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## 1. Introduction

Glass plate photographic negatives are valuable artifacts of historical and cultural heritage, but they are highly susceptible to fungal contamination, which can cause irreversible damage. Ionizing radiation has been explored as a potential method for decontamination due to its biocidal effect, ensuring both microbiological safety and the preservation of the material. This study evaluates the efficacy of **gamma radiation (6 kGy)** in eliminating fungal contamination on glass negatives under **room temperature (RT)** and **dry ice conditions (DI)**. Additionally, it presents a new methodology for bleaching glass plate negatives that become darkened after decontamination by irradiation.

## 2. General experimental

### 2.1. Sample selection

Four glass plate photographic negatives measuring 9 x 6.5 cm and 2mm thick were selected for the study.



### 2.2. Irradiation process by gamma rays

The samples were exposed to gamma radiation using IPEN's Cobalt-60 Multipurpose Gamma Irradiation Facility. The applied dose was **6 kGy** (disinfection) at a rate of **1 kGy.h<sup>-1</sup>**. Two samples were processed at **room temperature (25°C)** and two samples processed with **dry ice (-73°C)**.

### 2.3. Microbial analysis

Microbiological assessments included colony-forming unit (CFU) counting, performed using contact plates with selective culture media. Samples were divided into two groups: (1) irradiated at room temperature (6 kGy, RT) and (2) irradiated with dry ice (6 kGy, DI). The effectiveness of decontamination was determined by comparing microbial growth pre- and post-treatment.

### 2.4. Bleaching treatment

For the bleaching treatment, the irradiated samples were exposed for 20 hours inside an expanded polystyrene box with a UVB lamp to restore transparency.

## 3. Results

### 3.1. Microbial Identification and Reduction

The glass plate photographic negative samples exhibited significant microbial growth, confirming active contamination before irradiation.

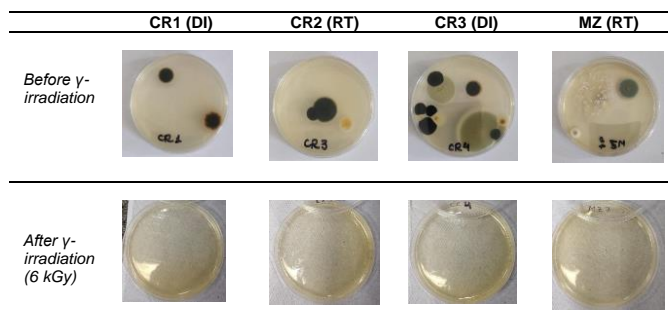
Dose (kGy)	Sample	Temperature (°C)	Pre-irradiation (CFU/m <sup>2</sup> )	Identification morphology	Post-irradiation (CFU/m <sup>2</sup> )
6	CR1	-73	2	1 <i>Cladosporium</i> spp. 1 NSF*	0
	CR2	25	3	2 <i>Cladosporium</i> spp. 1 NSF*	0
	CR3	-73	12	6 <i>Cladosporium</i> spp. 2 <i>Penicillium</i> spp. 4 NSF*	0
	MZ	25	4	1 <i>Penicillium</i> spp. 3 NSF*	0

\*non-sporulating fungi

## 4. Conclusions and Acknowledgements

- Ionizing radiation at **6 kGy** successfully eliminated microbial contamination from **glass plate photographic negatives**, with or without the use of dry ice. This study reinforces the potential of radiation processing in the **preservation of historical photographic materials**, preventing biodeterioration while maintaining the integrity of the artifacts. Future research should investigate the long-term stability of irradiated materials and optimize protocols for heritage conservation.
- This research is part of the doctoral studies at **Nuclear and Energy Research Institute (IPEN)** and was conducted with support from the **University of São Paulo (USP)**. Special thanks to the technical team involved in the irradiation experiments.

All samples irradiated at **6 kGy (RT and DI)** showed a **complete reduction** in fungal growth, demonstrating the effectiveness of gamma radiation for decontamination.

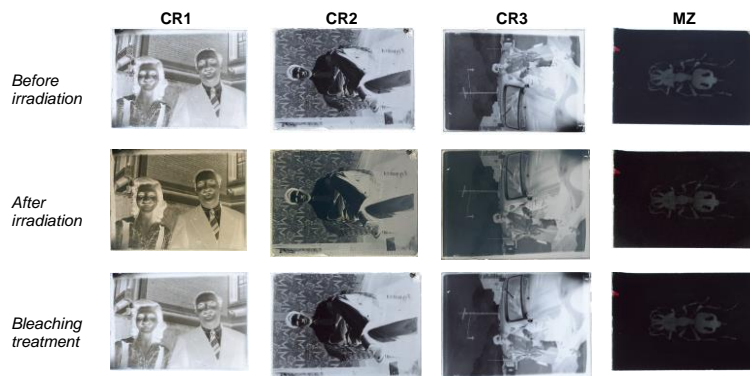


The use of **dry ice during irradiation** did not affect the fungal decontamination but may contribute to the mitigation of undesirable optical effects in vitreous materials.

### 3.2. Impact on Image Quality

Due to the ionizing radiation processing, it was expected that the glass plate negative samples would have a slightly darkened appearance of the glass of the photographic negatives.

After 20 hours of exposure to UVB radiation, the irradiated samples lightened and became close to their original state.



### 3.3. Protocol for ionizing radiation processing for fungal decontamination treatment of glass plate photographic negatives

- In the historical collection of glass plate photographic negatives, separate the materials contaminated by fungi.
- Store the glass plate negatives in sealed packages and inside boxes, preferably with dry ice.
- Process by gamma irradiation with a dose of 6 kGy, at a rate of 1 kGy.h<sup>-1</sup>.
- Wait 24 hours after processing and expose the glass plate negatives to UVB light, in a closed expanded polystyrene chamber, for 20 hours.