Visual degradation in Leonardo da Vinci's iconic self-portrait: A nanoscale study

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The discoloration of ancient paper, due to the development of oxidized groups acting as chromophores in its chief component, cellulose, is responsible for severe visual degradation in ancient artifacts. By adopting a non-destructive approach based on the combination of optical reflectance measurements and time-dependent density functional theory *ab-initio* calculations, we describe and quantify the chromophores affecting Leonardo da Vinci's iconic self-portrait. Their relative concentrations are very similar to those measured in modern and ancient samples aged in humid environments. This analysis quantifies the present level of optical degradation of the Leonardo da Vinci's self-portrait which, compared with future measurements, will assess its degradation rate. This is a fundamental information in order to plan appropriate conservation strategies. © 2014 AIP Publishing LLC. [http://dx.doi.org/10.1063/1.4879838]

A typical sheet of paper is mainly composed of a network of cellulose fibres. In the early 16th century, when the Leonardo's self-portrait[1] was made (Fig. 1), papermakers sourced fibres from cotton, linen, or hemp rags.[2–4] Pristine cellulose is a linear homopolymer composed of β-D-gluco-pyranose units $(C_6H_{10}O_5)_n$, which are linked together by β-(1,4)-glycosidic bonds to form chains, with n ranging from $\sim 10^2$ to $\sim 10^4$. Cellulose chains have a strong tendency to aggregate into highly ordered structural entities through an extended network of hydrogen bonds which link up to form fibres whose diameters range from ~ 1 to $\sim 10\,\mu m$.[5] Isolated cellulose possesses the most stable thermodynamic conformation among carbohydrates, a characteristic which endows it with excellent durability over time. The ageing of paper-based cultural heritage is mainly due to the degradation of cellulose as a result of its exposure to external agents, such as water (i.e., humidity), ultraviolet (UV) and visible (Vis) radiations, pollutants, and microorganisms, often acting in a synergistic way.[6,7] The overall degradation process can therefore be seen as a combination of two interconnected chemical reactions, namely, acidic hydrolysis of β-(1,4)-glycosidic bonds, which shortens the cellulose chains and as a result weakens the mechanical properties of sheets, and oxidation, with the subsequent development of various chemical products.[6–10] Among such oxidation products are carbonyl groups of various kinds (ketones, aldehydes, and carboxylic acids); those responsible for yellowing are called chromophores. Pristine cellulose does not absorb light above about 200 nm (below 6 eV).[7] The white appearance of paper is due to its light scattering effect.[11,12] The yellowing seen in ancient papers is mainly due to the fact that chromophores in cellulose[13] absorb in the higher energy range of visible light (corresponding to violet and blue) and largely scatter the yellow and red portion, thereby producing the characteristic yellow-brown hue.

In this Letter, we apply an innovative and non-destructive method using an improved version of the Kubelka-Munk (K-M)[14] model for light propagation in turbid media[15,16] in combination with Time-Dependent Density Functional Theory (TDDFT).[13,17] In this way, we have been able to correlate the optical reflectance spectra of Leonardo's self-portrait with the
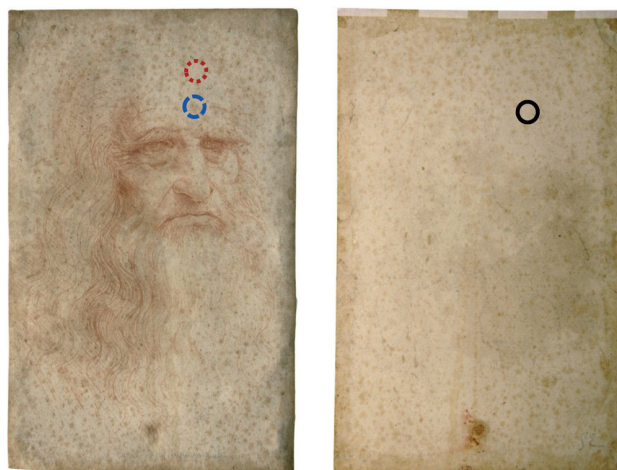

FIG. 1. Verso and recto of Leonardo da Vinci's self-portrait as acquired during the diagnostic studies carried out at the Central Institute for the Restoration of Archival and Library Heritage in Rome, Italy. The spots where analyses were carried out are shown: Pr1 (red short dashed circle), Pr2 (blue long dashed circle), and Pv (black solid circle).

absolute concentration of chromophores in the paper it was made on, and hence the extent of optical degradation.

Thanks to an extraordinary concession made by Turin's Royal Library in 2012, the self-portrait was made available for the purpose of carrying out diagnostic investigations at the Central Institute for the Restoration of Archival and Library Heritage in Rome, Italy. This privileged access to Leonardo's famous drawing enabled us to quantify its current level of optical degradation. The diffuse absolute reflectance (R) spectra of the artwork were measured in two 6 mm diameter spots (designated Pr1 and Pr2) on the recto (front) of the sheet, and in one spot (Pv) of the same size on its verso (back), as shown in Fig. 1. The positions of the spots were settled on because they are representative of a high degradation area that coincides with a foxing blemish (Pr2); an area affected by intermediate degradation (Pr1), and a low degradation area (Pv). The reflectance spectra were acquired over a wavelength (λ) range from 250 to 1050 nm (4.96–1.18 eV of photon energy). By applying the Kubelka-Munk (K-M)[14] model,[18,19] R_∞ (Fig. 2(a)), the reflectance that an infinitely thick paper sample would have, was ascertained[11,12] for full explanation). R_∞ spectra are representative of the intrinsic optical properties of the paper that Leonardo chose to draw his self-portrait on. In the three areas of the artwork that were studied, R_∞ is at its most intense in the near infrared (NIR) region of the spectrum, while it decreases in the Vis and UV regions. Overall, Pr1 and Pr2 show lower values for R_∞ with respect to Pv. In addition, Pr2 yields the lowest values, below 700 nm (above 1.77 eV), in particular. The behavior of the R_∞ spectra in the Vis region, with a range of approximately 400–700 nm (1.77–3.1 eV), is in agreement with the progressive darkening observed as one passes from Pv to Pr1, and then to Pr2.

From the reflectance spectra, we recovered the experimental intrinsic absorption coefficient of the constituent cellulose fibers of t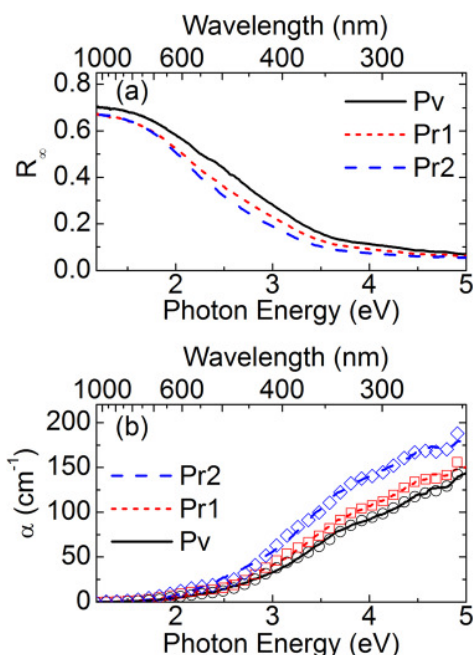he paper of Leonardo self-portrait. This was achieved by extending Yang and Miklavcic's[15,16] improved version of the K-M model (the method is described in detail in Refs. 11 and 12) to strongly absorbing samples. Fig. 2(b) reports the spectra of Pr1, Pr2, and Pv. Overall, the absorption coefficients reach their maximum values at the shorter wavelength end of the spectra, but subsequently decrease and show a first shoulder (more evident in Pr2) at about 264–270 nm (4.6–4.7 eV), and a second shoulder at about 335–344 nm (3.6–3.7 eV). At longer wavelengths, they decrease smoothly with a tail extending well into the visible region up to 620 nm (2 eV). Therefore, the progressive darkening seen when passing from Pv to Pr1, and then to Pr2, can be correlated to the higher values of α in the Vis region. Since pristine cellulose does not absorb above 200 nm (below 6 eV), the wide energy range of optical absorption that was observed in the Leonardo self-portrait indicates the presence of several chromophores with different absorption bands.

Here, it should be pointed out that the complexity of the local chemical environment in cellulose precludes the possibility of identifying individual chromophores in the optical spectrum by drawing analogies with reference compounds.[11–13] For this reason, in order to interpret the results (and following on from our previous work[11–13]), we modelled oxidized cellulose using eight different configurations of ketone, diketone, and aldehyde groups within the β-D-glucopyranose units (see Figs. 4–6 of Ref. 11). Some representative oxidized groups are shown in Fig. 3. The eight chromophores of our model are selected following Ref. 9. They are among the most abundant oxidized groups appearing in cellulose aged below 90 °C and, according to our TDDFT calculations, are those optically active below 4.8 eV. It should be emphasized that the aldehydic group is expected to be the transient product of the subsequent oxidation of the alcohols located on the C(6) atoms to a carboxylic group (COOH) in moderate cellulose oxidation.[10] Interestingly, simulations performed with COOH showed instead no absorption bands in the UV-Vis range up to

FIG. 2. (a) R_∞ spectra measured in the spots shown in Fig. 1. (b) Experimental (lines) and theoretical (symbols) absorption spectra of spots Pv, Pr1, and Pr2 on Leonardo's self-portrait.
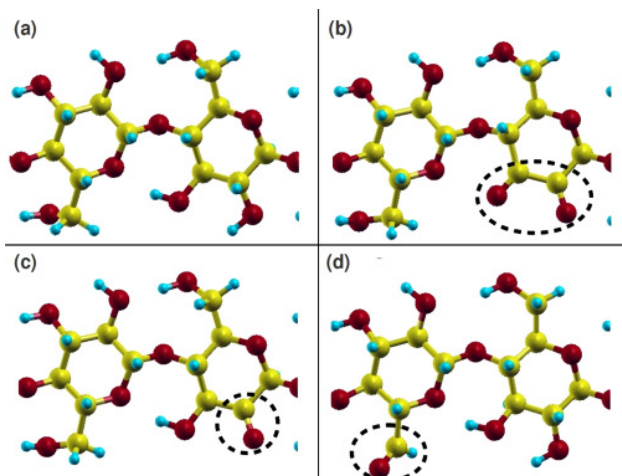

FIG. 3. Chemical structures of some representative oxidized groups of cellulose whose optical properties have been simulated using TDDFT: (a) pristine cellulose (unaged), (b) diketone, (c) ketone, and (d) aldehyde. Light gray (yellow), dark gray (red), and small (cyan) spheres represent carbon, oxygen, and hydrogen atoms, respectively.

258 nm (4.8 eV) and therefore no contribution to paper yellowing.

For each structure, the optical spectra were calculated according to TDDFT (see Figs. 4–6 of Ref. 11). Being the relative concentration of chromophores in paper samples unknown, theoretical spectra (symbols in Fig. 2) were ultimately obtained through a linear combination of single oxidized group spectra whose coefficients are determined by a best fit algorithm so as to minimize the mismatch with experimental spectra (lines in Fig. 2(b)). The linear combination coefficients obtained give an estimation of the concentration of chromophores inside aged paper,[20,21] and make it possible to obtain chemical information on the self-portrait by entirely non-destructive and non-invasive optical measurements.

To analyze our results, we divided the chromophores into three main categories according to their optical response: visible (VisAG), low-ultraviolet (LUVAG), and ultraviolet (UVAG) absorbing groups. The VisAG category contains the diketones (Figs. 3 and 4 of Ref. 11). The LUVAG category contains aldehydes and single ketones (Figs. 3 and 5 of Ref. 11). The latter, in position C(3) of a glucopyranose ring, are characterized by strong H-bonds with the hydroxyl group on the C(6) atom. Although the chromophores of the LUVAG category mainly absorb in the low UV range, they also display a smooth tail extending into the short end of the Vis region (blue-violet). Chromophores classified as VISAG and LUVAG are the ones that are the chief ones responsible for the yellowing of paper. Finally, the UVAG category contains single ketones free of hydrogen bond-type interactions which do not directly give rise to visible damage (Figs. 3 and 6 of Ref. 11).

In an attempt to quantify the chromophores which develop on the cellulose chains, the total concentrations of all of them (Fig. 4(a)), as well as the absolute and the relative concentrations of single categories of chromophores (Figs. 4(b) and 4(c)), were calculated for Leonardo's self-portrait and for a selection of reference samples. Reference samples A1, A2, and A3 are ancient papers (produced in Italy and France in the 15th century) in different states of degradation. Samples D, C, and V are modern papers which have been artificially aged in different environmental conditions, namely, dry air (sample D); humid air (at 59% relative humidity) (sample C); and humid air in a closed vessel (sample V)[2,8–10,13,20–22] (see supplementary material[28] for samples description). The total amounts of chromophores for Pv, Pr1, and Pr2 reported in Fig. 4(a) (respectively, 3.3, 4.0, and 6.1 mmol/100g of cellulose) are similar to those found in the ancient reference samples of a comparable age (3.1, 3.4, and 6.3 mmol/g for A1, A2, and A3 respectively). For samples D, C, and V, the total chromophore amounts are 0.1, 0.8, and 1.5 mmol/100g of cellulose, respectively. These values are lower than the concentrations of oxidized groups recovered using chemical analysis methods on samples aged under similar conditions.[9,23] This finding suggests that optically active oxidized groups represent a small fraction of the total number of oxidized groups.

The total concentration values of Pr1 and Pr2 are, respectively, about 20% and 80% greater than that of Pv. This points to the recto of the work having been heavily exposed to external degradation agents.[24–26] Unfortunately, this observation implies that any further exposure of the self-
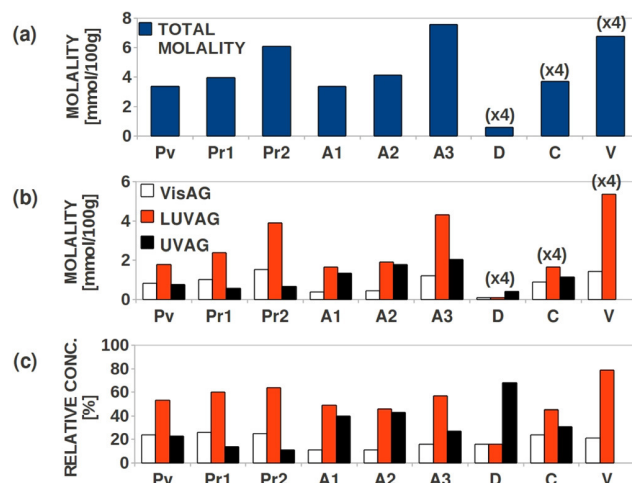

FIG. 4. Calculated total concentration (panel (a)), concentrations of each absorbing category (panel (b)), and relative concentrations (panel (c)) of UVAG, LUVAG, and VisAG chromophores in Pv, Pr1, and Pr2 spots on Leonardo da Vinci's self-portrait, on ancient reference samples (A1, A2, and A3), and artificially aged modern (D, C, and V) samples (multiplied by 4 for clarity).

portrait to such agents will only increase the difference in optical degradation between the recto and verso of the sheet. The important role played by moisture and pollutants in paper degradation is highlighted by the total concentration trend in the artificially aged D, C, and V samples, which strongly increases with exposure to humidity and degradation by-products during ageing (Fig. 4(a)).

Panels (b) and (c) of Fig. 4 offer a deeper insight into the chemical changes seen at the molecular level, since they show that optical damage induced by ageing is much more severe in the self-portrait in comparison to samples A1 and A2 due to the higher concentration of LUVAG and VisAG chromophores. LUVAG and VisAG total and relative concentrations in the da Vinci drawing are comparable to those measured in sample A3 (Figs. 4(b) and 4(c)). A3 is characterized by the presence of clear signs of damage caused by water spots,[2,8] evidence which highlights the important role played by humidity in the degradation of paper. It is known that water molecules provide additional oxygen radicals which facilitate the formation of VisAG and LUVAG in β-D-glucopyranose units.[27] The detrimental effect of humidity on paper is also supported by the selective formation of LUVAG chromophores upon passing from sample D to sample C, and finally to sample V. The worst affected is sample V, where ageing in a closed humid environment caused the volatile products of paper degradation[8] to linger, thereby increasing the formation of LUVAG oxidized groups. The high concentration of LUVAG chromophores in Leonardo's self-portrait tallies with accounts in Ref. 24 regarding the deterioration which has occurred during its documented history, its misguided conservation behind glass in a closed frame, in particular. The presence of trapped moisture and degradation products within a sealed housing may have catalyzed further oxidative processes in addition to those which were already underway.[25] It should be noted, however, that a certain amount of moisture is necessary for effective paper conservation, since it prevents so-called hornification.

The sadly poor state of preservation that characterizes Leonardo's self-portrait today is the result of the inappropriate conditions in which it was historically stored. Unfortunately, a lack of sufficient knowledge, both physical and chemical, of the mechanisms responsible for paper degradation prevented good conservation strategies from being adopted in the past. Our non-destructive and non-invasive diagnostic method provides a quantification of this priceless artwork's present state of optical degradation. A periodic repetition of the same analysis not only would provide an ongoing quantitative assessment of its rate of degradation (and therefore an estimation of its "life expectancy") but also increase our understanding of the inevitable degradation processes that are underway. Needless to say, such information is invaluable to restorers and conservators. Our method has allowed us to gain a clear insight into this iconic artwork's current state of preservation and represents an important contribution to ensuring that it will survive and be appreciated by future generations.

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