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## Optimization of radiation treatment of biological objects

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To meet the demands of irradiation centers and extend the range of biological objects which can be irradiated for different purposes, our research team has conducted a series of experimental studies to estimate the influence of irradiation parameters as well as physical and chemical properties of bio-objects on the irradiation efficiency. The study focuses on determining the criteria for choosing optimal dose range which would destroy pathogens while preserving essential molecules, such as proteins, lipids, and enzymes.

The study uses real-life objects, such as beef, turkey, chicken, salmon and trout, potato tubers, cereal and oil seeds as well as model objects, such as bacteria, fungi, phytopathogens, standard samples of volatile organic compounds and bovine serum albumin. The methodology of the research involves irradiation of bio-objects using the 1 MeV electron accelerator UELR-1-25-T-001 (SINP MSU, Russia) and X-ray apparatus DRON YM-2 with X-ray tube BSV 23 with copper anode and X-ray apparatus RAP 100-10 with X-ray tube 1BPV 23-100 with molybdenum anode (Burnazyan Federal Medical Biophysical Center of FMBA, Russia). Irradiation of bio-objects is simulated using GEANT 4 toolkit to estimate the dose uniformity and linear energy transfer (LET) throughout the objects in order to find the most effective irradiation method depending on the objective of irradiation and the distribution of irradiation parameters in the objects.

To determine the optimal dose range limits we applied current physical methods to investigate the biochemical and biophysical changes in biological objects after irradiation. Gas chromatography-mass spectrometry method was used to trace the change in the concentrations of volatile organic compounds in bio-objects since some of these concentrations are highly sensitive bio-markers of lipid, protein oxidation as well as bacterial activity in the irradiated objects. To estimate the change in the native structure of proteins we used high-performance liquid chromatography-mass spectrometry method with tandem mass spectrometric detection and spectrophotometric method for estimation of myoglobin derivative concentrations. Microbiological analysis of bio-objects was carried out to estimate the efficiency of suppressing pathogens by irradiation. The lack of cost-efficient express methods for detecting irradiated objects with a high-water content caused us to apply the kinetic fluorometric fingerprinting technique for recognition of irradiated and non-irradiated bio-objects. The experiments involved pre-planting irradiation of seeds and root crops to assess the effect of irradiation on the growth and phytosanitary status of agricultural crops. The plants grown from irradiated seeds and root crops were planted and monitored at the experimental sites of the Siberian Federal Scientific Center of Agricultural-Biotechnology of the Russian Academy of Sciences to determine the optimal dose range limits for crops irradiation.

Following the experiments performed by our team it was established that the efficiency of irradiation of biological objects is determined as a function  $\boxtimes(D) = F(K1(D), K2(D), K3(D))$ , where K1 is a value determined by the dose uniformity throughout the object and the dose needed to suppress pathogens to the required degree; K2 determines the fraction of pathogens which are suppressed in the biological object irradiated with a certain dose; K3 is radiosensitivity heterogeneity of pathogens across the statistical ensemble.

We have found clear dose and time dependencies of the concentration of lipid and protein oxidation aldehydes as well as the concentration of ethanol alcohol. With a higher irradiation dose ranging from 250 Gy to 10000 Gy a higher peak of lipid and protein oxidation derivatives is detected on day 1 and day 2 after irradiation. On the contrary, the higher the dose the lower ethanol content in biological objects during 4 days of storage after irradiation. Therefore, the concentrations of lipid and protein oxidation aldehydes can serve as markers of lipid and protein peroxidation, while the concentration of ethanol is a marker of efficiency of bacterial suppression as a result of irradiation. For example, when beef tenderloin is irradiated with 1 MeV electron beam with the dose rate of 4 Gy·sec-1, lipid and protein peroxidation is observed in the beef samples irradiated with the doses of 500–1000 Gy and higher, while in the beef samples irradiated with 250–350 Gy the ethanol concentration is 2 times lower than in the non-irradiated beef samples. Therefore, as dose and time dependencies of volatile organic compound markers suggest, the lowest limit of the optimal dose range is 250–350 Gy, and the highest limit is 500–1000 Gy.

The spectrophotometric method for calculating the metmyoglobin concentration in bio-objects containing myoglobin as well as the trypsin hydrolysis of the native structure of bovine serum albumin allow us to quan-

tify the impact of electron beam and X-ray irradiation with different physical parameters, such as dose, dose rate, and the type of irradiation, on protein native structure.

Counting of viable cells in bio-objects after irradiation and assessment of the quantitative damage of protein native structure by irradiation show that the limits of the optimal dose range for beef tenderloin irradiated with 1 MeV electron beam with the dose rate of 4 Gy·sec-1 is 220-854 Gy assuming that the upper limit is determined by the myoglobin oxidation. At the same time, the optimal radiation dose range for beef is 204-755 Gy assuming that the upper limit is determined by the damage of protein native structure. Therefore, the dose ranges determined by measuring the number of viable cells in bio-objects and the damage of protein native structure align well with the ranges obtained by measuring the volatile compound concentrations.

Considering that the rate of indicator reactions involving carbocyanine dyes and oxidizing agents varies depending on physical and chemical properties of bio-objects, kinetic fluorometric fingerprinting technique, which measures the absorption spectrum and fluorescence intensity of extracts made from bio-objects, has proved to be the most suitable express method for recognition of non-irradiated and irradiated animal and plant biological tissues.

Our experimental studies allow to develop practical recommendations on how to improve the efficiency of the bio-object irradiation.

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## Section

Applications of nuclear methods in science, technology, medicine and radioecology

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