International Biodeterioration & Biodegradation 90 (2014) 152–160

Contents lists available at ScienceDirect

International Biodeterioration & Biodegradation

journal homepage: www.elsevier.com/locate/ibiod

Fungal biodeterioration of stained-glass windows

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ARTICLE INFO

Article history:
Received 25 November 2013
Received in revised form
26 February 2014
Accepted 3 March 2014
Available online 25 March 2014

Keywords:
Stained glass windows
Biodeterioration
Fungi
Heritage

ABSTRACT

Biodeterioration of stained-glass windows by fungi was studied using historically accurate glass production methods. Glass reproductions were made according to the chemical composition determined by micro energy dispersive X-Ray Fluorescence of two historical glass windows belonging to King Ferdinand II's collection dating from the 15th and 17th centuries. Three distinct glasses compositions with different colours were selected and reproduced: i) a mixed-alkali colourless glass: ii) a purple potash-glass with manganese as chromophore, and iii) a brown potash-glass coloured by iron ions. The reproduced glass samples, with two initial surface morphologies (corroded and non-corroded), were inoculated with fungi previously isolated and identified on the original stained-glass windows as species of the genera *Penicillium* and *Cladosporium*. Physical and chemical glass surface alterations were analysed by means of optical microscopy, Raman microscopy, micro Infrared spectroscopy, and scanning electron microscopy with energy dispersive spectroscopy analysis. Results showed that fungi produced clear damage on all glass surfaces, present as spots and stains, fingerprints, biopiting, leaching and deposition of elements, and formation of biogenic crystals. Therefore, the inoculated fungi were able to biodeteriorate glasses with distinct compositions. Regarding the biodeterioration degree, there were no differences between the initial non-corroded and corroded glass surfaces.

1. Introduction

Conservation and restoration of historical stained-glass windows is recognized as a complex problem (Drewello and Weissmann, 1997), since not only physical and chemical attack can occur, but also microbial deterioration, known as biodeterioration. Generally, the biological corrosion of historical glass has been underestimated (Garcia-Vallès et al., 2003). There are several studies on chemical corrosion of stained-glass windows, located in cathedrals and churches all over Europe (e.g. Leissner, 1996; Orlando et al., 1996; Garcia-Vallès et al., 1996, 2003; Sterpenich and Libourel, 2001; Melcher and Schreiner, 2004; Farges et al., 2007; Tournie et al., 2008; Gentaz et al., 2011; Vilarigues et al., 2011). However, the biodeterioration of those windows has often been undervalued, and little is known about the damage produced by microorganisms (Rölleke et al., 1999; Schabereiter-Gurtner et al., 2001; Carmona et al., 2006; Piñar et al., 2013). A few works stated that the biodeteriorative role of microorganisms enhances deterioration and accelerates the physical–chemical processes leading to glass decay (Drewello and Weissmann, 1997; Gorbushina and Palinska, 1999; Marvasi et al., 2009). Fungi are among the most harmful microorganisms associated with biodeterioration of organic and inorganic materials, including glass (Drewello and Weissmann, 1997). The resistance of fungal spores to desiccation, their adhesion ability to the hydrophobic substrata, as well as the

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http://dx.doi.org/10.1016/j.ibiod.2014.03.007
0964-8305/

ability to metabolize a wide range of carbon sources and the preferred acidic to neutral pH values can be important factors to the development of fungi on glass surfaces. Moreover, fungal metabolic versatility enhances their efficiency to colonize a wide range of substrata, such as stained-glass windows that were exposed to high relative humidity and temperature fluctuations, water, and other environmental factors such as aerosols and dust that carry out inorganic and organic matter (e.g. microorganism spores). Thereupon, the main conditions that promote fungal growth on stained glass windows are: i) the presence of organic matter of various origin, such as dust deposits, dead fungal and bacterial materials, and microbial metabolites; and ii) high temperature and relative humidity values that allow glass to hold adsorbed water. The amount of adsorbed water depends on the type of glass and on the relative humidity. According to Walters and Adams (1975), soda-lime glass adsorbs more water than borosilicate glass or lead silicate glass. The water activity (a_w) on the surface increases in parallel to the formation of the gel layer and the incorporation of water molecules. This higher supply of water makes the glass more attractive for microbes (Drewello et al., 2000).

Fungal biodeterioration studies on historical stained glass are important in understanding its processes and to help find a way to avoid and/or control this type of deterioration. Therefore, the main objective of this work was to appraise the biodeterioration of stained-glass reproductions by fungi under laboratory conditions. In order to achieve this goal:

- two historical stained-glass windows, belonging to King Ferdinand II's collection, with known chemical composition were selected;
- the fungi that colonized the panels under study were identified and the more common genera were selected to be used in a glass biodeterioration experiment;
- glass reproductions with three distinct compositions and with non-corroded and corroded surfaces (simulating recently produced and weathered glass surfaces) were manufactured and were then inoculated with the selected fungi;
- different analytical techniques were used to study biodeterioration of the inoculated glass surfaces.

2. Materials and methods

2.1. Stained glass windows, storage conditions and its environment

King Ferdinand II (1816–1885) of Portugal gathered a vast collection of art works, including a collection of stained-glass windows. This collection was divided between his two principal residences: the Pena National Palace (Sintra, Portugal) and the Necessidades Palace (Lisbon, Portugal). In the Necessidades Palace, the king had the majority of his collection of stained-glass panels mounted in five windows and in three transom windows. He had a further assemblage installed in the great hall of Pena National Palace. In 1910, the stained-glass panels from the Necessidades Palace were removed from their original position and sent to the Ajuda National Palace (Lisbon, Portugal), where they remained in storage for several decades. In 1948, the complete collection arrived at Pena National Palace with the aim of being installed in the windows of the Stag Room. This plan was never implemented, and the windows were kept in a storage room in a poor state of preservation at the Pena National Palace for the next six decades (Rodrigues et al., 2013). Two panels belonging to this collection were selected for this study. One of the panels is from the 15th century and is thought to be of German origin (GP – German Panel). The other is a Swiss panel from the 17th century (SP – Swiss Panel) (Martinho and Vilarigues, 2011).

Pena National Palace, located in Sintra (38_47°N, 9_25°W), is influenced by oceanic conditions (it is only 8–9 km from the sea), and surrounded by a vast forested area. Temperature and relative humidity were measured inside the storage room at the Pena National Palace, using a humidity/temperature data logger (Rotronic HW3), from January to August 2011. Determinations of these two parameters are important in order to reproduce, under laboratory, the approximate environmental conditions to which the glass windows were kept in the Palace.

2.2. Manufacture of glass reproductions

Micro energy dispersive X-ray Fluorescence (μ-EDXRF) analyses were performed on the two stained-glass panels (Rodrigues et al., 2013). Glass reproductions were made based on the chemical compositions determined for the German and Swiss historical stained-glass panels (Rodrigues et al., 2013). From these data, three distinct compositions, with three different colours, were selected to be reproduced and used in the fungal biodeterioration experiment. Table 1 presents the chemical composition of the selected glasses. From this table it can be noticed that the colourless glass is a mixed-alkali while purple and brown glasses are potash-glass. The amount of Mn oxide in the purple glass is higher when compared to the other two glasses, since this is the colouring element of this glass. The colouring elements in brown glass are manganese (Mn) and iron (Fe).

For the glass synthesis, pure raw laboratory materials were used, and each glass was melted in a refractory ceramic crucible at 1300 °C, over 24 h. The glass obtained was blown using traditional off-hand glass tools and techniques analogous to the production period of the original samples. The resulting roundels (discs) kept a fire polished surface nearly identical to the historic glass production.

The discs were annealed at 500 °C (for 4 h), which was followed by slow cooling (approx. 20 h), and then they were cut into pieces with no further polishing. The pieces had the approximate dimensions of (10 × 10 × 2) mm^3.

In order to understand glass biodeterioration and the influence of surface texture, a set of 18 glasses for each composition (9 with non-corroded surfaces and 9 with corroded surfaces) were produced. The glass corrosion was performed by immersion of the samples in 40 mL of distilled water for periods of 4–6 months. The reaction containers were made of inert material (polyethylene) in order to avoid any reactions between the electrolyte and container walls, and sealed to prevent interactions with the atmosphere.

After glass reproductions were made, their chemical compositions were analysed using micro energy dispersive X-Ray Fluorescence (μ-EDXRF).

2.3. Biological sampling

Samples for microbial identification were collected in four distinct areas in each glass window. The panel surfaces were swabbed using sterile tubes swabs (one for each sample) that were

Table 1
Chemical composition used to reproduce the three distinct glasses with different colours.

Oxide components (w/w)	Swiss panel (SP)	German glass panel (GP)	
	Colourless	Purple	Brown
SiO_2	58.5	64.4	62.5
CaO	18.0	16.8	17.0
K_2O	5.0	12.6	11.2
MnO	0.7	1.2	0.8
Fe_2O_3	0.8	0.3	0.5
Na_2O	17.0	4.7	8.0