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# The effectiveness of the microbiological radiation decontamination process of agricultural products with the use of low energy electron beam

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## ABSTRACT

The effectiveness of the radiation decontamination process was tested for electron beam of energy 200 keV and 300 keV. The energy of electrons was controlled by the measurements of its penetration ability in stack of B3 dosimetric film. In the presented work, the reduction of total aerobic bacteria count was observed, depending on time of irradiation for samples of dried black pepper, onion flakes and bay leaves. The results were compared with the effect observed for the process where high energy electron beam was used.

## 1. Introduction

Spices are often contaminated with high levels of bacteria, moulds and yeasts originating from the plant environment, namely soil, water and air. Also pathogenic microorganisms may grow on some herbs and spice plants. As reported by Banach et al. (2016), pathogenic microorganisms, particularly *Salmonella* spp. and pathogenic *Bacillus* spp., were identified as a potential concern in black pepper and dried herbs. If untreated, the herbs and spices will result in rapid spoilage of the products, or when contaminated with pathogenic bacteria can also cause serious food-borne illnesses. In order to reduce the microbiological load, spices are usually steam treated or irradiated (Chmielewski and Migdał, 2005). Although the commercial application of ionising radiation for food processing began in the 1950's, it is still developing today (Ehlermann, 2016). Usually it is gamma sources, or high energy electron beam accelerators, that are used to eliminate microorganisms from food (Chmielewski and Haji-Saeid, 2004). Typically used e-beam accelerators for food treatment work in the range of electrons energy 8–10 MeV, which enables to irradiate the layer of thickness from a few to a dozen centimetres, depending on the properties of irradiated material.

A new approach to radiation processing of food and agricultural products is associated with a limited penetration ability of electrons having energy below 300 keV, defined as “low energy electrons”. In the process where high energy electrons are used, the whole volume of food is irradiated. Since microorganisms reside mostly on the surface of dry food, the irradiation of the external layer should be sufficient to eliminate food-borne microorganisms. The effectiveness of low energy electron beam (100 and 80 keV) in comparison with high energy

electron beam and gamma rays was proved in the elimination of *Bacillus pumilus* spores on test filters (Tallentire et al., 2010). The application of 170 keV electron beam effectively improved the quality of soybeans for further processing (Kikuchi et al., 2003; Todoriki et al., 2002). The low energy electron beam was also proposed as a method of microbiological decontamination of seeds. It was proved that it didn't negatively influence germination of seeds but the effectiveness of elimination of bacteria was lower in comparison with gamma rays (Fan et al., 2017).

The aim of the work presented was to estimate the effectiveness of the microbiological decontamination process of spices using electron beam of energy below 300 keV.

## 2. Experimental

### 2.1. Materials

Three types of spices were selected for the experiments: dried black pepper grains, dried onion flakes and dried bay leaves. The samples were analysed using the Digital USB Microscope AG118 to characterise its structure. All samples used in the experiments were naturally contaminated with microorganisms.

### 2.2. Irradiation

#### 2.2.1. Irradiation with high energy electron beam

Irradiation with electrons having energy 10 MeV was performed using the Elektronika accelerator (Migdał et al., 1995). 10 g of each sample was packed in plastic bags and irradiated in single layer to avoid

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dose gradient within irradiated sample. The defined doses were controlled using graphite calorimeters from RISO High Dose Reference Laboratory, which measure average dose absorbed by calorimetric body.

### 2.2.2. Irradiation with low energy electron beam

For irradiation with low energy electron beam the Accelerator ILU-6 was used (Zimek et al., 1995). This machine, installed in 1988 at INCT for the purposes of research and technology applications using the electron beam within the range of energy 0.5–2 MeV, had to be examined for the possibility of changing operating parameters in order to lower the energy of accelerated electrons while ensuring the safety and stable operation with regard to design requirements and constraints. Analysis of the accelerator equipment work conditions have shown that reducing the electron energy level to 0.15–0.3 MeV requires an alternative High Voltage modulator system. The new constructed device, based on transistor switches, ensured flexibility of the generated pulse parameters selection.

The considerations of the irradiation system configuration regarding the specific properties of low energy electron beam (short penetration range), ILU-6 electron window geometry, and the form of the samples to be irradiated, led to the design of the rotation drum made of thin stainless steel grid (Fig. 1.). The drum of diameter 7 cm and the height 8.5 cm was rotating with the constant speed 150 rpm. The constructed drum enables rotation of the samples under the beam during irradiation to ensure delivery of a uniform dose on the surface of the samples.

In the experiments, 10 g of each food sample was placed in the drum separately and irradiated for 2–30 min.

The dose measurements for irradiation with low energy electron beam were performed using B3 dosimetric foil from Riso High Dose Reference Laboratory, Denmark. This dosimeter is a 18  $\mu\text{m}$  thick film and can be used for measuring doses at low-energy electron irradiation using a calibration function that has been established using high energy electron irradiation (Helt-Hansen et al., 2005). The Risoscan software was used to read the dose. The penetration ability of the electron beam was measured using the stack of B3. However, ILU-6 has the built-in instrument for indicating the accelerating voltage, the real energy of electron beam that the material is exposed to, which is lower due to absorption in 50  $\mu\text{m}$  Ti foil of the accelerator extraction window, and about 10 cm air gap between the window and the bottom of the drum. The calculations taking into account the stopping power of Ti and air have shown that total energy degradation rate is 80–50 keV, and 150–300 keV in the initial range, respectively. The visualisation of the irradiated layer of thickness was observed in a cross-section photo of the PVC film, where the density was 1.4  $\text{mg}/\text{cm}^3$ , as shown in Fig. 2.

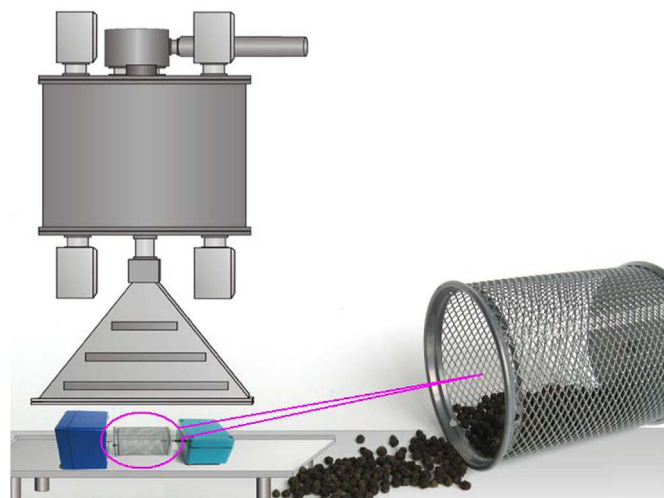


Fig. 1. Diagram of irradiation of loose materials in rotating drum using ILU-6 accelerator.

### 2.3. Microbiological analysis

The analysis were performed for control (not irradiated) samples, and for samples irradiated with different doses. After irradiation, 10 g of each sample was placed in sterile plastic bags with 90 ml of Ringer solution and homogenised in stomacher. The obtained suspension was used to prepare a series of sequential dilutions used to reduce a dense culture of cells to a more usable concentration. The growth of bacteria was observed after 72 h of incubation at 30 °C using Casein-Peptone Dextrose Yeast Agar. The values of total count of aerobic bacteria were transformed into log<sub>10</sub> values and presented as average from two replicates  $\pm$  standard deviation.

## 3. Results

### 3.1. Properties of food samples

The food samples selected for the experiments differed in structure, which is an important parameter when low penetrating radiation is used. Black pepper grains have a heterogeneous structure, which can be observed in the cross section of the grain as shown in Fig. 3. The thickness of the external layer of black pepper is from 200 to 500  $\mu\text{m}$ . The density of food samples was determined by the use of Archimedes' principle. The measurements were performed in ethanol. The average density of external and internal layer of black pepper was 0.9 and 1.5  $\text{g}/\text{cm}^3$  respectively. The samples of onion flakes and bay leaves are homogenous but have different average densities 1.2 and 0.7  $\text{g}/\text{cm}^3$  respectively. Onion flakes have higher density than bay leaves, which are highly porous, as shown in Fig. 4.

### 3.2. The penetration ability of the low electron beam

The penetration ability of the low energy electron beam was measured using the stack of B3 film. The depth-dose profiles for electron beam at two energy levels are presented in Fig. 5.

The penetration ability of the electron beam depends on the thickness of titanium foil and the distance between the accelerator window and the surface of the irradiated sample. In the experimental conditions the electrons having energy 200 keV could penetrate about 8  $\text{mg}/\text{cm}^2$  of irradiated samples whereas the beam of energy 300 keV could penetrate about 32  $\text{mg}/\text{cm}^2$ .

### 3.3. Microbiological decontamination

#### 3.3.1. The effectiveness of the high energy electron beam

The effectiveness of the microbiological decontamination process was tested on selected food samples irradiated with the electron beam of energy 10 MeV. The results of the microbiological analysis are presented in Table 1.

As shown in Table 1, the reduction of the microbiological load depends on the dose of irradiation. In presented experiments, the highest reduction of the microbiological load was observed for samples of bay leaves. Applying a dose of 2.5 kGy resulted in a 3 log reduction in the total number of aerobic bacteria. More resistant to irradiation were samples of dried onion flakes. The lowest reduction of the microorganism population was observed for samples of black pepper. Applying a dose of 2.5 kGy caused only about a 1 log reduction in the number of aerobic bacteria.

#### 3.3.2. The effectiveness of the low energy electron beam

The effectiveness of the microbiological decontamination process was tested on selected food samples irradiated with the electron beam of energy 200 keV and 300 keV. The results of the microbiological analysis of onion flakes and bay leaves are presented in Table 2.

A higher irradiation time was used for treatment of black pepper. The results of the microbiological analysis of black pepper are

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