

# Supplementary Materials: Application and Monitoring of Oxidative Alginate–Biocide Hydrogels for Two Case Studies in “The Sassi and the Park of the Rupestrian Churches of Matera”

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## Characterization of Biocidal Hydrogels

Alginate-based hydrogels were characterized through viscometry, FTIR-ATR spectroscopy and differential scanning calorimetry (DSC). All measurements were performed in triplicate on pure alginate hydrogel (HY), 5 wt.% of sodium alginate crosslinked with 0.3% CaCl<sub>2</sub> and on the biocidal hydrogels BH\_1 (hydrogel containing calcium hypochlorite) and BH\_2 (hydrogel containing sodium dichloroisocyanurate).

*Viscometry measurements.* The relative viscosities of alginate hydrogels were measured using a Viscolead rotational viscometer (mod. ADV “L”, Fungilab, Barcelona, Spain). Viscosity of the hydrogels was measured at 25 °C by setting the rotational speed at 5 rpm and the recorded values were reported in Table S1.

*FTIR-ATR spectra.* The IR spectrum were acquired using a FTIR Spectrum Two (Perkin-Elmer, Waltham, MA, USA) equipped with a UATR module at room temperature in a scan range between 4000 and 450 cm<sup>−1</sup>, with 4 accumulations and at a resolution of 4 cm<sup>−1</sup>. Infrared spectra of alginates allow to estimate the M/G values by means of the ratio between the maximum intensity of the absorption bands centered at 1030 and 1080 cm<sup>−1</sup> (A<sub>1030</sub>/A<sub>1080</sub>); these two bands are typical of the mannuronate (C–O–H bending) and guluronate (C–O–C stretching) subunit respectively [1,2]. The IR spectra were reported in Figure S1(A), the extrapolated M/G ratios for HY, BH\_1 and BH\_2 were listed in Table S1. All the M/G ratios resulted higher than that of the sodium alginate powder (1.7).

*Water content.* The water content of the three hydrogels were determined by weighing according to:

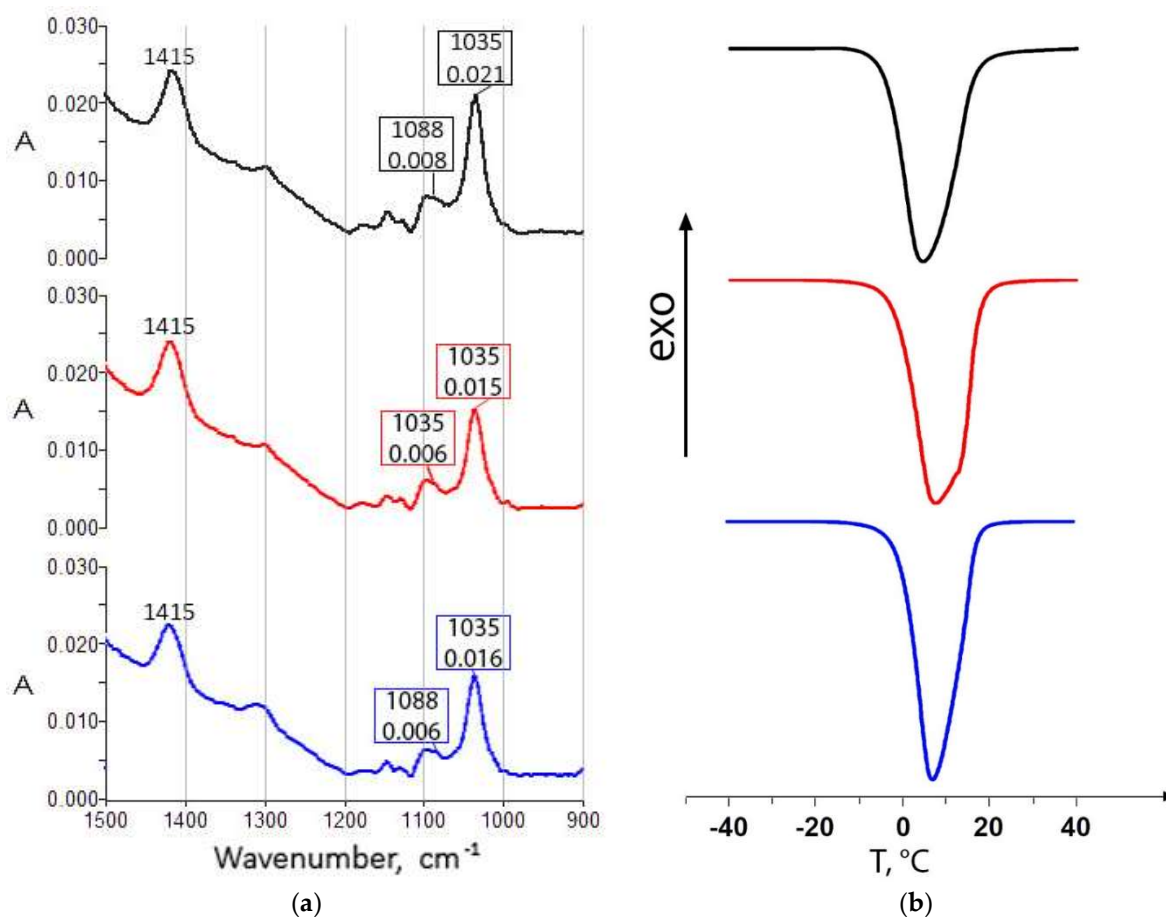
$$WC = \frac{W_w}{W_{HG}} \quad (1)$$

Where W<sub>w</sub> is the mass of water added in the hydrogel and W<sub>HG</sub> represent the mass of the hydrogels prepared with dry constituents [3]. The resulting values of WC were reported below in Table S1.

*Thermal analysis.* The thermal analyses were performed using a DSC 3 (Mettler Toledo, Schwerzenbach, Switzerland) differential scanning calorimeter. The hydrogels were placed in aluminum pans and the thermograms were acquired between −40 °C and +40 °C with a heating rate of 10 °C/min under a nitrogen atmosphere with a flow rate of 50 mL/min. The free water index (FWI), i.e., the fraction of water that exhibits bulk behavior in the gel medium, can be calculated as [4]:

$$FWI = \frac{\Delta H_{exp}}{\Delta H_{melt} \cdot WC} \quad (2)$$

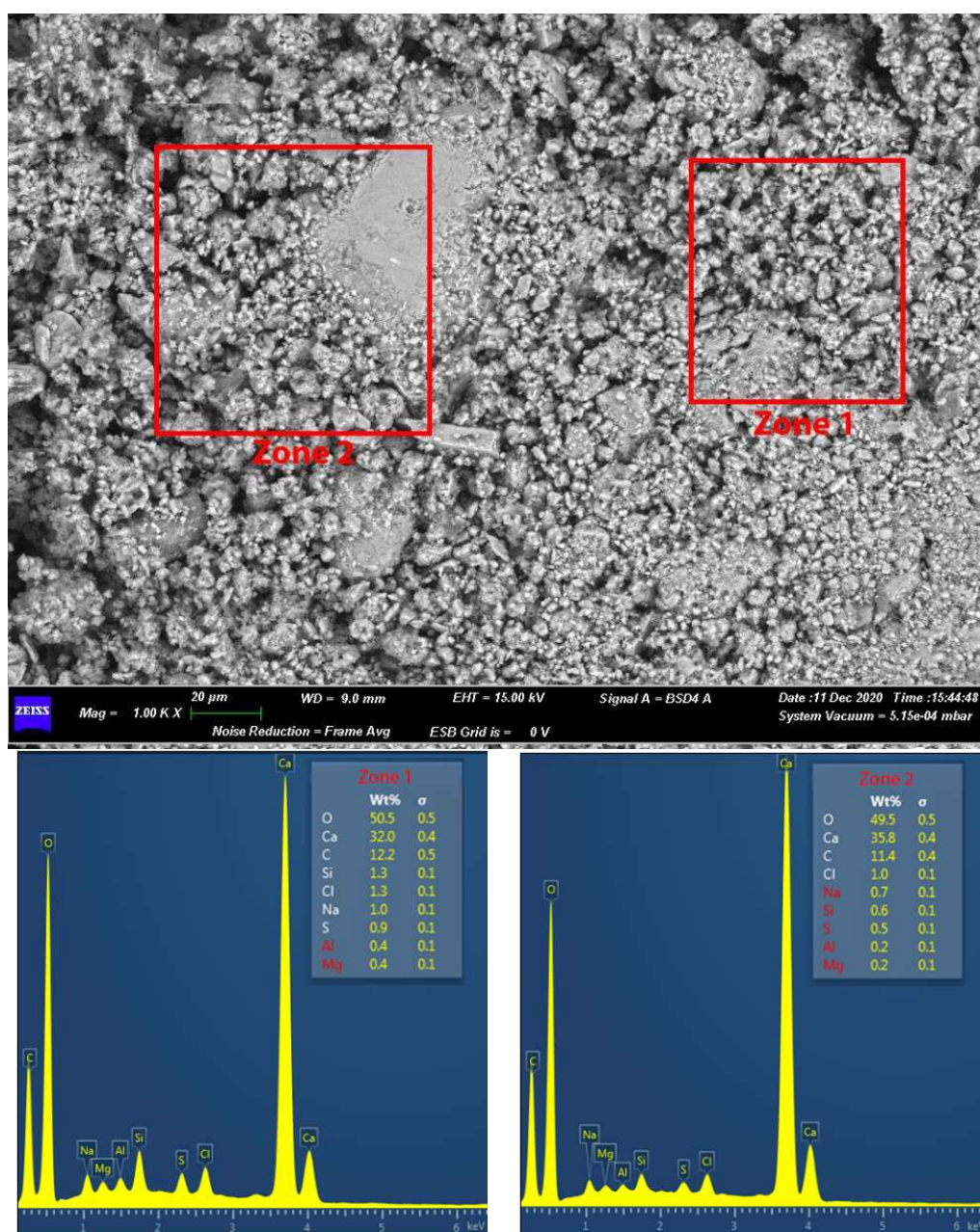
Where  $\Delta H_{\text{exp}}$  is the experimental enthalpy of the melting transition (J/g),  $\Delta H_{\text{melt}}$  is the theoretical melting enthalpy of water (333.61 J/g). FWI data were reported in Table S1 and thermograms were showed in Figure S1 (B).



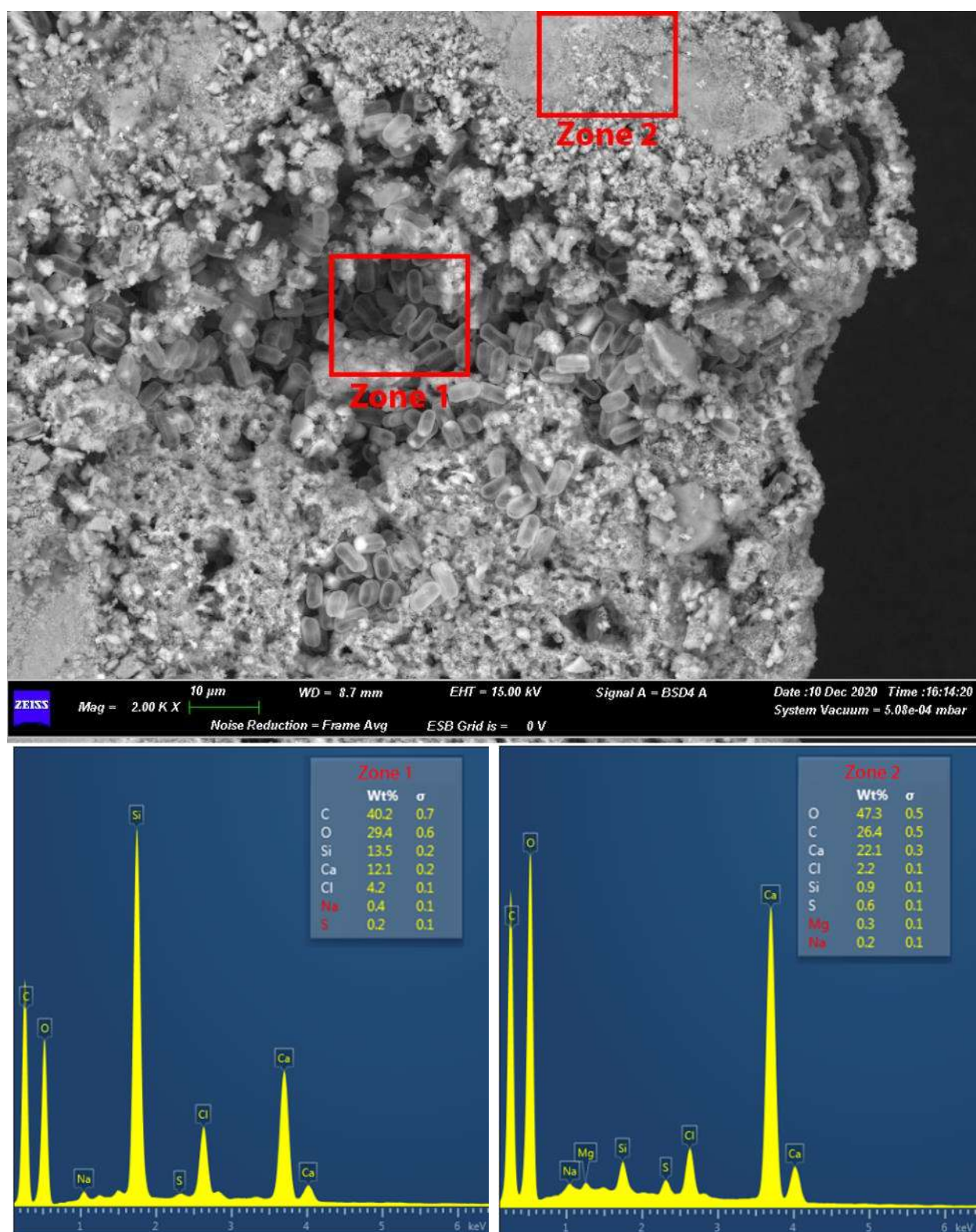
**Figure S1.** (A) FTIR-ATR spectra in the region comprise between 1500 and 900  $\text{cm}^{-1}$  and (B) DSC thermograms of 5 wt.% alginate hydrogel (—), BH\_1 (—) and BH\_2 (—).

**Table S1.** Summary of the investigated properties of pure alginate hydrogel (HY) and biocidal gels containing calcium hypochlorite (BH\_1) and sodium dichloroisocyanurate (BH\_2).

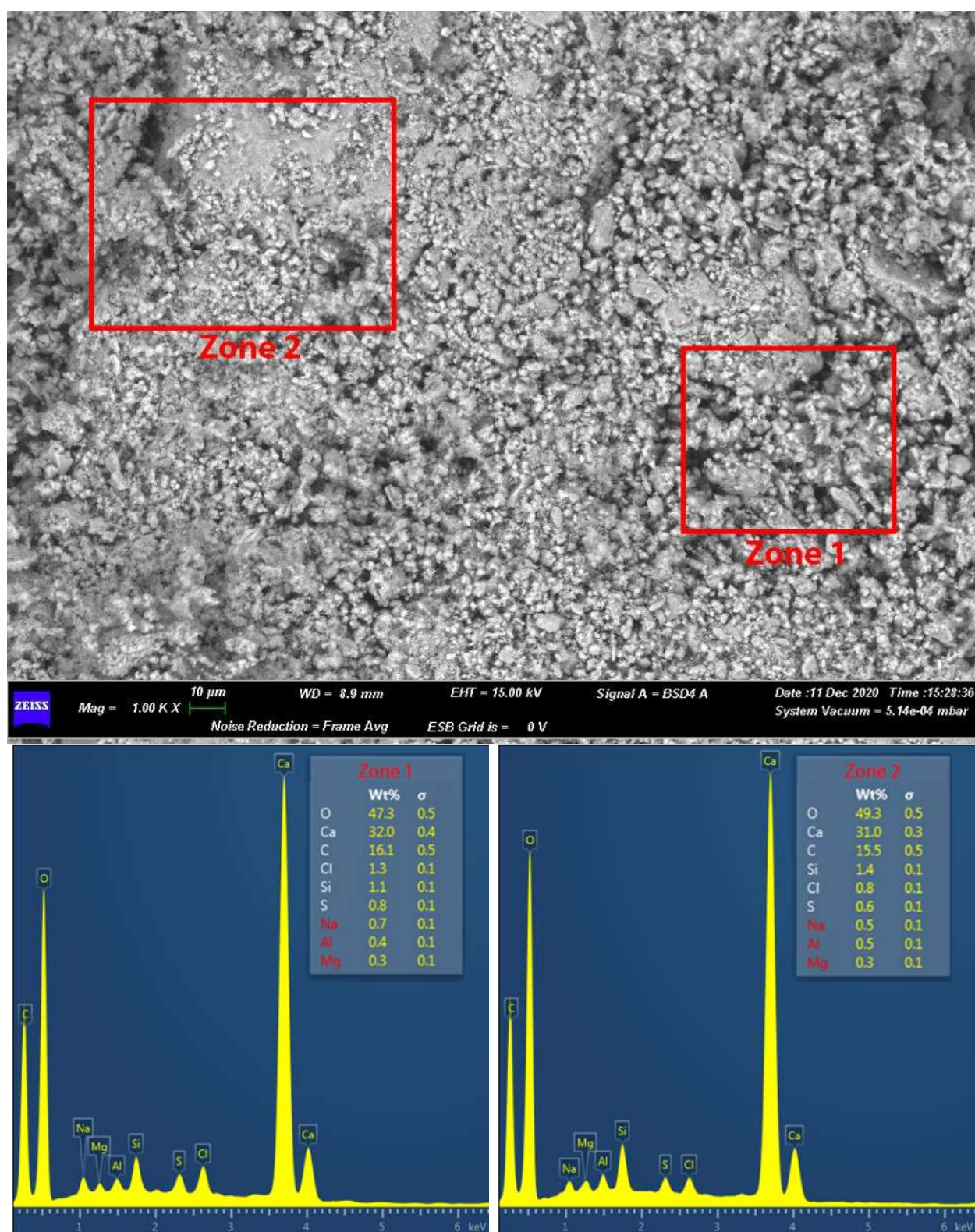
Sample	$\eta, P$	M/G	WC	FWI
HY	1036	2.6	0.95	0.86
BH_1	662	2.5	0.94	0.91
BH_2	916	2.7	0.94	0.92



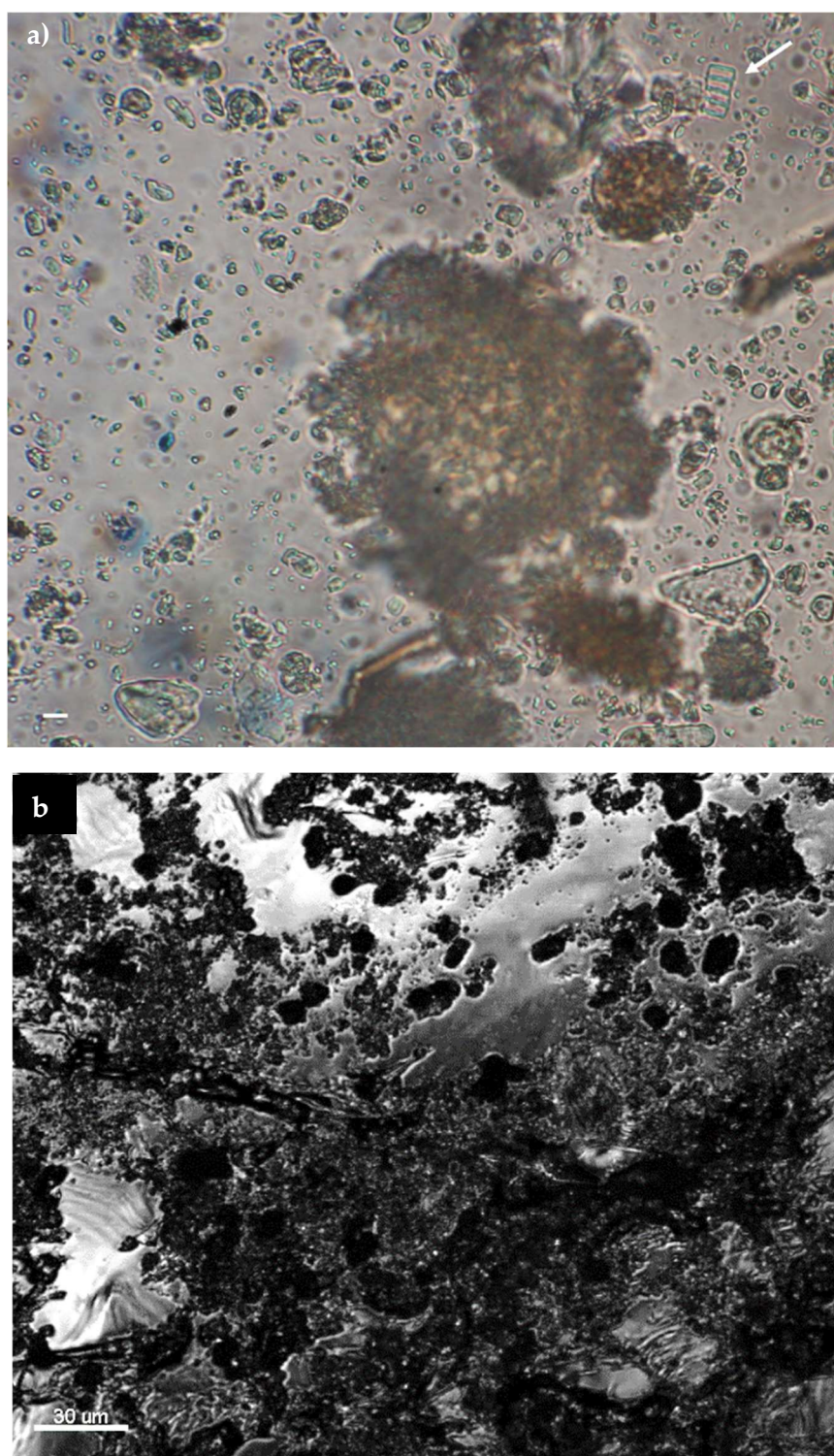
**Figure S2.** SEM images of reference stone materials fragments collected inside the Church of the “Madonna dei Derelitti” and EDS analysis of two selected areas.



**Figure S3.** SEM images of colonized stone materials fragments collected inside the Church of the “Madonna dei Derelitti” and EDS analysis of two selected areas.



**Figure S4.** SEM images of stone materials fragments collected after cleaning treatment inside the Church of the “Madonna dei Derelitti” and EDS analysis of two selected areas.



**Figure S5.** Images of samples collected at different areas of the Church of “Madonna dei Derelitti”, after the cleaning procedures obtained after observations at (a) the light microscope (bar = 5 µm, 40×) and (b) CLSM. It is evident the absence of living microorganisms; mineral material and cellular debris are visible in (a) among which empty frustules of diatoms (arrow).

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## References

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