Role of protolytic interactions in photo-aging processes of carminic acid and carminic lake in solution and painted layers

Gianna Favaro,* Costanza Miliani, Aldo Romani and Manuela Vagnini

Università di Perugia, Dipartimento di Chimica, 06123 Perugia, Italy.
E-mail: favaro@phch.chm.unipg.it; Fax: 039 075 585 5598

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In this paper absorption and fluorescence spectra and emission quantum yields and lifetimes of the red colorants, carminic acid and its metal complex, carminic lake, were studied in solution and on painted surfaces. Accelerated photo-aging of carminic acid and lake was investigated in solution, in the presence and absence of a binder (arabic gum) commonly used in water-colour painting, while natural ageing was followed for several months on water-colour painted paper.

The study of carminic acid in water as a function of pH showed that the absorption spectrum changes with pH. Four acid–base dissociation steps were detected and the corresponding p*K*s were determined from spectrophotometric and fluorimetric titrations. The fluorescence quantum yields (in the 10^{-2}–10^{-4} range) and the lifetimes (on the sub-nanosecond timescale, 90–1000 ps) were markedly dependent on the pH of the medium. Excited state p*K**s were calculated by means of the Förster cycle. The acidity decreased upon excitation for the first deprotonation step involving the –COOH group ($\Delta pK^* = -1.9$), but increased slightly for the successive deprotonation steps involving three phenolic hydroxy groups ($\Delta pK^* = 0.6$, 2.0 and 0.2, respectively).

The results obtained from the aging experiments indicate that both carminic acid and lake are bleached during irradiation. While the binder prevents lake from fading, it destabilises the carminic acid. These findings are discussed in the light of the interactions of the dye with the solvent and matrix.

Introduction

Lake pigments are translucent dyes prepared by precipitating or adsorbing an organic dye onto an inert inorganic substrate. These coloured substances were essential constituents of the artist's palette from the fourteenth to the nineteenth centuries.[1] In this work, absorption and luminescence properties of carminic acid (CA) and its metal complex (carminic lake, CA–Al) were investigated in solution and on painted surfaces.

Carminic acid is a β-*C*-glycopyranosyl derivative of anthraquinone (Fig. 1) which is extracted from the cochineal, a

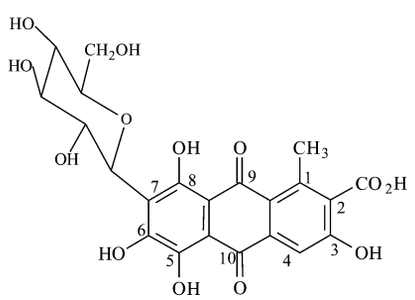

Fig. 1 Structural formula of carminic acid (CA).

tropical American insect that feeds on certain species of cactus.

The related lake is an aluminium two-ligand complex, in which the Al atom co-ordinates two dye molecules through the 5-hydroxy oxygen and the adjacent carbonyl oxygen, forming a six-membered chelate ring structure.

This molecule belongs to the class of anthraquinonic dyes, which have been studied extensively due to their practical application as colorants (*e.g.*, in paints, foods, cloth, *etc.*). In addition, there is basic interest in these molecules because modest structural changes induce drastic modifications in the absorption and fluorescence characteristics.[2–4] The peculiar spectral behaviour of these compounds has been explained by considering the stability of intramolecular hydrogen-bonding in the ground and excited states, which may or may not undergo excited state intramolecular proton transfer (ESIPT) from the phenolic hydroxy group to the carbonyl oxygen upon excitation.[5]

Over the last decade, great interest has been devoted to carminic acid, mainly in relation to its extensive use as a natural food and cosmetic pigment. Various studies have investigated photodegradation,[6] fluorescence properties in both aqueous solution[7] and organic solvents,[8] reorientation dynamics,[8] self-assembly,[9] and acid–base behaviour.[6,10]

In the present paper, attention is focused on the use of carminic acid and carminic lake in paints and the possible colour changes or colour fading that they may undergo with aging.

Paintings are exposed to atmospheric pollutants and solar irradiation, and undergo natural aging. This exposure may cause deterioration, particularly by changing the original colours. It is important to know which chromatic changes occur as a result of environmental changes. Acid rain is one form of pollution that attacks painted artefacts. By studying protolytic equilibria involving the multiple acidic centres of this molecule and the accompanying colour changes, information can be obtained about the environmental effects as well as about possible interactions of the dye with the binders and varnishes present in the painted layers. Even though several papers from the literature report results on acid-base titration and fluorescence measurements of carminic acid,[6–10] some discrepancies between the literature data and our findings induced us to reproduce and present both spectral and titration data in this paper.

Light can also alter colours and damage painted surfaces. Light-induced effects may have dramatic consequences on the chromatic features of paintings. In order to evaluate the aging

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that might occur indoors in a museum, painted layers and dye solutions (for the purpose of comparison) were irradiated with a lamp to simulate daylight. Their spectral changes were periodically analysed.

Experimental

Materials

The carminic acid was purchased from Aldrich and used without further purification. The carminic lake was purchased from Zecchi (Florence, Italy). The organic solvent used, DMSO, was a Fluka spectrograde product. Arabic gum was from Winsor & Newton, London. To determine the p*K* values, Britton buffers were used in the 2–12 pH range; diluted $HClO_4$ and NaOH solutions were used in the acidic and alkaline regions, respectively. Re-distilled water was used to prepare the buffers.

To prepare the samples of painted surfaces, the dye was mixed with the binder and painted onto a piece of paper.

Apparatus

The absorption spectra in solution were recorded with a Perkin Elmer Lambda 16 spectrophotometer. The Perkin Elmer accessory for reflectance measurements was used to record absorption on the painted surfaces.

Corrected emission spectra were obtained using a Spex Fluorolog-2 FL 112 spectrofluorimeter controlled by Spex DM3000F spectroscopy software.

To record temperature and solar radiation values during the natural aging experiments, a data-logger instrument for recording and statistically elaborating temperature and radiance data (Babuc, from LSI, Milan, Italy) was used. Typical histograms of solar radiance and temperature distribution during the period of observation (eight months) are reported in Fig. 2.

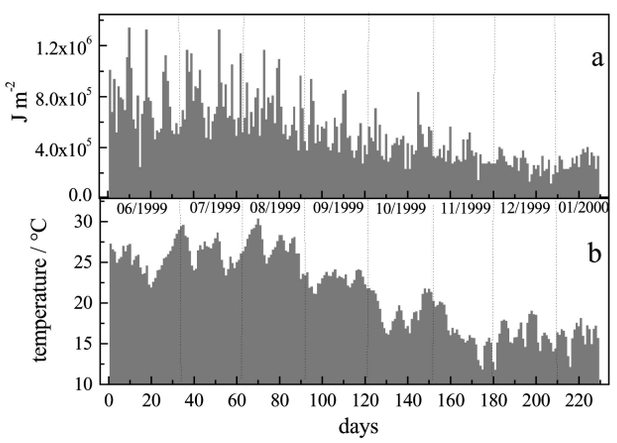

Fig. 2 Radiation intensity (a) and temperature (b) distributions during an eight-month period.

For the accelerated photo-aging trials, carried out on the dye in DMSO solution, a 150 W Xenon lamp filtered through a Pyrex filter was used. The exposure times did not exceed 50 hours.

Measurement conditions

The emission quantum yields (Φ_F) were obtained by comparing corrected areas of the sample ($area_S$) and the standard ($area_{ST}$) emissions, using quinine sulfate in 0.5 M H_2SO_4, $\Phi_{ST} = 0.546$, as the standard.[11] The formula in eqn. (1)

$$\Phi_F = \Phi_{ST} \times (A_{ST}/A_S) \times (area_S/area_{ST}) \times (n_S^2/n_{ST}^2) \quad (1)$$

was used; this accounts for the differences between the absorbance and the refraction index of the sample (A_S, n_S) and standard (A_{ST}, n_{ST}) solutions.

Sample concentrations were adjusted in order to keep the absorbance within 0.2–0.5. This is a compromise between needing to have a detectable signal and the requirement to be within the linear range of the absorbance. The relatively large Stokes shifts militate against self-absorption of the fluorescence emission. The solvent contribution to the emission signal was subtracted, when necessary. The accuracy in the Φ_F values is estimated to be within 10% for $\Phi_F \geq 10^{-3}$ and within 20% for $\Phi_F < 10^{-3}$.

The emission lifetimes (τ_F) were determined by using the Pockel's cell of the Spex spectrofluorimeter accessory, which is based on the phase-shift method (time resolution *ca.* 20 ps). The light scattered by an aqueous glycogen solution (undetectable fluorescence) was used as the standard. The accuracy of the data obtained depended on the intensity of the emission. The data were processed using Global Unlimited™ 3.0 software, which allows the analysis of multiple decays of up to four components.

Spectrophotometric and fluorimetric titrations

Low ionic strength Britton buffers (μ = 0.01 mol dm^{-3}) were used to determine the p*K*s. The pH values were measured with an Orion 9103 pH-meter.

The dissociation constants were obtained from the inflection points in the absorbance *vs.* pH curves at the wavelengths of the maximum difference in absorbance of the equilibrating species. In the fluorimetric titrations, the excitation was carried out at an isosbestic point between the spectra of the equilibrating species; the inflection points of the emission intensity *vs.* pH curves at suitable wavelengths gave the dissociation constant values. Sample concentrations were on the order of 10^{-5} mol dm^{-3}. Given the low concentration and low ionic strength of the buffer solutions, the activity coefficient ratio of the acid–base couples was considered to be one.

Results and discussion

Effect of pH on the spectral characteristics of carminic acid

The effect of pH on the absorption and emission spectra of carminic acid was investigated in the 0.9–13 pH range. Dilute solutions (<5 × 10^{-5} mol dm^{-3}) were used in order to avoid aggregation. The lake could not be investigated in aqueous solution due to its poor solubility.

Absorption spectra. Four species were detected from the absorption spectra of CA in the pH range explored (Fig. 3). Each of them was stable within a limited pH interval and

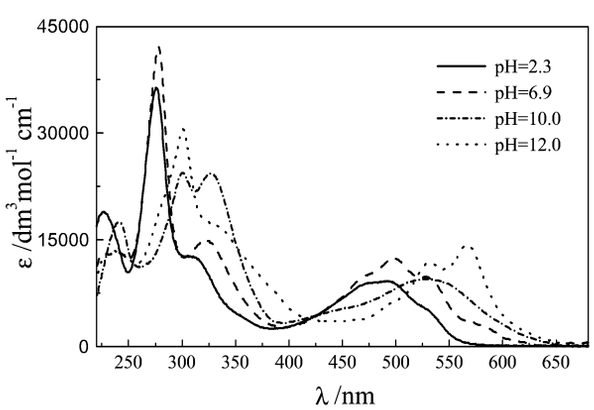

Fig. 3 Absorption spectra of CA in aqueous solution at various pH values.

was the precursor for the formation of the successive one as the pH changed. This was supported by the presence of well-defined isosbestic points in the absorption spectra and the reversibility of the transformation. The absorption maxima shifted to the red as the pH increased (from 490 nm at pH = 2 to 567 nm at pH = 12). The whole spectral shift corresponds to a colour change from yellow-orange (pH = 2) to violet (pH = 12). The spectral region covered by the absorption is approximately the same as that of other hydroxyanthraquinones (*e.g.*, alizarin and purpurin),[12] that is, the glycosyl moiety has little effect on the energy of the lowest singlet excited state.

Fluorescence spectra. As found for other hydroxyanthraquinones,[12] carminic acid was fluorescent over the whole pH interval explored upon excitation with monochromatic light between 475 and 525 nm, depending on the pH. The fluorescence quantum yield and lifetime are critically dependent on the pH. The most important finding obtained from the fluorimetric measurements was the detection of a fifth species, in the acidic region (pH ~ 1), in addition to the four species observed from the absorption spectra. This species, assigned to the fully protonated neutral form, has absorption and emission bands positioned like those of the mono-anion, but its fluorescence quantum yield is only about one third. The emission intensity increases from the neutral molecule ($\Phi_F = 1.0 \times 10^{-3}$) to the mono-anion ($\Phi_F = 3.3 \times 10^{-3}$) and reaches a maximum value for the di-anion ($\Phi_F = 1.3 \times 10^{-2}$). It becomes almost undetectable at pH = 10 ($\Phi_F = 6 \times 10^{-4}$) but then increases at pH ≥ 12 ($\Phi_F = 2.5 \times 10^{-3}$). These results do not completely agree with the literature data,[7] which report that relative fluorescence quantum yields decrease and are almost completely quenched in alkaline solution (pH > 10). These discrepancies are probably due to the higher ionic strength used and lower alkaline limit explored. The fluorescence spectra are broad and structureless. The maxima shift to the red with increasing pH. This finding is in contrast with the literature report that the emission response shifts to higher energies with increasing pH.[9] The Stokes shifts, calculated using the maxima of the absorption and fluorescence spectra, decrease from 3500 cm^{-1} in acidic solution to 2000 cm^{-1} in alkaline solution. This reflects a smaller difference in stabilisation energy, and therefore in dipole moment, of the excited and ground states when the negative charge on the molecule increases. Absorption and fluorescence spectra (normalised on the maximum) are shown in Fig. 4; the spectra of the neutral form are not reported in the figure since they overlapped with those of the mono-anion when normalised.

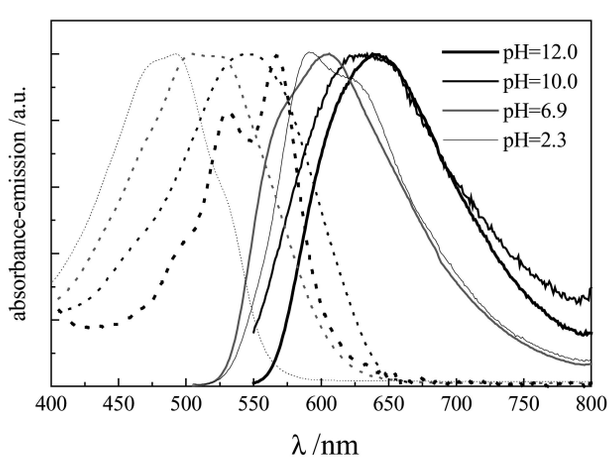

Fig. 4 Fluorescence spectra (solid line) normalised to absorption spectra (dotted line) of the four species spectrally distinguishable when the pH changes.

The fluorescence lifetimes, determined by the phase-shift method, are of the order of hundreds of picoseconds. At certain pH values, the decay kinetics fit a bi-exponential function well; contributions from the two species are pH dependent. Detection of two distinct lifetimes is a good indication that acidic and basic excited species do not equilibrate within the excited state lifetime. The rate parameters for emission, $k_F = \Phi_F/\tau$, and radiationless deactivation, $k_{NR} = (1/\tau) - k_F$, were determined from the experimental Φ_F and τ data and are reported in Table 1.

The Φ_F and τ values were strongly pH dependent. They were in the 10^{-2}–10^{-4} and 90–1000 ps ranges, respectively. The k_F values were of the order of 10^7 s^{-1} over the entire pH range explored, while k_{NR} varied from 1×10^{10} to 10×10^{10} s^{-1} depending on the pH value. The k_{NR} rate constant includes both internal conversion to the ground state and intersystem crossing to the triplet state. Triplet–triplet absorption was, in fact, detected in a butanol solution by laser flash-photolysis (λ_{exc} = 470 nm). The transient produced, with a lifetime on the microsecond timescale (τ = 3 μs), was quenched by oxygen at a diffusion-controlled rate.

Acid–base properties

To determine the p*K*s of the protolytic dissociations, spectrophotometric and fluorimetric titrations were employed. Inflection points were observed within the pH intervals where isosbestic points were maintained between the two equilibrating species.

As for the 2-hydroxyanthraquinones,[12] the fluorimetric titration curves showed inflection points at the same pH values as the spectrophotometric ones, for pK_1, pK_2 and pK_3. Therefore, these curves also provided the ground state p*K*s, since the protolytic equilibrium in the excited state was not established within the lifetime of that state.[13] The deprotonation step occurring in the acidic range (pK_0 = 2.9), which was only detected by fluorescence, could have been assigned to excited state acid–base equilibrium. However, comparison with literature values[6,10] indicates that pK_0 corresponds to the deprotonation of the most acidic site in the ground state molecule. The p*K*s determined are reported in Table 2 and are compared with values from the literature;[6,9,10] the fourth p*K* (~13) was only roughly estimated.[9]

The excited state p*K*s*, calculated by use of the thermodynamic Förster cycle,[14] are also reported. The frequency differences between 0–0 levels, needed to calculate the p*K**, were obtained from the intersections of the normalised absorption and fluorescence spectra. The acidity may increase or decrease upon excitation. For carminic acid, the acidity decreased in the excited state for the first deprotonation, while, for the successive deprotonations, a slight increase in acidity was observed upon excitation, even though the increase was much smaller than for other hydroxyanthraquinones.[12,15]

These results give a complete view of the acid–base equilibria of this molecule and allow the spectra to be assigned to the molecular species that interconvert one into the other as the pH changes. In carminic acid, where several acidic centres are present, the first dissociation step (pK_0 = 2.9) definitely occurs at the carboxylic group in the 2 position, which is the most acidic site. It is more acidic than 2-anthraquinoic acid (p*K* = 4.2),[16] due to the inductive electron-attracting effect of the phenolic OH group in the 3 position, which decreases the charge density in the 2 position and consequently lowers the p*K* of the COOH group. Deprotonation at the carboxylic group does not produce appreciable spectral changes in this molecule, where the chromatic properties are essentially determined by the hydroxyanthraquinone chromophore. Further support for this assignment is the p*K* increase in the excited state, which generally decreases for phenolic hydroxy groups. Accordingly,

Table 1 Fluorescence quantum yields and dynamic parameters of the singlet excited state of carminic acid at pH values typical of different deprotonated forms. Only the main fluorescence components (corresponding to emission from the status indicated in parentheses) are reported

pH (status)	Φ_F	τ/ps	$k_F/10^7\ s^{-1}$	$k_{NR}/10^9\ s^{-1}$
0.9 (Neutral)	1.0×10^{-3} (1.33×10^{-3})[a]	(93)[a]	(1.4)[a]	(11.0)[a]
2.3	1.8×10^{-3}	94	1.9	10.6
3.3 (Mono-anion)	3.3×10^{-3}			
6.9 (Di-anion)	1.3×10^{-2}	940	1.4	1.0
10.0 (Tri-anion)	6.2×10^{-4}	Not measurable		
12.4 (Tetra-anion)	2.5×10^{-3}	330	0.75	2.9

[a] Data taken from ref. 7.

Table 2 Dissociation constants (pK_0, pK_1, pK_2 and pK_3) of carminic acid obtained from spectrophotometric titration (a), fluorimetric titration (b) and from the literature (c). Excited state pK*s calculated by means of the Förster cycle. ΔpK^*s represent the differences between the ground state pK and the corresponding excited state pK^*.

	pK_0	pK_1	pK_2	pK_3
Ground state (a)		5.47	8.47	12.10
(b)	2.90	5.31	8.85	12.37
(c)	2.91[a]	5.62[a]	8.30[a]	~13[b]
	2.81[c]	5.43[c]	8.10[c]	
Excited state	4.8	4.8	6.7	12.0
ΔpK^*	−1.9	0.6	2.0	0.2

[a] Data taken from ref. 10. [b] Data taken from ref. 9. [c] Data taken from ref. 6.

the acidity increases in the analogous 2-anthraquinoic acid upon excitation ($\Delta pK^* \sim -2$).[16]

The following acid–base equilibration ($pK_1 = 5.4$) involves the 6-hydroxy group since the 5- and 8-OH groups are less acidic due to their position with respect to the carbonyl, which reduces the hydrogen mobility through hydrogen bonding. Moreover, the 6-hydroxy group is far enough away from the negatively charged site, COO^-, that its ionisation is only weakly influenced by the repulsive electrostatic potential. This assignment is in contrast with that reported in the literature, where the $pK = 5.62$ is attributed to dissociation of the OH in the 5 position.[10]

Even though the double negative charge of the di-anion should lower the acidity of the other hydroxy groups, two further deprotonation steps ($pK_2 = 8.7$ and $pK_3 = 12.2$) were observed upon decreasing the acidity, assigned to the dissociation of the 3- and 8-hydroxy groups. The absorption spectrum of the tetra-anion, which is stable at pH > 12, is structurally similar to that of the purpurin di-anion, which involves 2- and 4-hydroxy deprotonations.[12] Since the 2 and 4 positions in purpurin are equivalent to the 6 and 8 positions in carminic acid, the latter are probably both deprotonated at pH = 12.

The photophysical data reported in Table 2 indicate that the variations in Φ_F and τ_F with deprotonation essentially depend on the different contributions of non-radiative relaxation, since all the k_F values are of the same order of magnitude. The relatively long lifetime of carminic acid at pH = 6 (di-anionic form) is the result of the intramolecular hydrogen bonds of the 5- and 8-hydroxy groups that keep the ion rigid. The clear-cut decrease of emission intensity of the tri-anion (stable at pH ~ 10) favours the deprotonation at the 8 position. The further increase of the emission quantum yield of the tetra-anionic form is probably related to the loss of relaxation paths through OH vibrations. For this molecule, acid–base equilibria in the excited state involve intramolecular hydrogen-bonded forms, as in the ground state, due to minute changes in the geometry upon excitation. Semiempirical calculations have shown that there are no marked changes in dipole moment upon excitation.[17] This also explains the small increase in acidity upon excitation.

Effects of environment and aging on the spectral properties of CA and CA–Al

An important aspect of studies on the effect of pH on colorants used in artistic painting is its potential to interpret the interactions of colorants with atmospheric pollutants, as well as within their own chemical environment. In painting, dyes and pigments are usually dispersed in a binder and protected by a top layer of transparent varnish. The spectral features of the absorption and emission bands of the colorant depend on both the aging and interaction of the dye with the additives. Thus, spectra can be used as a diagnostic test of the medium used by the artist and can help to evaluate how the durability of the colorant can be increased or decreased by the presence of additives.

Study in solution. Investigating solutions where the colorant was dissolved in the presence or absence of a binder was the first approach used to study the interactions of the dye with the additive and to detect colour fading, if any, due to photo-aging. Carminic acid is soluble in most polar solvents, while lake could only be dissolved in DMSO. For this reason DMSO was the solvent chosen for the study. Spectra and spectral evolution under irradiation of carminic acid and its complex in the presence or absence of additive are shown in Figs. 5 and 6. It

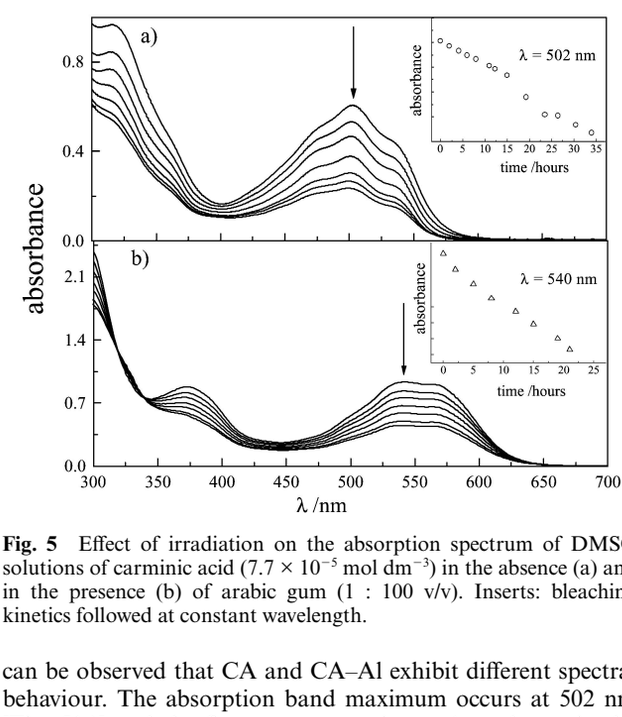

Fig. 5 Effect of irradiation on the absorption spectrum of DMSO solutions of carminic acid (7.7×10^{-5} mol dm^{-3}) in the absence (a) and in the presence (b) of arabic gum (1 : 100 v/v). Inserts: bleaching kinetics followed at constant wavelength.

can be observed that CA and CA–Al exhibit different spectral behaviour. The absorption band maximum occurs at 502 nm [Fig. 5(a)] and the fluorescence maximum (not shown in the figure) at 581 nm for CA, while both the absorption [$\lambda_{max} = 527$ nm, Fig. 6(a)] and emission spectra ($\lambda_{max} = 616$ nm) are red-shifted by about 30 nm for CA–Al. In DMSO, the emission quantum yield is one order of magnitude lower for CA–Al than for CA ($\Phi_F = 10^{-3}$ *vs.* 10^{-2}). The spectrum of CA in DMSO is

Table 3 Photobleaching parameters ($t_{½}$, Φ_{rel}) of CA and CA–Al in the absence and in the presence of arabic gum in DMSO solution

	CA	CA–Al	CA–arabic gum	CA–Al–arabic gum
$t_{½}$ (hours)	25	38	18	220
Φ_{rel}	0.873	0.339	1	0.057

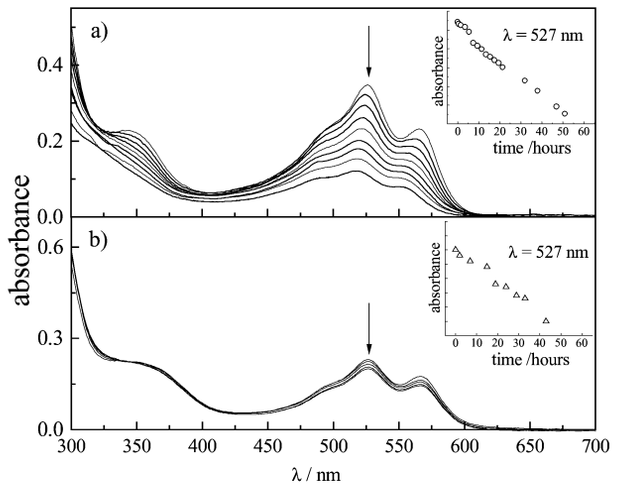

Fig. 6 Effect of irradiation on the absorption spectrum of DMSO solutions of carminic lake (2.8×10^{-5} mol dm^{-3}) in the absence (a) and in the presence (b) of arabic gum (1 : 100 v/v). Inserts: bleaching kinetics followed at constant wavelength.

similar to that observed in water around pH = 7; this corresponds to the di-anion which also has the highest fluorescence yield. The CA–Al spectrum does not resemble any of the spectra of the poly-anions obtained from the carminic acid in water, but, rather, it resembles the absorption spectrum of the 1,2-dihydroxyanthraquinone in the di-anionic form (pH ~ 14).[12] It is conceivable that this latter absorption could be assigned to an anionic-like form, involving the 5-OH group, which is engaged in the complex (note that the 5-OH of CA does not deprotonate in the pH range explored since it is the most basic one). Prolonged irradiation of these solutions with a Xenon lamp filtered through a Pyrex filter bleached the colour band in both cases [Figs. 5(a) and 6(a)].

When arabic gum, usually used as a binder with hydrosoluble dyes, was added to the solutions, the CA spectrum changed further [Fig. 5(b)], while that of CA–Al remained substantially unchanged [Fig. 6(b)]. The red-shift ($\lambda_{max} \approx 550$ nm) exhibited by the CA spectrum in the presence of the binder suggests that a further deprotonation occurs. The spectrum is, in fact, very close to that observed in aqueous solution at pH ≈ 10. Upon irradiation, the solution containing CA and arabic gum showed a very marked decoloration [Fig. 5(b)], while that containing the lake was highly photostable [Fig. 6(b)]. Thus, it has been established that arabic gum has a photo-stabilising effect on lake, by reducing the degradation compared with a solution of lake alone. Photobleaching quantum yields were estimated relative to the most photodegradable system, that is, CA + binder ($\Phi = 1$) by using the relationship given in eqn. (2):

$$\Phi \propto \frac{0.1 \times C_0}{t \times I_0 \times \int \varepsilon_\nu \mathrm{d}\nu} \qquad (2)$$

where C_0 is the initial dye concentration and I_0 is the intensity of the irradiating source. The absorbed photons were considered proportional to the integral of the absorption spectrum over the emission range of the source (340–700 nm) and referred to the same amount of bleaching (10%) produced during an irradiation time of t hours for all samples. Due to the low transformation percentage, the absorbed quanta can be considered to be approximately constant. The relative quantum yields are reported in Table 3 along with the half-lives ($t_{½}$) of the colour intensity, determined (or extrapolated) from the bleaching kinetics in the visible absorption region. It can be seen from the table that the solution of carminic acid containing arabic gum has the maximum degradation yield, while the additive has an efficient stabilising effect on the lake solution. Arabic gum plays a twofold role: it acts as an inner filter of UV light ($\lambda \leq 350$ nm) and increases the basicity of the medium. The latter effect is analogous to that found for aqueous glucose solutions of carminic acid.[9] The red shift of the CA absorption spectrum in the presence of binder indicates that prototropic interaction occurs between CA and binder, in which the latter behaves like a base with respect to the dye. This interaction, causing further deprotonation, increases the sensitivity of the colorant to photodegradation. In the lake, the bond to the metal through the 5-hydroxy group lowers the acidity of the free hydroxy groups, and the spectrum remains unchanged. In this case the photoprotection is provided by the binder filtering the UV radiation.

A study on the photodegradation of CA as a function of pH, showed that photodegradation increases as the pH increases.[6] This is in agreement with the above measurements, since the greatest degradation occurred under conditions where the absorption spectrum showed the presence of a poly-deprotonated form, that is, for the CA-arabic gum system.

Study on painted surfaces. To investigate the effects of the binder and aging on painted surfaces, samples were prepared with the dye and binder spread over pieces of paper, using the "water painting" technique. In the previous investigations on accelerated aging (some tens of hours) of the dyes in solution, the temperature effect was not important due to the relatively short irradiation times. Photo-aging on painted surfaces was carried out under ambient conditions (natural aging) over a period of eighteen months. Under these conditions, temperature may also have had an important effect. Both temperature and solar irradiation intensity were recorded by the instrument (Babuc) described in the Experimental section. Reflectance spectra of water-painted papers, determined from time to time, are shown in Figs. 7 and 8, for CA and CA–Al, respectively.

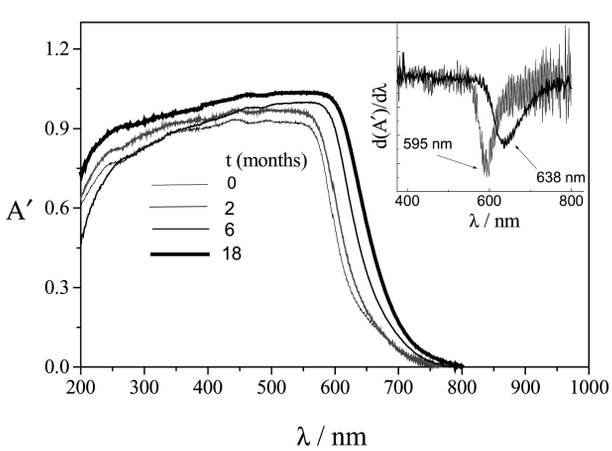

Fig. 7 Effect of irradiation on the reflectance spectrum of a water-painted paper with carminic acid. $A' = \log 1/R$ (R is the reflectance signal).

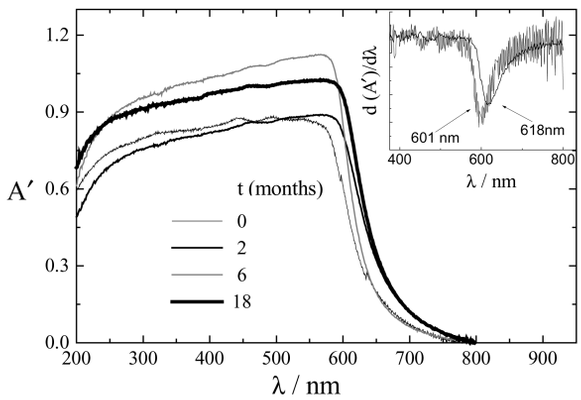

Fig. 8 Effect of irradiation on the reflectance spectrum of water-painted paper with carminic lake. $A' = \log 1/R$ (R is the reflectance signal).

The reflectance spectra are less informative than normal absorption spectra. In this case, the inflection point, obtained from the first derivative (see inserts), is meaningful. Red shifts were observed for both the CA and CA–Al spectra with aging. The amount of degradation was estimated from the shift to the red of the minimum of the derivative. It can be seen from the figures that, during the same aging time (eighteen months), the spectrum of the CA painted paper red-shifted 43 nm, whereas that painted with lake shifted only 17 nm. This confirms that the aluminium complex is the most stable colorant, as indicated from the study in solution.

Conclusions

A study of the pH effect on absorption and fluorescence spectra of carminic acid allowed the ground and excited state p*K*s to be determined. In the pH range explored, four acid–base dissociation steps were detected. By assigning the absorption and fluorescence spectra, the different forms, stable at certain pH values, were characterised (neutral form, mono-anion, di-anion, tri-anion and tetra-anion). This provides a valuable analytical tool for investigating interactions involving the hydrogen transfer processes between the dye and its chemical environment.

Carminic acid behaves like an acidichromic molecule since its aqueous solutions change colour as the pH changes. Emission is less sensitive to changes in the pH than absorption, since the fluorescence spectra of the differently protonated forms widely overlap. The fluorescence quantum yields and lifetimes are markedly dependent on the pH of the medium. Radiationless processes (internal conversion and intersystem crossing to the triplet) contribute greatly to excited singlet deactivation.

In the excited state, the neutral molecule becomes a weaker acid upon excitation, since the carboxy group is involved in the first dissociation step, while the hydroxy species, involved in the successive dissociation steps, are slightly stronger acids in the excited state. However, the p*K* decrease upon excitation is not as great as in other hydroxyanthraquinones.

The information about the spectral properties and their pH dependence has shed light on the spectral changes occurring in the presence of the binder. The absorption spectrum of the dye, before and after irradiation, depends on its environment. Thus, colour can be a diagnostic test of the medium used by the artist. All solutions were bleached upon irradiation and the degree of bleaching depended on the environment. The photoproducts absorb mainly in the UV region, thus photodegradation causes colour fading, not colour changes. When binder (arabic gum) was added to the solution, different effects were produced on the CA and CA–Al spectra and their photobleaching; the durability of CA decreased, while lake was efficiently stabilised. This explains why lakes have been widely used as colorants in painting. Finally, the spectral changes induced by photo-aging on painted surfaces may indicate the transformations that might occur indoors in a museum and may be used to interpret this effect in terms of the interactions of the dye with its environment.

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