



B29 Decontamination of Human Autopsy Specimens by ⁶⁰Co Gamma-Photon Irradiation and Human DNA Identification by Short Tandem Repeat Analysis of Irradiated Tissues

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After attending this presentation, the participant will understand 1) the use of Cobalt 60 (⁶⁰Co) gamma-photon ionizing radiation for decontaminating human autopsy specimens and 2) the effect of decontaminating doses of gamma radiation on Short Tandem Repeat (STR) DNA analysis of tissue and bone specimens.

This presentation will impact the forensic community by demonstrating irradiation as a way to decontaminate human remains found in an incident involving contaminating weaponized microbial agents prior to submission of the laboratory for human identification by STR DNA analysis.

Short Tandem Repeat (STR) DNA typing has been the primary tool for identification of human remains in military and mass disaster incidents. The majority of the military specimens submitted to the Armed Forces DNA Identification Laboratory (AFDIL) include fresh tissues and bones collected by medical examiners during autopsies.

The potential use of military force in nations that may have biological weapons of mass destruction, such as weaponized *Bacillus anthracis* spores, and the potential for large numbers of fatalities from inhalation anthrax in these nations suggested that we study a method for decontaminating human remains. We investigated ⁶⁰Co gamma-photon irradiation as a method of decontamination.

In this study, 11 different human cadaveric finger specimens were collected from 6 female and 5 male deceased donors. DNA was organically extracted and then quantitated using the Quantiblot® method. STR profiles were obtained from a 3 to 5-mm piece of tissue from each of the donor fingers for a known DNA STR typing control prior to gamma irradiation. The organic DNA extraction and Profiler Plus™ STR profiling methodologies are rapid, reliable and robust technologies that have been validated for casework at AFDIL, accepted in the forensic community, and published in the forensic literature.

A STP-350 biological shipment container (SAF-T-PAK, Edmonton, AB, Canada) was chosen to safely contain biological specimens, endure the rigors of transport from a military theater of operation and the process of irradiation, and meet the biohazardous materials transportation regulations. The large orange suitcase-like shipment container has the internal capacity to fit eight biohazard canisters in a fitted polystyrene holder. The biohazard canisters can hold up to five specimen transport tubes and absorbent paper.

Each of eight specimen transport tubes contained 0.5 g of dry, freeflowing *Bacillus subtilis* var. *niger* (*B. globigii*) spores, i.e., biological indicators (BI), as a surrogate for *B. anthracis*, at a concentration of 5.5×10^{11} colony-forming units (CFU) per gram. A radiation dosimeter (Dalanine pellets) was placed in three of these tubes with the spores. Each of the 11 donor fingers was placed in a specimen tube containing 99.99% isopropanol. The BI's and the tissue specimens were then distributed in the eight canisters in mapped locations within the case.

Gamma irradiation of the shipment container and dosimetry were performed in the Cobalt-60 Irradiation Facility at the Armed Forces Radiobiology Research Institute. Electron paramagnetic resonance (EPR) dosimetry measurements of irradiated alanine pellets were performed at the National Institute of Standards and Technology (NIST, Gaithersburg, MD) using published procedures to determine the dose delivered. The case and contents were given three fractionated doses of gamma-photon radiation at a constant rate in order to achieve a decontamination assurance level (DAL) for the BI. The DAL is the dose of gamma radiation required to kill all spores in 99% of the BIs. Before and after irradiation, BI spore viability was determined by standard microbiological methods as defined in the 2002 United States Pharmacopeia 25th edition (USP25).

The gamma-photon radiation dose, which was delivered inside of the case, was an average of 51.7 kGy as measured by alanine dosimetry and was sufficient to kill all spores in test BI samples. DNA isolated from 2gram bone specimens and 3 to 5-mm cubed tissue specimens from each of the irradiated fingers yielded full Profiler Plus™ STR profiles that were consistent with the STR profiles obtained from the original non-irradiated donor source fingers. Mutations, extraneous alleles, and allelic dropouts were not observed in the Profiler Plus™ STR DNA profiles from the irradiated finger tissues and bones.

Conclusions: This study shows that gamma-photon irradiation decontaminates potentially contaminated human specimens and that the process does not adversely affect STR DNA analyses for human identification purposes.

Short Tandem Repeat DNA, Human Identification, Radiation