Gamma-irradiation for cultural heritage – treatment of selected fungi on linen textile

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BACKGROUND OF THE STUDY

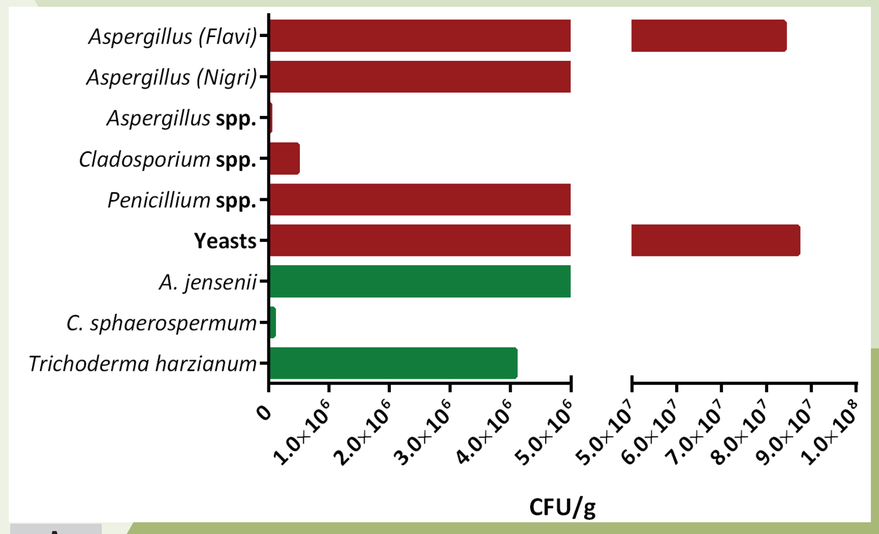
A common carrier for paintings is glue-coated linen that is vulnerable to fungal biodeterioration. Gamma-irradiation has been proposed as one of the physical methods for control of fungal contamination of cultural heritage objects but it could initiate unwanted side effects on organic polymers in textile when applied in high and repeated irradiation doses.
The purpose of the study was to assess antifungal effect of gamma-irradiation doses and dose rates against naturally occurring mycobiota and artificially inoculated primary, secondary and tertiary fungal colonizers common for cellulose materials like linen. The composition of natural mycobiota on glue-coated linen (initial level) and eventual post-irradiation recovery of mycobiota were analyzed.

METHODOLOGY

The initial level of common fungal colony-forming mycobiota on model glue-coated linen textile was determined by plate count method on Malt Extract Agar (MEA) upon 7 days of incubation (at 25°C and 70-80% relative humidity -Rh) and the data expressed as the number of colony-forming units per gram (CFU/g). Next, linen samples were separately inoculated with selected primary (*Aspergillus jensenii*), secondary (*Cladosporium spaherospermum*) and tertiary colonizers (*Trichoderma harzianum*) at concentration of 104 CFU/g. Inoculated linen and controls were incubated at 25 C and 70-80% of Rh for 7 days. One group of samples was analysed immediately upon the incubation while the rest of the samples were irradiated at ^{60}Co gamma source at RCDL to doses of 2, 7, 20 and 50 kGy, at dose rates of 0.1 and 9.8 Gy/s and analysed after incubation for 0, 7, 14 and 28 days.

RESULTS

Alternaria spp., *Aspergillus* spp., *Cladosporium* spp. *Fusarium* spp., *Penicillium* spp. and yeasts comprised naturally occurring mycobiota, in initial concentrations of 10^3 CFU/g (molds) and 10^4 CFU/g (yeasts). These fungi were non-homogeneously dispersed on glue-coated linen. Upon 7 days of incubation in humid atmosphere the concentration of mycobiota increased for four orders of magnitude. Similar increase was obtained for non-irradiated artificially inoculated primary, secondary and tertiary colonisers (Fig.1A).
All applied doses and dose rates were effective against primary and tertiary colonizers but not for secondary colonizers and linen mycobiota. Doses of 2 and 7 kGy was ineffective in reduction of linen mycobiota to the initial level; after 28 days of incubation fungi were recovered in concentrations up to 10^6 and 10^5 CFU/g, respectivelly (Table 1-2). Dose of 20 kGy (0.1 Gy/s) reduced *Cladosporium* spp., and *Alternaria* spp. to 10^4 CFU/g; *Penicillium* spp. was reduced to the initial level while yeasts, *Aspergillus* spp., and *Fusarium* spp. recovered in concentrations below initial (Table 2). For both 7 and 20 kGy dose rate of 9.8 Gy/s was more effective in fungal elimination than 0.1. Gy/s while for 2 kGy the dose rate effect was inconsistent. Upon exposure to 50 kGy sterile white mycelia was recovered on few plates after incubation periods.
All applied doses and dose rates were effective against artificially inoculated primary (A. *jensenii*) and tertiary colonizers (T. *harzianum*). Secondary colonizer, C. *sphaerospermum* survived radiation with 2, 7 and 20 kGy (Fig.2), and showed the similar recovery pattern as obtained for *Cladosporium* spp. After treatment with 7 and 20 kGy (0.1 Gy/s) *Cladosporia* recovered between 7th (or 14th) and 28th day in concentrations between 10^3 and 10^6 CFU/g. The same doses with dose rate 9.8 Gy/s inhibited recovery of C. *sphaerospermum*.



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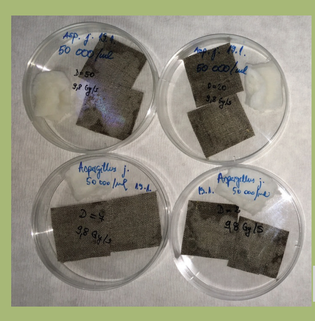

B

**Figure 1. A) Mean concentration of naturally occurring (red) and artificially inoculated (green) mycobiota after 7 days of incubation at 25 C and 70-80% of Rh;
B) Linen textile samples inoculated with A. *jensenii* , prior to gamma-irradiation.**

Table 1. Levels of naturally occurring mycobiota after irradiation at 2 kGy and dose rates of 0.1 and 9.8 Gy/s.

Fungi	Mean fungal concentration (CFU/g)							
	0th day		7th day		14th day		28th day	
	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s
Alternaria spp.	$4.5x10^3$	10^3	-	-	$2x10^5$	$8x10^4$	$6x10^3$	$6.6x10^3$
Aspergillus (Flavi)	-	-	-	-	$3.4x10^4$	-	$1.8x10^4$	$6.4x10^6$
A. fumigatus	-	-	-	-	$2x10^4$	-	-	-
Aspergillus (Nigri)	-	-	-	-	10^4	-	-	-
Aspergillus spp.	-	-	-	-	-	-	10^5	-
Cladosporium spp.	$7.6x10^3$	$2x10^3$	$2x10^5$	-	2.510^5	$8.3x10^5$	$9x10^5$	$1.3x10^6$
Epicoccum spp.	-	-	-	-	$1.8x10^3$	-	-	-
Fusarium spp.	-	-	-	-	$1.5x10^5$	$1.7x10^3$	-	$4x10^4$
Mucor spp.	-	-	-	-	-	-	$2x10^3$	-
Penicillium spp.	-	-	-	-	-	$4.8x10^5$	$5x10^4$	-
Phoma spp.	-	$6x10^3$	-	-	-	-	-	-
Stemphylium spp.	-	-	-	-	-	$2x10^4$	-	-
Yeasts	$2.3x10^3$	$1.7x10^4$	$2x10^4$	-	-	-	-	$2.3x10^4$
Other fungi	10^3	$2x10^3$	-	-	$5.4x10^3$	-	-	-

Table 2. Mean fungal concentration of naturally occurring mycobiota (CFU/g) after irradiation at 7 kGy and 20 kGy , both applied at dose rates of 0.1 and 9.8 Gy/s.

Fungi	CFU/g at 7 kGy								CFU/g at 20 kGy	
	0th day		7th day		14th day		28th day		28th day	
	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s	0.1 Gy/s	9.8 Gy/s
Alternaria spp.	-	-	-	-	$3.9x\ 10^4$	-	$2x10^5$	-	10^4	-
Cladosporium spp.	-	-	$8.1x10^3$	-	$9x10^5$	-	$1.6x10^6$	-	$1.5x10^4$	-
Fusarium spp.	-	-	-	-	-	-	$4.5x10^5$	-	-	625
Penicillium spp.	-	-	-	-	-	-	-	10^4		
Yeasts	$1.4x10^4$	$1.5x10^4$	700	-	-	$6x10^4$	-	$2.3x10^4$	-	150
Other fungi	10^3	-	100	-	$2x10^5$	$1.5x10^2$	-	-	-	100

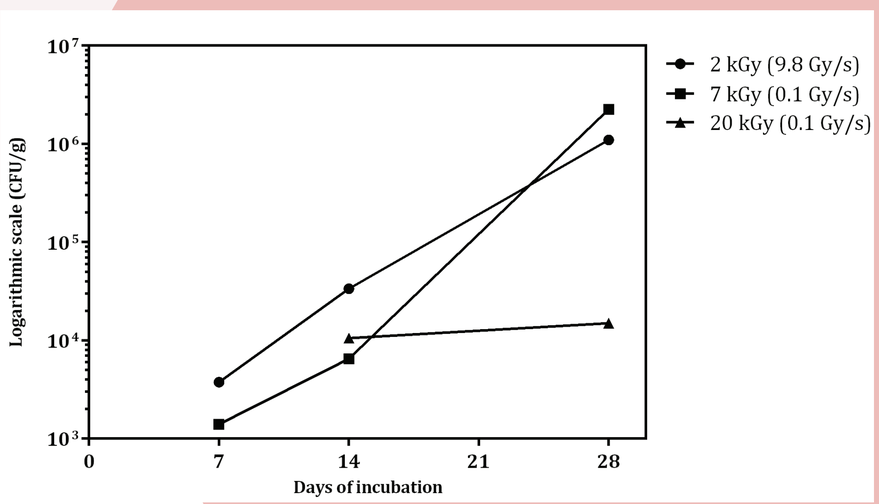

Figure 2. Gamma-irradiation treatment survival of secondary coloniser *Cladosporium sphaerspermum* grown on glue-coated linen

Aspergillus flavus (Flavi)

Cladosporium spp.

CONCLUSION

Results indicated that species of *Cladosporium* and yeasts seem to be the most resistant fungi to gamma irradiation. For successful gamma-radiation reduction of fungal contamination on cultural heritage objects it is essential to determine mycobiota composition and to asses optimal irradiation conditions appropriate for reduction of the contamination to an acceptable level.

ACKNOWLEDGEMENTS
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