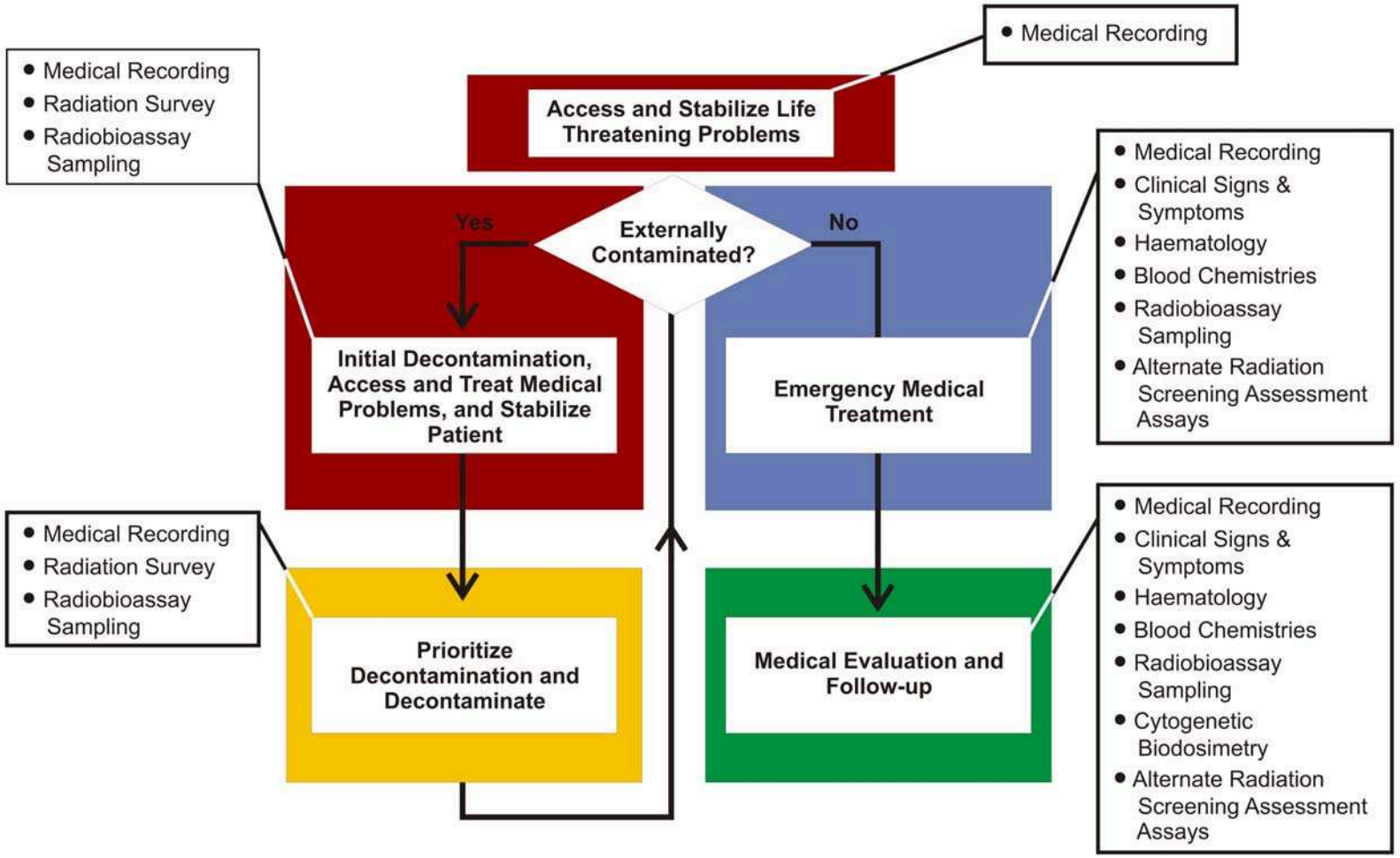

FIG. 3. Biodosimetry concept of operations during management of radiation incident with trauma or illness. Biodosimetry functions are illustrated for the individual action steps of the REAC/TS and AFRRI "Radiation Patient Treatment" algorithm.

A Joint Interagency Working Group (JIWG) under the auspices of the U.S. Department of Homeland Security Office of Research and Development conducted a technology assessment of emergency radiological dose assessment capabilities [22]. Gaps were identified to provide rapid radiation exposure triage. Current approaches and emerging technologies that offer potential to contribute in radiation injury and dose assessment response were identified. Research and development are needed to establish a diagnostic pyramid triage concept to facilitate a functional biodosimetry concept of operations in a mass-casualty radiation emergency (Figure 4). The initial screening radiation assay must be rapid (1 assay per minute or less), use a hand-held device, and ideally involve a self-use test. Secondary and tertiary radiation assay may require more expertise and take longer (<1 day) for use but have higher radiation specificity.

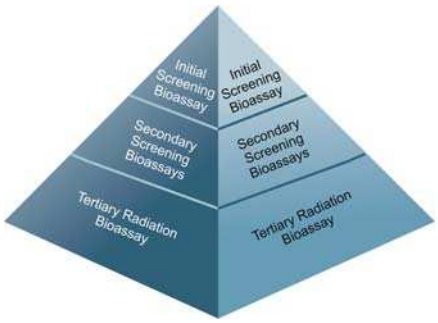

FIG. 4. Illustration of the diagnostic pyramid triage concept for mass-casualty radiation emergencies.

4. Early-response multiple parameter biodosimetry

4.1. Medical recording for radiation incidents

Medical recording is essential for effective diagnosis and medical management of radiation incidents. This is applicable at the incident scene as well as during transport to and while at the medical treatment facility. Medical recording guidance supporting the management of radiation casualties is available from the IAEA (see worksheets of IAEA's Generic Procedures for Medical Response During Nuclear and Radiological Emergency, EPR-Medical) [23]. AFRRI has approached this requirement using medical recording forms in annotable PDF format and medical recording and dose assessment software (Figure 5).

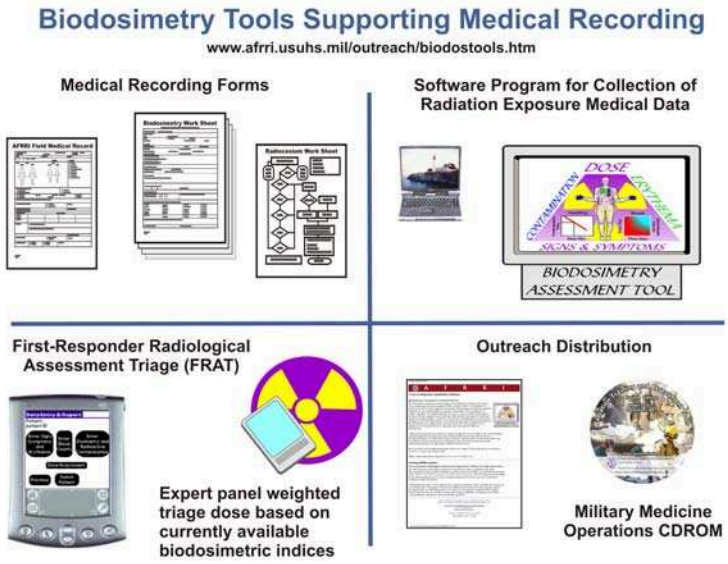

FIG. 5. AFRRI's biological dossimetry tools supporting medical recording.

4.1.1 Medical recording forms

Medical recording for radiation incidents should be consistent with an "all hazards" approach used by first responders. AFRRI's Adult/Pediatric Field Medical Record (AFRRI Form 330) provides a medical record template in a convenient one-page form for gathering emergency medical information in the field. It is applicable to both adult and pediatric cases. The form is provided as Appendix A.

The AFRRI Biodosimetry Worksheet (AFRRI Form 331) represents a comprehensive data entry worksheet, recently expanded from four to six pages to accommodate a modified version of METRPOL ARS severity scoring system. It provides a place for recording the facts about a case of radiation exposure, including the source and type of radiation, the extent of exposure, relevant biodosimetry diagnostic information, and the nature of the resulting injuries. The form is applicable to both adult and pediatric cases and is provided as Appendix B.

4.1.2 Medical recording and dose assessment software

The Biodosimetry Assessment Tool (BAT) [1, 24-25] program (version 1.0) for Windows XP was released on September 21, 2007, to the Board of Governors of the AFRRI, an agency of the U.S. Department of Defense. BAT was developed by AFRRI scientists as a tool to record and deliver diagnostic information (clinical signs and symptoms, physical dosimetry, etc.) to federal health care providers responsible for the management of radiation casualties. It is designed primarily for early use after a radiation incident and permits collection, integration and archiving of data obtained from patients accidentally exposed to ionizing radiation. Collection of relevant data is facilitated by use of structured templates and user-friendly software. This enables the generation of diagnostic indices for the development of a multiparameter dose assessment. The BAT program is NOT a substitute for treatment decisions by physicians and other trained health care professionals. Additional clinical parameters (i.e., infection, treatments, etc.) useful for casualty management also are assessed. The resulting display of patient diagnostic information provides treating health care providers with concise and relevant information on which to base clinical decisions. This information can be archived for further use in radiation protection management. An integrated, interactive human body map permits recording radioactivity detected by an appropriate radiation detection device. BAT is distributed on-line upon review of a download request application accessible at website www.afrri.usuhs.mil.

The First-responders Radiological Assessment Triage (FRAT) [1, 26] program will enable first responders to triage suspected radiation casualties based on the initial, or prodromal, features listed in the Emergency Radiation Medicine Response—AFRRI Pocket Guide. FRAT is being developed initially for the Palm operating system and may eventually be available for other PDA devices. With minimum text entry, FRAT will provide (1) signs and symptoms, (2) blood lymphocyte counts, and (3) dosimetry data. The program will assess the multiparameter triage dose or the exposure without an assigned dose, or it will indicate there is no evidence of overexposure. Additional FRAT output features include triage dose-specific messages addressing (1) reliability and diagnostic information, (2) hospitalization estimations, and (3) mortality projections. At this time AFRRI solicits individuals to contribute in beta testing the application.

4.2 Triage biodosimetry

4.2.1 Radioactive contamination

The body location of radioactive contamination, internal contamination information, the dose estimation based on location, and the dose based on personnel dosimeters, if available, should be recorded by first responders and medical personnel. The BAT application provides templates for recording these and other relevant parameters (location and activity of radiation source, patient location relative to radiation source, etc.) that can contribute to medical management and dose reconstruction. Metallic (or other) fragment samples should be collected for isotope classification, as appropriate, for identifying the radiation exposure scenario. In addition, biological samples (i.e.,

urinalysis, fecal, wound, swipes from body orifices) should also be collected for determining the committed dose. AFRRI's FRAT application uses information about radioactive contamination not eliminated after removal of clothes and washing as evidence for radiation exposure in a triage dose assessment algorithm.

4.2.2 Clinical signs and symptoms

The time onset and severity of early prodromal phase signs and symptoms can provide some valuable information regarding the absorbed "dose range." The early or prodromal phase response from exposure to ionizing radiation is characterized by a dose-dependent expression of a constellation of signs and symptoms [27] including nausea, vomiting, anorexia, and central nervous system function impairment. The FRAT application integrates these prodromal signs to provide a triage dose assessment. Progressive increases in radiation dose result in an increased percentage of both the incidence and the constellation of prodromal signs and symptoms. The appearance of acute symptoms, such as vomiting, is directly dependent on the radiation dose to an overexposed individual [27] and contributes to the multiparameter diagnostic index (Table III) used for assessing dose. Following photon and criticality accident exposures, the BAT program can be used to record prodromal symptoms and access dose prediction models for the prodromal symptom, time onset of vomiting [27-30]. An acute photon exposure dose of 2 Gy would cause ~50% of individuals to exhibit emesis approximately 4.6 hours post-irradiation. However, since potential confounders (flu epidemic, etc.) can also induce similar symptoms, caution is warranted when using selective prodromal symptoms alone to assess dose for efficient treatment of the accident victim. For example, the incidence of psychogenic vomiting would likely be elevated during stressful events such as a radiological mass casualty incident.

The location and time-course of radiation-induced cutaneous injury should be recorded. Reddening of the skin, or initial erythema, is generally seen within a few hours to a few days following exposure to a high radiation dose (>2 Gy) and lasts only for a day or two. This information provides diagnostic information concerning partial- or whole-body exposures and can later help define the boundary of the radiation exposure area when skin graphs are necessary. The AFRRI Biodosimetry Worksheet (Appendix B) and BAT program provide data templates for this purpose. The skin's response to radiation is biphasic, and this type of skin reaction is largely due to capillary dilation caused by the release of histamine-like substances. Erythema increases during the first week following exposure and then generally subsides during the second week. It may return 2–3 weeks after the initial insult and last up to 30 days, and additional changes, such as desquamation, bullae formation, or even skin sloughing may follow, all of which make even a crude estimation of radiation dose almost impossible.

4.2.2. Haematology

Haematological responses are an early response biomarker for radiation dose assessment and also contribute in the assessment of the severity of haematology ARS. Fliedner advocates the use of blood cell changes after whole-body radiation exposures are reliable bioindicators of injury and a critical aid to plan therapeutic treatments [31]. An approximate 50% decline occurs in peripheral blood lymphocyte counts over 12 hours that fall below normal vales (1.4×10^9/L) is indicative of a potential severe radiation overexposure [12]. Goans and colleagues introduced lymphocyte depletion kinetic models for dose estimates based on human radiation accident registry data for whole-body acute gamma exposures [28] and more recently for criticality accidents [29]. Immediately following exposure, a complete blood cell count (CBC) with white cell differential should be obtained and then taken three times a day for the next 2–3 days and twice a day for the following 3–6 days. The BAT program permits the recording of peripheral blood lymphocyte counts and then converts them into dose predictions using lymphocyte depletion kinetic models based on previous dose responses in

Table III. Biodosimetry Based on Acute Photon-Equivalent Exposures*

Dose Estimate	Time to Onset of vomiting		Absolute Lymphocyte count (x10^9/liter)[b] (Day)						Lymphocyte depletion rate[c]	Relative increase in serum amylase activity at 1 d compared with normals[d]	Number of dicentrics[e]	
Gy	%[a]	Time (Hr)	0.5	1	2	4	6	8	Rate constant		Per 50 metaphases	Per 1000 metaphases
0	--	--	**2.45**	**2.45**	**2.45**	**2.45**	**2.45**	**2.45**	--	1	0.05 – 0.1	1-2
1	19		**2.30**	**2.16**	**1.90**	**1.48**	1.15	0.89	0.126	2	4	88
2	35	4.63	**2.16**	**1.90**	**1.48**	0.89	0.54	0.33	0.252	4	12	234
3	54	2.62	**2.03**	**1.68**	1.15	0.54	0.25	0.12	0.378	6	22	439
4	72	1.74	**1.90**	**1.48**	0.89	0.33	0.12	.044	0.504	10	35	703
5	86	1.27	**1.79**	1.31	0.69	0.20	0.06	.020	00.63	13	51	1034
6	94	0.99	**1.68**	1.15	0.54	0.12	0.03	.006	0.756	15		
7	98	0.79	**1.58**	1.01	0.42	.072	.012	.002	0.881	16.5		
8	99	0.66	**1.48**	0.89	0.33	.044	.006	<.001	1.01	17.5		
9	100	0.56	1.39	0.79	0.25	.030	.003	<.001	1.13	18		
10	100	0.48	1.31	0.70	0.20	.020	.001	<.001	1.26	18.5		

* Table modified from version reported by Waselenko and colleagues [12]. Depicted above are the four most useful elements of biodosimetry. Dose range is based on acute photon-equivalent exposures. The first column indicates the percent of people who vomit, based on dose received and time to onset. The middle left section depicts the time frame for development of lymphopenia. Two or more determinations of blood lymphocyte counts are made to predict a rate constant which is used to estimate exposure dose. The middle right section shows the relative increase in serum amylase activity in humans 1 day after radiation exposure. The final column represents the current "gold standard" which requires several days before results are known. CSF therapy should be initiated when onset of vomiting, lymphocyte depletion kinetics, and/or serum amylase suggests an exposure dose for which treatment is recommended. Therapy may be discontinued if results from chromosome dicentrics analysis indicate lower estimate of whole-body dose.

[a] Cumulative percentage of victims with vomiting.

[b] Normal range: 1.4-3.5x10^9/L. Numbers in bold fall within this range.

[c] The lymphocyte depletion rate is based on the model Lt = 2.45 x 109/L x e-k(D)t where Lt equals the lymphocyte count (x10^9/L), 2.45 x 10^9/L equals a constant representing the consensus mean lymphocyte count in the general population, k equals the lymphocyte depletion rate constant for a specific acute photon dose, and t equals the time after exposure (days).

[d] Relative increases in serum amylase activity compared with normals [42].

[e] Number of dicentric chromosomes in human peripheral blood lymphocytes.

radiation accidents [27, 28, 32]. Lymphocyte cell counts and lymphocyte depletion kinetics provide dose assessment predictions that fall in the equivalent photon dose range of 1–10 Gy; see Table III.

4.2.4 Blood chemistry assay

Blood biochemical markers of radiation exposure have also been advocated for use in early triage of radiation casualties [26, 33-35]. An increase in serum amylase activity (hyperamylasemia) from the irradiation of salivary tissue has been proposed as a biochemical measure of early radiation effect in a normal tissue [36-37]. Several studies have also advocated it use as a candidate biochemical dosimeter in man [26, 38-40]. A few hours after irradiation injury, cells in the salivary gland show acute inflammation and degenerative changes resulting in increases in serum amylase activity. Histochemical, isozyme analysis, and partial-body exposure studies confirm that the increase in serum amylase activity originated from the salivary glands. Serum amylase activity increases occur early after head and neck irradiation of humans [41] and generally show peak values between 18-30 h after exposure, returning to normal levels within a few days [42]. Sigmoidal dose-dependent increases in the early (1 day) hyperamylasemia are supported by radio-iodine therapy [43-44], radiotherapy [38, 39, 45-46], and from limited data from three individuals exposed in a criticality accident [40]. Table III shows a dose response for relative increases in serum amylase activity 1 d after exposure. Significant inter-individual variations are reported in dose-response studies [37, 42, 45-46], which represent a potential major confounder for use of serum amylase activity alone as a reliable biodosimeter. This inter-individual variation in biochemical response is not unexpected, since it is well known that the radiation level causing irreversible failure of the hematopoietic system varies among individuals and may reflect genetic and physiological differences and relative differences in the radiosensitivity of hematopoietic stem/progenitor cells [47] as well as radiation exposure parameters (i.e., partial-body exposures, shielding, dose-rate, etc.) [48].

4.2.5 Triage chromosome aberration cytogenetics

The quantification of the yield of chromosome aberrations (e.g., dicentric and rings) in lymphocyte metaphase spreads is one of the principal methods for estimating radiation dose to exposed individuals. The generation of dose-response calibration curves is required for the proper evaluation of the lymphocyte-dicentric changes, which requires a high level of expertise and is typically performed in expert reference laboratories.

Dr. David C. Lloyd (National Radiological Protection Board, UK) suggested that cytogenetic triage using a lymphocyte metaphase-dicentric assay could be especially useful in providing evidence of non-uniform exposure and confirmation of individuals in a high-dose, exposed triage category [49]. In individuals with a high-dose estimate based on the mean number of dicentrics per cell, a significant fraction of metaphase spreads free of dicentrics suggests that surviving functional stem cells are present and that these individuals would be potential candidates for cytokine therapy versus a bone marrow transplant.

In managing radiation accidents, it is important to triage patients into broad, 1-Gy dose windows, especially when there are mass casualties and limited resources. Chromosome-aberration analysis using the conventional cytogenetic metaphase-spread dicentric bioassay is useful for the initial triage of mass casualties [4, 7, 12, 50-52]. In the triage mode only 40 to 50 metaphase spreads per subject are scored (fewer if the aberration yield is high) instead of the typical 500 to 1000 scored in a routine analysis. Table III demonstrates the expected triage (50 metaphases) and reference (1000 metaphases) yield of dicentrics for acute photon doses from 1 to 5 Gy. After the initial results are communicated to the treating physician, additional scoring is advisable so that potential dose-assessment conflicts can be resolved, and assistance can be provided for physicians considering marrow stem cell transplants in high dose cases. Specialized cytogenetic biodosimetry laboratories need to develop and routinely practice emergency cytogenetic biodosimetry triage procedures.

4.2.6 Provisional and emerging triage, clinical, and definitive dose assessment methods

Several provisional and emerging approaches have been considered as methods to provide triage, clinical, and/or definitive dose assessment. For a review of these and other established dose assessment methods see reports by Alexander and colleagues [2] and Joint Interagency Working Group [22] and Table IV. Electron paramagnetic resonance (EPR)-based detection of free radicals is a well accepted and validated method for measurement of dose to dental enamel from biopsy teeth [2; 53] and has recently been extended to measure absorbed dose from teeth *in vivo* and nail clippings *ex vivo* [2]. Radiation causes injury to various tissues and organs resulting in time- and dose-dependent increases in blood. These proteins are bioindicators for radiation injury of relevant ARS organ systems (i.e., bone marrow, gastrointestinal system) as well as early bioindicators of absorbed dose. Gene array methods have been used to identify candidate radiation-response gene expression targets derived from blood lymphocytes and then measured by quantitative real-time reverse transcriptase polymerase chain reaction (QRT-PCR) methods. See Table 4 below for a select listing and status of these provisional and emerging methods.

Table IV. Select List of Provisional and Emerging Radiation Injury and Dose Assessment Methods

Method	Status	References
EPR		
- teeth (*in vivo*)	EPR L-band is potentially able to measure doses as low as 2 to 3 Gy but needs additional development	2; 54-56
- nails (*ex vivo*)	EPR X-band shows a lower limit of detection of 0.5 - 1 Gy	2; 57-59
Blood protein immunoassay		
- C-reactive protein	Acute-phase reaction protein derived from liver and demonstrated both as a biodosimeter and bioindicator of hematology ARS	60-62
- Flt-3 ligand	Bioindicator of bone marrow injury	63-64
- Citrulline	Bioindicator of injury to small intestine epithelial tissue	65-67
- γH2AX	Protein associated with DNA double strand break repair	68
- Multiple proteins	Candidate multiple protein biomarkers proposed for biodosimetry; multiple protein biomarkers demonstrated using multivariate discriminant or linear regression analyses methodology for radiation injury	69-70
Blood lymphocytes gene expression		
- QRT-PCR assay of multiple targets	Multiple radiation responsive gene targets identified and used in the development of consensus dose-response calibration curves using an *ex vivo* blood radiation model system	71-75

5. Recommended biodosimetry enhancements for mass-casualties radiological incidents

In general first responders and medical care providers at hospitals have limited capabilities for assessing radiation injury, especially in the event of a radiological mass casualty incident. Further no single radiation bioassay at present is sufficient to provide robust dose-assessment capabilities for potential radiation exposure scenarios including mass casualties. A multiple parameter approach is necessary for triage, clinical, and definitive radiation biodosimetry [1]. A biological dosimetry

approach for medically managing of a mass-casualty radiological emergency should include local, national, and international cooperation to: i) train and equip local responders with tools to provide early radiological triage capability, ii) establish deployable teams (equipped with hand-held and licensed devices to assess radiation exposure) and iii) access specialized reference laboratories (equipped with automation technologies to enhance their throughput).

Nations need access to expert radiobioassay, cytogenetic biodosimetry, molecular biodosimetry, and EPR dose assessment reach-back service laboratories. These capabilities provide nations with definitive dose assessment supporting radiation protection programs. Implementation of automation methods for these dose assessment methods have merit and should be linked with established service laboratories. Efforts to establish laboratory networks composed of national and international reference laboratories able to respond to a sudden surge of analysis requests from a mass-casualty incident should be encouraged and facilitated.

Radiological teams able to rapidly deploy to the emergency incident are essential to respond to mass casualty events and should follow guidelines recommended by IAEA's RANET [16] and/or WHO's REMPAN [17] programs. These deployable teams capable of performing triage biodosimetry assays (blood cell counts, signs and symptoms assessments, and radioactivity biosampling) should also be exercised. The teams will need training [76] and be equipped with necessary supplies and equipment.

The development of biomarkers for biodosimetry has been identified as priority efforts to help the U.S. prepare for the possibility of a terrorist attack using radiological or nuclear devices [12, 75]. Sustained research support is needed to identify, optimize, and validate novel hematological, cytological, and molecular radiation responsive biomarkers and biophysical dose assessment methods. The results from this effort should provide the basis for development of rapid and high-throughput applications leading towards licensed and effective hand-held and laboratory devices for assessing radiation exposure.

Finally, the countermeasures to enhance national and international medical response capabilities for radiological incidents, including mass casualty events, need to be integrated into the first-responder community "all hazard" response concept in order to be effectively assimilated and sustained. Future hand-held and deployable laboratory devices used to measure radiation exposure based on a biological sample should have dual-use capabilities for assessing exposure to other threat agents (i.e., chemical and biological), which is consistent with an "all hazard" approach. These research developments supporting biological dosimetry should be rapidly incorporated into local, national, and international "radiological exercises" to enhance training for responders, and to increase knowledge of policy managers and the general public for the important role of biodosimetry in mass-casualty radiological emergencies.

6. Summary

- A coordinated integration of local and national radiological response capabilities that are supplemented with international cooperation can provide critical biological dosimetry capabilities to support the medical management of a mass-casualty radiological emergency.

- Major gaps in the biodosimetry response capability for mass-casualty radiological emergencies have been identified and include:
  - the capability to rapidly identify exposed individuals using licensed diagnostic hand-held or field-laboratory systems;
  - protocols to measure radioisotopes likely used by terrorists from contaminated individuals;
  - enhance assess to deployable radiological teams with capabilities to perform on-site haematology, assessment of clinical signs and symptoms, and sampling for radiobioassays;
  - funding to establish and sustain functional global networks of expert reference laboratories performing dose assessment.

- International cooperation will enhance biological dosimetry capabilities through sharing of research discoveries, nations participating in U.N. agencies radiological assistance programs, and research efforts focusing on applications for applied radiological biodosimetry.

7. Acknowledgments

The views expressed here are those of the authors; no endorsement by AFRRI has been given or inferred. AFRRI supported this research under work units RAB4AL and RAB4AM. The Technical Support Working Group also helped support this research under work unit TSWG-132DI. The authors gratefully acknowledge the assistance of the present (COL L. Andrew Huff, M.D.) and previous (COL John Mercier, Ph.D.; COL William E. Dickerson, USAR ret; COL John Jacocks, USA ret., COL David Jarrett, USA) Directors and co-workers of AFRRI's Military Medical Operations Department, present co-workers (Natalia I. Ossetrova Ph.D.; Ira H. Levine; David. K. Sandgren, MAJ James A. Ross, COL Patricia Lillis-Hearne M.D., Gregory L. King, Ph.D.) and past AFRRI co-workers (William E. Jackson, Pataje G.S. Prasanna, Ph.D.; Alexandra C. Miller, Ph.D.), collaborators (Ronald E. Goans, Ph.D, MJW Corporation and Tulane University, New Orleans, Louisiana, USA; David Holt, PhD, Institute of Naval Medicine, United Kingdom; Diana Wilkinson, Ph.D., Defence Research & Development Canada, Ottawa, Canada; and George H. Anno, Ph.D., Pacific-Sierra Research Corp., Los Angeles, California, USA; Marcy B. Grace Ph.D, Washington, D.C., USA), and present (HM3 Sergio Gallego) and former (Karen Salomon) research assistants. The author also wishes to acknowledge and thank Dr. Bobby Scott (Lovelace Respiratory Research Institute, Albuquerque, New Mexico, USA) for use of Figure 2A.

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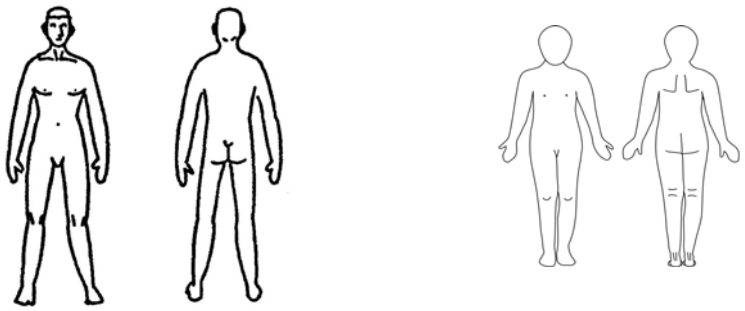
APPENDIX MATERIAL

Pages

1. Name (last, first)		Rank/Grade	☐ Male ☐ Female
SSN	Specialty code		Religion
2. Unit	Force		Nationality
☐ A ☐ AF ☐N ☐ MC ☐ Civilian			

Adult/Pediatric Field Medical Record

Adapted from DD Form 1380, U.S. Field Medical Card

☐ BC ☐NBI	☐ Disease	☐ Psych
3. Injury Adult Front Back Child Front Back 		☐ Airway ☐ Head ☐ Wound ☐ Neck/back injury ☐ Burn ☐ Amputation ☐ Stress ☐ Other (specify)

4. Level of consciousness				
☐ Alert			☐ Pain response	
☐ Verbal response			☐ Unresponsive	
5. Pulse	Time	**6.** Tourniquet ☐ No ☐ Yes		Time
7. Morphine ☐ No ☐ Yes	Dose	Time	**8.** IV	Time
9. Treatment/observations/current medication/allergies/NBC (antidote)				
10. Disposition	☐ Returned to duty ☐ Evacuated ☐ Deceased			Time
11. Provider/unit				Date (YYMMDD)
12. Reassessment				
Date (YYMMDD)		Time of arrival		
Time				
BP				
Pulse				
Resp				
Date/time	**13.** Clinical comments/diagnosis			
	14. Orders/antibiotics (specify)/tetanus/IV fluids			
15. Provider				Date (YYMMDD)
16. Disposition	☐ Returned to duty ☐ Evacuated ☐ Deceased			Time
17. Religious services	☐ Baptism ☐ Anointing ☐ Confession	☐ Prayer ☐ Communion ☐ Other	Chaplain	

Biodosimetry Worksheet
(Medical Record of Radiation Dose, Contamination, and Acute Radiation Sickness Response)

Reporting Authority (person(s) creating this page of the report)			
Name (last, first):			
Unit:	Country of origin:		
Phone:	Fax:	E-mail:	
Location:	Date (yymmdd):	Time:	

Casualty		
Name (last, first):		Rank:
Parent unit:		Parent unit location:
Parent unit phone:	Country of origin:	Parent unit e-mail:
Parent unit FAX:	Location of casualty:	
History of presenting injury (conventional and/or radiation): (Use page 6 for additional space.)		
History of previous radiation exposure: (Use page 6 for additional space.)		
Past medical history (general): (Use page 6 for additional space.)		
Medical countermeasures (e.g., antiemetics, transfusion), specify: (Use page 6 for additional space.)		
Administered (where, when, route):		

Exposure conditions	
Date of exposure (yymmdd):	Exposure location:
Time of exposure:	Weather conditions (at time of exposure):
Exposure results	
Description of incident: (Use page 6 for additional space.)	
External exposure overview	
Body exposure: ☐ Total ☐ Partial ☐ Uncertain	Shielding confounder: ☐ Yes ☐ No
Contamination overview	
External contamination: ☐ Yes ☐ No	Contaminated wound: ☐ Yes ☐ No
Internal contamination: ☐ Yes ☐ No	If yes, describe (Use page 6 for additional space.):

Biodosimetric assays overview	Sampling date, time (yy:mm:dd:time)	Estimated time post-exposure (h)	Dose (Gy)	Reference radiation
Time onset of vomiting				
Lymphocyte counts or depletion kineti				
Urine bioassay				
Cytogenetic biodosimetry				
Other				

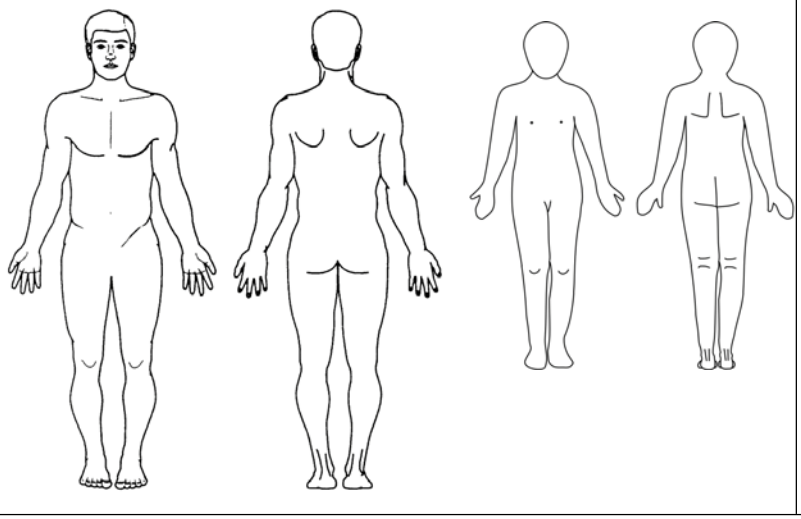
Patient's identification code: _____

Contamination: Dose Assessment (person(s) creating this page of the report)		
Name (last, first):		Unit:
Phone:	Fax:	E-mail:
Country of origin:	Place:	
Date dose assessed (yymmdd):	Time and method dose assessed:	

Contamination: external/internal contamination	
Substance trademark (if applicable):	
Solid: ☐ Yes ☐ No	Gaseous (G): ☐ Yes ☐ No
Particulate (P): ☐ Yes ☐ No	Aerosol (L/G): ☐ Yes ☐ No
Liquid (L): ☐ Yes ☐ No	Aerosol (P/G): ☐ Yes ☐ No
Radionuclide(s):	Chemical compound(s):
Activity (Bq):	

Contamination distribution

Adult **Child**



Comments:
(Use page 6 for additional space.)

Route of intake (in case of internal contamination)	
Inhalation: ☐ Yes ☐ No	Injection: ☐ Yes ☐ No
Cutaneous: ☐ Yes ☐ No	Other: ☐ Yes ☐ No
Ingestion: ☐ Yes ☐ No	If yes, specify:

Contamination assessment	
Contamination measurement:	Detection device:
Counts per minute:	Estimated activity:
Decontamination measures:	Residual contamination:
Measures taken to prevent uptake:	
Measures taken to minimize reabsorption:	
Measures taken to increase excretion:	

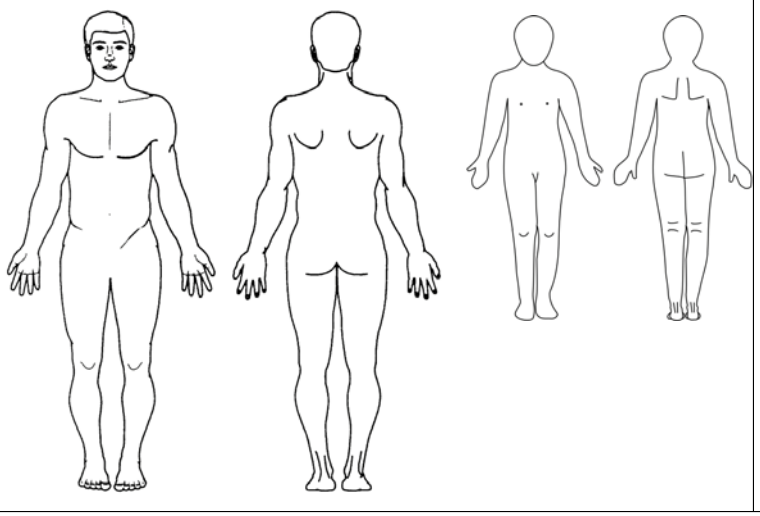
Patient's identification code: _____

External Exposure: Dose Assessment (person(s) creating this page of the report)		
Name (last, first):		Unit:
Phone:	Fax:	E-mail:
Country of origin:	Place:	
Dose assessment date (yymmdd):	Dose assessment time:	

Nature of exposure: radiation source		
Alpha (α): ☐ Yes ☐ No	Beta (β): ☐ Yes ☐ No	Neutron (n): ☐ Yes ☐ No
Gamma (γ): ☐ Yes ☐ No	X-ray (x): ☐ Yes ☐ No	Mixed n/γ: ☐ Yes ☐ No
Dose rate (at distance measured from):		Distance to source:
Activity of source (if known):		Duration of exposure:
Confounding factors used in dose reconstruction (e.g., shielding): ☐ Yes ☐ No		
Type of dosimeter (if applicable):		Body location of dosimeter:
Facility where dosimeter was read:		Dosimeter reading:
Biological dosimetry type and facility where performed (if applicable):		

Dose distribution

Adult **Child**



Comments:
(Use page 6 for additional space.)

Blood chemistry analysis				
	First	**Second**	**Third**	**Fourth**
Date collected (yymmdd)				
Time collected				
Date analyzed (yymmdd)				
Time analyzed				
Serum amylase (U/L): (Reference value: 21–160 U/L)				
Serum C-reactive protein (mg/L): (Reference value: ~1 mg/L)				
Other				

Patient's identification code:_____

ARS Responses Assessment: (person(s) creating this page of the report)		
Name (last, first):		Unit:
Phone:	Fax:	E-mail:
Country of origin:	Place:	

Signs and Symptoms (none: 0; degree of max. severity: 1 (mild) to 4 (severe); see page 5 for severity degrees)						
Date assessed (yymmdd)						
Time assessed						

Neurovascular system	**Degree of severity**					
Nausea:						
Vomiting: (Note time of initial onset on page 6)						
Headache:						
Anorexia:						
Fever:						
Hypotension:						
Tachycardia:						
Neurological deficits:						
Cognitive deficits:						
Fatigue/weakness:						
Maximum grading N:						

Cutaneous system	**Degree of severity**					
Erythema: (Note body location on page 6)						
Pruritis (itching):						
Edema:						
Bullae (blisters):						
Desquamation:						
Ulcer or necrosis:						
Hair loss:						
Onycholysis:						
Maximum grading C:						

Gastrointestinal system	**Degree of severity**					
Diarrhea: • Frequency:						
• Consistency:						
• Melena (bloody stools):						
Abdominal cramps or pain:						
Maximum grading G:						

Hematopoietic system	**Blood cell counts and degree of severity**											
(C=cell count D=ARS degree)	C	D	C	D	C	D	C	D	C	D	C	D
Lymphocytes (× 10^9)/liter:												
Granulocytes (× 10^9)/liter:												
Neutrophils (× 10^9)/liter:												
Platelets (× 10^9)/liter:												
Blood loss:												
Infection:												
Maximum grading H:												

RC =						
Days after exposure:						

Patient's identification code: _____

Appendix. Grading System for Response of Neurovascular, Gastrointestinal, Cutaneous, and Hematopoietic Systems

Symptom	Degree 1	Degree 2	Degree 3	Degree 4
Neurovascular system				
Nausea:	Mild	Moderate	Intense	Excruciating
Vomiting:	Occasional (one per d)	Intermittent (2–5 times per d)	Persistent (6–10 times per d)	Refractory (> 10 times per d)
Headache:	Minimal	Moderate	Intense	Excruciating
Anorexia:	Able to eat & drink	Intake decreased	Intake minimal	Parenteral nutrition
Fever:	< 38°C	38–40°C	> 40°C for < 24 h	> 40°C for > 24 h
Hypotension:	Heart rate >100 beats/m; blood pressure > 100/70 mm Hg	Blood pressure < 100/70 mm Hg	Blood pressure < 90/60 mm Hg: transient	Blood pressure < 80/? mm Hg; persistent
Neurological deficits:	Barely detectable	Easily detectable	Prominent	Life-threatening, loss of consciousness
Cognitive deficits:	Minor loss	Moderate loss	Major impairment	Complete impairment
Fatigue/weakness:	Able to work	Interferes with work or normal activity	Needs assistance for self care	Prevents daily activities
Cutaneous system				
Erythema:	Minimal, transient	Moderate (< 10% body surface area)	Marked (10–40% body surface area)	Severe (> 40% body surface area)
Pruritis (itching):	Sensation of itching	Slight and intermitten pain	Moderate and persistent pain	Severe and persistent pain
Edema:	Persistent, asymptomatic	Symptomatic, tension	Secondary dysfunction	Total dysfunction
Blistering:	Rare, sterile fluid	Rare, hemorrhage	Bullae, sterile fluid	Bullae, hemorrhage
Desquamation:	Absent	Patchy dry	Patchy moist	Confluent moist
Ulcer or necrosis:	Epidermal only	Dermal	Subcutaneous	Muscle/bone involvement
Hair loss:	Thinning, not striking	Patch, visible	Complete, reversible	Complete, irreversible
Onycholysis:	Absent	Partial	Partial	Complete
Gastrointestinal system				
Diarrhea:				
• Frequency, stools/d	2–3	4–6	7–9	≥ 10; refractory diarrhea
• Consistency	Bulky	Loose	Very loose	Watery
• Melena (bloody stools)	Occult	Intermittent	Persistent	Persistent with large amount
Abdominal cramps/pain:	Minimal	Moderate	Intense	Excruciating
Hematopoietic system				
Lymphocyte changes: (reference value, 1.4–3.5 × 10^9 cells/L)	1–2d: ≥ 1.5	1–2d: 1–1.5	1–2d: 0.5–1	1–2d: < 0.5
	3–7d: ≥ 1	3–7d: 0.5–1	3–7d: 0.1–0.5	3–7d: < 0.1
Granulocyte changes: (reference value, 4–9 × 10^9 cells/L)	1–2d: ≥ 2	1–2d: 4–6; mild	1–2d: 6–10; moderate	1–2d: > 10; marked
	3–7d: ≥ 2	3–7d: > 2	3–7d: > 5	3–7d: > 5
Thrombocyte (platelets) changes: (reference value, 140–400 × 10^9 cells/L)	1–2d: ≥ 100	1–2d: 50–100	1–2d: 50–100	1–2d: 50–100
	3–7d: ≥ 100	3–7d: 50–100	3–7d: 20–50	3–7d: < 20
Blood loss:	Petechiae, easy bruising, normal hemoglobin level	Mild blood loss with < 10% decrease in hemoglobin level	Gross blood loss with 10%–20% decrease in hemoglobin level	Spontaneous bleeding or blood loss with > 20% decrease in hemoglobin level
Infection:	Local, no antibiotic therapy required	Local; only local antibiotic therapy required	Systemic; p.o. antibiotic treatment sufficient	Sepsis; i.v. antibiotics necessary

Comments: (Use this page for additional space.)

Adapted from:

1. NATO Standardization Agreement (STANAG 2474). Determination and Recording of Ionizing Radiation Exposure for Medical Purposes. Appendix 1, 2003.
2. Fliedner TM, Friesecke I, Beyrer K, eds. Medical Management of Radiation Accidents: Manual on the Acute Radiation Syndrome. Oxford: British Institute of Radiology; 2001. p. 1–66.
3. Gorin N-C, Fliedner TM, Gourmelon P, *et al*. Consensus conference on European preparedness for haematological and other medical management of mass radiation accidents. Ann Hematol. 2006; 85(10):671–679.
4. Radiation Event Medical Management (REMM). Guidance on Diagnosis & Treatment for Health Care Providers. Accessed 24 Oct 2007, from http://www.remm.nlm.gov/ars.htm.
5. Waselenko JK, MacVittie TJ, Blakely WF, *et al*. Medical management of the acute radiation syndrome: recommendations of the Strategic National Stockpile Radiation Working Group. Ann Int Med. 2004; 140:1037–1051.

Patient's identification code: _____