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Photophysics and photochemistry of tetrahydrocurcuminoids

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Abstract

The photochemical properties of 10 tetrahydrocurcuminoids (THCs) have been studied for the first time. They display very low fluorescence in ethanol solution at room temperature with quantum yields between 0.9 and 13×10^{-3} whereas their phosphorescence quantum yields in ethanol glass at 77 K are between 0.025 and 0.5. Their disappearance quantum yield measured in ethanol solution, between 5×10^{-4} and 10^{-2}, follows the same trend as their antiradical power efficiency (ARP) previously measured. The reactivity of THCs is enhanced if they include a phenol group in meta or para of the linking chain and a phenol or methoxy group as neighbor. UV absorption difference spectra of the irradiated solutions, HPLC and GC–MS analyses performed on 4-propylguaiacol (4PG), which displays similar behavior as tetrahydrocurcumin (THC2) indicates the formation of quinone methides and *ortho*-quinone. The formation of a phenoxy radical by photooxidation appears to be the first reaction step. This demonstrates that the general photodegradation mechanism of phenols found for guaiacyl lignins applies also to simplest molecules of natural origin such as THCs. The irradiation of the THCs in carboxymethylcellulose films follows the same trends as the solution studies, except that the quinone methides and *ortho*-quinones are not detected, probably because they react with the cellulosic matrix. The knowledge obtained in this study probably will help to promote the use of tetrahydrocurcuminoids in various applications where it is necessary to develop antioxidant properties in presence of solar light.

Keywords: Tetrahydrocurcuminoids; Fluorescence; Phosphorescence; Phenol; Photochemistry; Antioxidant

1. Introduction

Tetrahydrocurcuminoids are obtained from curcuminoids extracted from the roots of *Curcuma longa* L., commonly called turmeric. Curcumin of natural origin contains in addition to curcumin, (1,7-bis(4-hydroxy-3-methoxyphenyl)-1,6-heptadiene-3,5-dione), demethoxycurcumin and bis-demethoxycurcumin in smaller quantities [1]. Turmeric is one of the major spices and food coloring, improperly called Indian saffron, much less expensive than the true saffron [2]. Structurally, curcumin belongs to the diarylheptanoids of naturally occurring 1,3-diketones in which the carbonyl groups are directly linked to olefinic carbons [3]. Curcumin shows remarkable pharmacological activity: it is a very strong but safe anti-inflammatory agent [4]; it displays some inhibition of the HIV proteases [5] and it seems to have anti-cancer activity [6]. Curcumin acts as a lipoxygenase substrate [7] and also inhibitor of cyclooxygenase enzymes [8]. The main action of curcumin is due to its ability to inhibit the formation of reactive oxygen species such as hydroxy radicals and superoxide anion [9–11].

Tetrahydrocurcuminoids (THCs), produced from curcuminoids by hydrogenation, are colorless which render these products useful in non-colored food and cosmetic applications that currently employ synthetic antioxidants [12]. Tetrahydrocurcuminoids appear to be the major active metabolites formed when curcuminoids are intraperitoneally administered to mice [13]. Several independent studies reported the significant antioxidant effects of the tetrahydrocurcuminoids obtained from turmeric [14–16]. They display also chemopreventive effects on mouse colon carcinogenesis [17].

We have previously synthesized and studied curcuminoid compounds as possible photoprotective molecules for lignocellulosic materials [18]. For this purpose, we examined the

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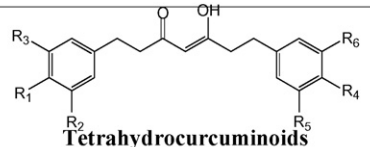


Tetrahydrocurcuminoids

Compounds	R1	R2	R3	R4	R5	R6
THC1	H	H	H	H	H	H
THC2	OH	OMe	H	OH	OMe	H
THC3	OMe	OH	H	OMe	OH	H
THC4	OMe	H	H	OMe	H	H
THC5	OMe	OMe	H	OMe	OMe	H
THC6	OMe	OMe	OMe	OMe	OMe	OMe
THC7	OH	OMe	OMe	OH	OMe	OMe
THC8	OH	H	H	OH	H	H
THC9	OH	OH	H	OH	OH	H
THC10	OH	H	H	OH	OMe	H

Scheme 1. Formulae and abbreviated names of the studied THCs.

photochemical behavior of curcumin, dimethylcurcumin and non-substituted curcumin [19]. Their similar behavior indicated a moderate role of phenol groups in the photodegradation process. Structural analysis of photodegradation products has shown among other products formation of a flavanone molecule. It represented a unique example of photochemical conversion of a diarylheptanoid molecule into a flavonoid, another very important class of natural products.

The phenolic group of curcuminoids plays the major role in their antioxidant and free scavenging activities [20]. Very recently [21], we have studied a series of curcuminoids and tetrahydrocurcuminoids, bearing various hydroxy and methoxy groups on their benzene subunits. We described the syntheses and a systematic determination of their antioxidant and hydrogen donating capabilities using the DPPH method at 25 °C in methanol. The results showed that the tetrahydrocurcuminoids were in general much more efficient than their curcuminoid analogs, if they include both a phenol group in meta or para of the linking chain and a phenol or methoxy group as neighbor. This efficiency gain of THCs by comparison to curcumins was not attributed to the presence of the beta-diketone moiety in the chain, as it was already proposed [15], but to the presence of benzylic hydrogens, which are involved in the oxidation process of these compounds and not in curcuminoids.

The photophysics and photochemical behavior of THCs have never been described. Considering their actual uses in color-free food and cosmetic products, the knowledge of the response of tetrahydrocurcuminoids to UV-light appears to have great interest. Moreover, the state of Minas Gerais is one of the main place producing turmeric in Brazil, and development of new applications for this type of compounds is searched. In this paper, we report on the fluorescence, phosphorescence and photochemical behavior of 10 tetrahydrocurcuminoids described in Scheme 1. Their photochemical reactivities in ethanol solution and carboxymethylcellulose (CMC) films will be compared to their antioxidant properties [21]. The information gained might be very helpful to use tetrahydrocurcuminoids to protect cellulosic materials.

2. Results and discussion

The syntheses of studied compounds have already been described [21]. As for curcumin [3], the ^{1}H NMR spectra of the studied tetrahydrocurcuminoids show that in methanolic solutions, they exist only in their enol form [21].

2.1. Photophysics of THCs

2.1.1. UV absorption in alcoholic solutions

All the studied tetrahydrocurcuminoids in ethanol solution present a strong absorption band centered at 280 nm, which extends to 330 nm (Fig. 1). The part above 300 nm is the most important region of their absorption related to their photostability against solar light emission which extends to 300 nm.

The absorption is due to two subunits: the polysubstituted benzenic ring and the enone chromophore of the chain linking the two ends. This is exemplified in Fig. 2 for THC2: the theoretical spectrum constituted by the sum of the spectra of acetylacetone (in its enolic form) and twice the absorption of 4-*n*-propylguaiacol is very similar to the experimental one, except the value of the extinction coefficient which is one third higher. Some interactions in fluid solution between the two ends, as it was observed for other bichromophores [22], might explain this discrepancy. The enone presents a symmetry forbidden $n \rightarrow \pi^*$ absorption band near 310 nm of very low intensity, and a $\pi \rightarrow \pi^*$ symmetry allowed band at about 280 nm [23]. The benzenic part displays near 280 nm the classical charge-transfer band, when strong electron donating groups are included in the ring [24]. In methanol, a protic-polar solvent, the $n \rightarrow \pi^*$ absorption band of the enone undergoes a hypsochromic shift (shorter wavelength displacement), whereas the charge transfer band of the benzene chromophore is shifted to longer wavelengths [25]. For these reasons, it appears that the low lying state of tetrahydrocurcuminoids originates from the benzene part of the molecule.

2.1.2. Fluorescence and phosphorescence of THCs

The fluorescence of the tetrahydrocurcuminoids THC1–THC10 has been measured at room temperature in dilute

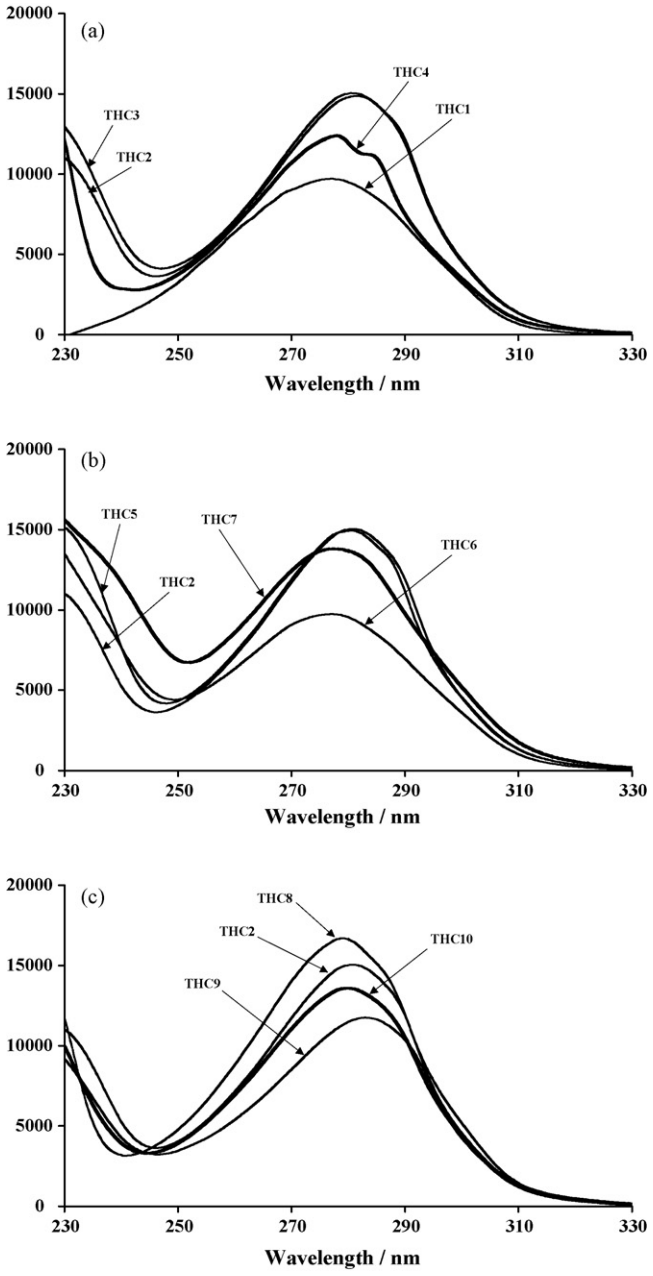


Fig. 1. Absorption spectra of the studied curcuminoids in ethanol solution at room temperature.

ethanol solution (concentration $\approx 10^{-5}$ mol L^{-1}). The emission was found very low, the spectra for compounds giving the most intense emission are reported in Fig. 3.

The fluorescence quantum yields of THCs, at room temperature in ethanol solutions ($\approx 10^{-5}$ mol L^{-1}), were estimated by reference to 9,10-diphenylanthracene [26]; they are reported in Table 1. All tetrahydrocurcuminoids display very low fluorescence emission with quantum yields at about 5×10^{-3}; this is one order of magnitude less than benzenic derivatives [27]. This indicates a strong deactivation of the singlet state by non-radiative processes and maybe by a non-fluorescent exciplex favored by an electron charge transfer between the enolic part as acceptor and the benzenic units as donor. This hypothesis is strengthened by the fluorescence decays (Table 2). They were fitted with a biexponential function with a short component near 2 ns and a longer one near 7 ns. The fluorescence decay of toluene in ethanol was found between 33 and 36 ns [27]. This shows that the fluorescence emission of THCs originates from both benzene rings of the molecules, one being in strong interaction with the enolic unit, the second one less. When electron donating groups are present in the THCs, these interactions become stronger and the lifetimes decrease: $\tau_1 = 2.6$ ns for THC1 *versus* less than 1 ns for THC6 and THC7 which include three donating groups on each benzene unit; $\tau_2 = 18.8$ ns for THC1 versus less than 8.6 ns for all other THCs. It is likely that there is some mixing between the $n\pi^*$ and $\pi\pi^*$ configuration in the low lying singlet state with a predominant $\pi\pi^*$ character.

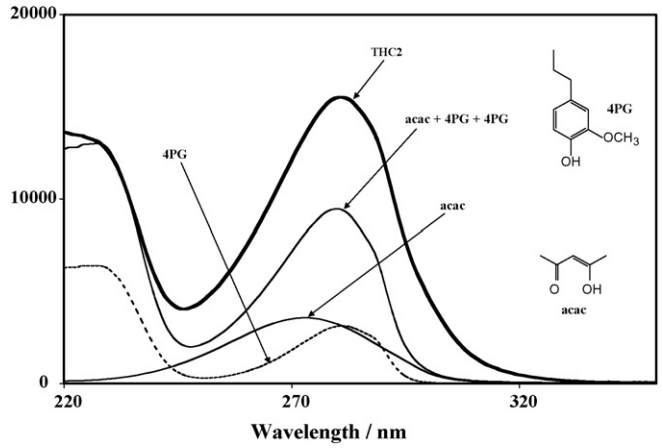


Fig. 2. Electronic absorption spectra of tetrahydrocurcumin, THC2, acetylacetone (enolic form), 4-*n*-propylguaiacol and calculated absorption spectrum of THC2 (ε(acetylacetone) + 2ε(propylguaiacol)). Solvent, methanol; room temperature.

The phosphorescence spectra were recorded at 77 K in ethanol glass. The quantum yields of phosphorescence (Φ_p) were estimated by comparison to the emission of naphthalene measured in the same conditions [28]. The values of Φ_p and some phosphorescence lifetimes are given in Table 1. The phosphorescence emission spectra for the most intense

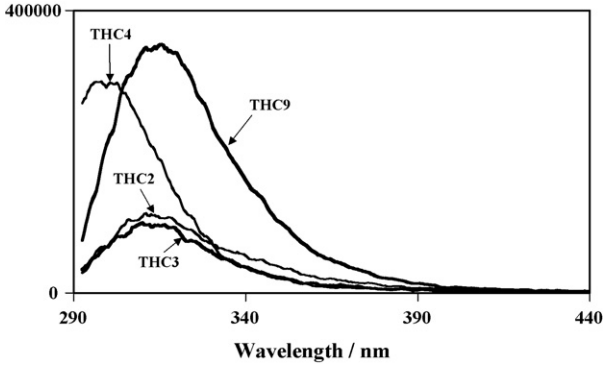


Fig. 3. Fluorescence emission of THC2, THC3, THC4 and THC9 at room temperature in ethanol solution (concentration $\approx 10^{-5}$ mol L^{-1}, $f_{exc} = f_{em} = 2.5$ nm, $\lambda_{exc} = 280$ nm, Ar degassed, corrected, see Section 4).

Table 1
Fluorescence and phosphorescence quantum yields of THCs

Compound	Fluorescence quantum yield RT (Φ_F)[a]	Phosphorescence quantum yield 77 K (Φ_P)[b]	Phosphorescence lifetime 77 K (τ_P) (ms)[c]
THC1	0.9×10^{-3}	0.025	–
THC2	7×10^{-3}	0.26	340
THC3	6×10^{-3}	0.50	400
THC4	9×10^{-3}	0.23	240
THC5	3×10^{-3}	0.06	–
THC6	2×10^{-3}	0.10	–
THC7	3×10^{-3}	0.05	–
THC8	2×10^{-3}	0.10	<20
THC9	13×10^{-3}	0.04	–
THC10	2×10^{-3}	0.035	–

[a] Room temperature; solvent, ethanol; concentration $\approx 10^{-5}$ mol L^{-1}, degassed by argon bubbling; reference, 9,10-diphenylanthracene [26], ±15%.
[b] 77 K; solvent, ethanol; concentration $\approx 10^{-5}$ mol L^{-1}, undegassed; reference, naphthalene [28], ±10%.
[c] ±10%.

Table 2
Fluorescence decays of THCs

Compounds[a]	Fluorescence decay parameters[b]			
	A_1	τ_1 (ns)[c]	A_2	τ_2 (ns)[c]
THC1	0.33	2.6	0.67	18.8
THC2	0.54	1.8	0.46	5.5
THC3	0.74	1.9	0.26	7.7
THC4	0.43	1.1	0.57	7.3
THC5	0.83	1.4	0.17	7.1
THC6	0.87	0.9	0.13	8.0
THC7	0.93	0.3	0.07	7.4
THC8	0.54	2.4	0.46	8.6
THC9	0.78	1.5	0.22	4.0
THC10	0.65	1.2	0.35	6.0

[a] Room temperature; solvent, ethanol; concentration $\approx 10^{-5}$ mol L^{-1}, degassed by argon bubbling.
[b] The fluorescence decays were fitted by the equation $I_F = I_F^0[A_1 \exp(t/\tau_1) + A_2 \exp(t/\tau_2)]$; see Section 4 [29].
[c] ±10%.

emissions (THC2, THC3, THC4 and THC8) are given in Fig. 4.

Compounds THC4 and THC8 present a phosphorescence emission blue shifted by comparison to those of THC2 and THC3. This is in accordance with the presence of additional electron donating methoxy groups in the latter two compounds, lowering the energy of the first triplet state, especially for THC2, which would essentially be of $\pi\pi^*$ character. The presence of some vibronic structure in the three other curcuminoids confirms the existence of some mixing between the $n\pi^*$ and $\pi\pi^*$ configurations in low lying triplet state, due to insufficient electron donating effect of the substituents. There is no simple correlation between Φ_P and the structure of the THCs: nevertheless the tetrahydrocurcumin and tetrahydroisocurcumin THC2 and THC3, respectively, present the highest values, comparable to those found for benzene [30]. The phosphorescence lifetimes measured for the THCs are two times shorter than the one of benzene [28]: the presence of electron donating groups affects strongly the energetic deactivation processes of the triplet state of the benzene rings.

A summary of the absorption (RT) and emission (77 K) of tetrahydrocurcumin THC2 is presented in Fig. 5. The unstructured nature of the bands might originate from some rapid acid–base dissociation process of the phenolic groups in the excited state, the acidity being more important in the excited states [31].

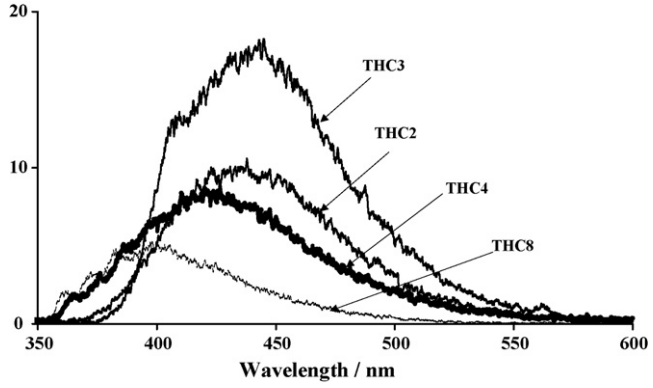


Fig. 4. Phosphorescence emission of THC2, THC3, THC4 and THC9 at 77 K in ethanol glass (concentration $\approx 10^{-5}$ mol L^{-1}, $f_{exc} = f_{em} = 2.5$ nm, $\lambda_{exc} = 280$ nm, undegassed, uncorrected, see Section 4).

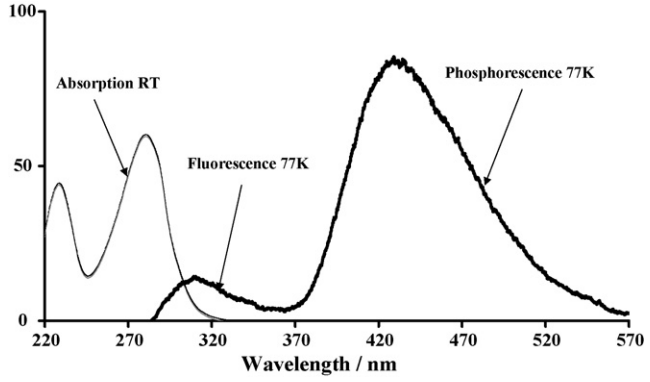


Fig. 5. Absorption (RT), fluorescence and phosphorescence emission of THC2 at 77 K in ethanol glass (concentration $\approx 10^{-5}$ mol L^{-1}, $f_{exc} = f_{em} = 2.5$ nm, $\lambda_{exc} = 280$ nm, undegassed, uncorrected).

2.2. Photochemistry of THCs

2.2.1. Alcoholic solutions

The 10 THCs molecules were irradiated in non-degassed ethanol (concentration near 10^{-4} mol L^{-1}) mainly with a low pressure Hg lamp, which mainly emits at 254 nm and not with wavelengths above 300 nm, to simulate daylight. It was shown previously on lignin [32], that irradiation at 254 nm gave results similar to irradiation with wavelengths filtered by Pyrex glassware, eliminating the short UV component ($\lambda < 300$ nm). The same observation applies to THCs (see further), indicating that there was no reactivity by upper excited states [33]; light energy dissipation by internal conversion to the lowest excited electronic state of the molecule occurred before the photochemical degradation processes started. Fig. 6a and c shows the UV-absorption spectra of THC2 in ethanol solution after irradiation for 0, 15, 30 and 90 min using low and medium pressure Hg lamps, giving emissions at $\lambda = 254$ and $\lambda > 300$ nm, respectively. Using low pressure Hg lamp, we observed a decrease of the band situated at 280 nm and apparition of a new band centered at 320 nm. The same type of observation was made when medium pressure Hg lamp is employed, except that the band centered at 320 nm is much less intense. It is likely that the photoproducts, absorbing between 300 and 380 nm, as exemplified by the difference curves in Fig. 6b and d, are photolyzed by the 313 and 366 nm emission of the medium pressure Hg lamp. The photochemistry appears much more cleaner with the low pressure Hg lamp, because an isobestic point near 305 nm is observed; this is in favor of the transformation of THC2 in a compound having its absorption maximum near 320 nm.

The photochemical behavior of other THCs was also monitored by UV-absorption spectrometry. The phenolic THCs, such as THC3, THC7–THC10, similarly to THC2, display a decrease of the main band situated at 280 nm with the consecutive apparition of a new absorption, centered at 320 nm and an isobestic point near 305 nm. The non-phenolic THCs present comparatively a much lower reactivity. As examples, we have reported in Fig. 7(a–c), the UV spectra obtained after different irradiation times for THC9 and THC5. For THC9, the difference spectrum shown in Fig. 7(b), when comparing to the one of THC2 shown in Fig. 6b, indicates that the same type of photoproducts is formed from the guaiacyl and catechol units. It might be some *ortho*-quinones formed by demethoxylation for THC2 or by the classical oxidation of catechol for THC9, as it was already observed on the photochemistry of lignin model compounds [34]; however, as it will be seen later, the formation of quinone methides is also an other route of evolution of the phenoxy radical which is the primary photoproduct. The low reactivity of non-phenolic THCs is in favor of the photoreactivity of the phenol. Moreover, we have measured the quantum yield of disappearance of the THCs (Table 2). They are compared to their antiradical power efficiency (ARP), which is a measure of their hydrogen transfer to DPPH radical to form, as primary product, a phenoxy radical [21]. The order of photoreactivity and of ARP, expressed in relative units in Table 2, are the same: THC9 > THC7 > THC2 > THC3 > THC10. This is not

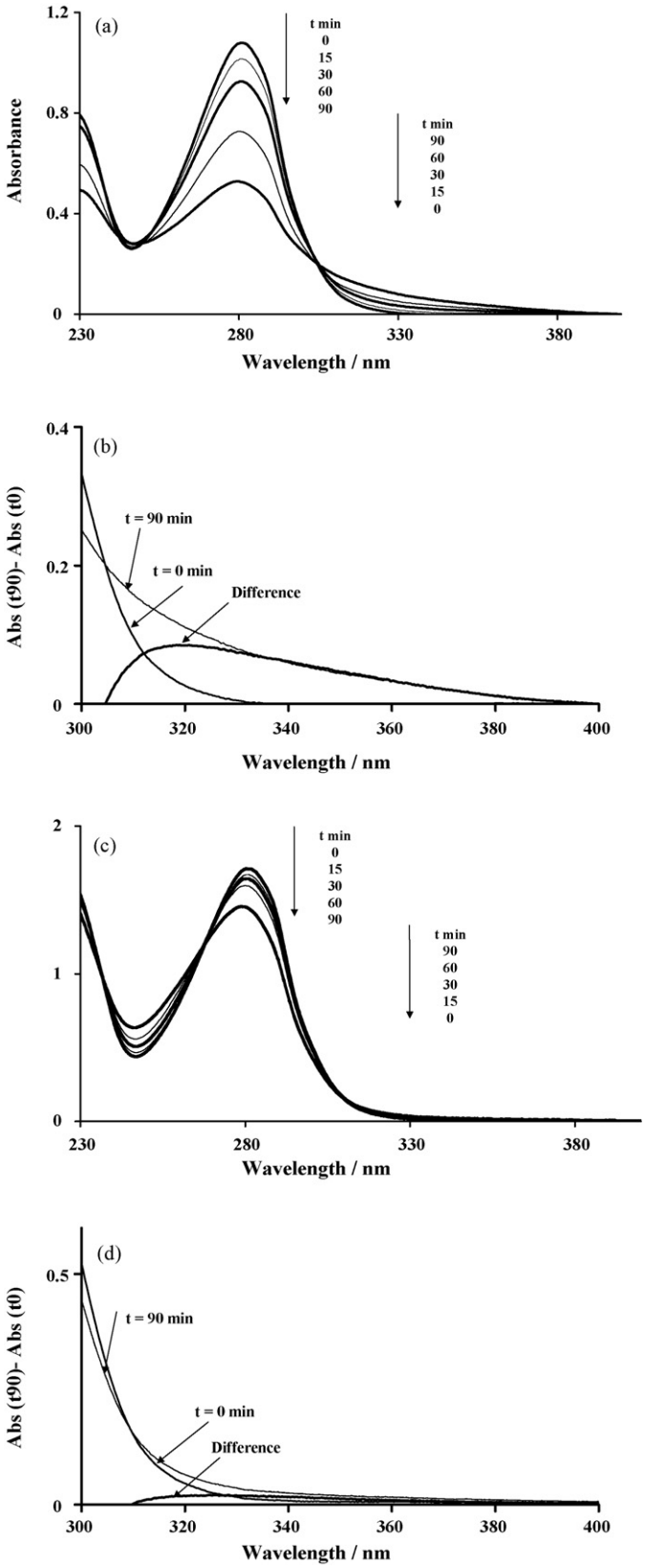


Fig. 6. Absorption spectra of THC2, after irradiation with: (a) low medium pressure Hg lamp ($\lambda \approx 254$ nm); (c) medium pressure Hg lamps ($\lambda > 300$ nm) (see Section 4) in non-degassed ethanol solution (concentration $\approx 10^{-4}$ mol L^{-1}, path length 1 cm, temperature $\approx 25\,^{\circ}C$); (b) difference spectra $t_{90} - t_0$ of curves (a) ($\lambda \approx 254$ nm); (d) difference spectra $t_{90} - t_0$ of curves (c) ($\lambda > 300$ nm).

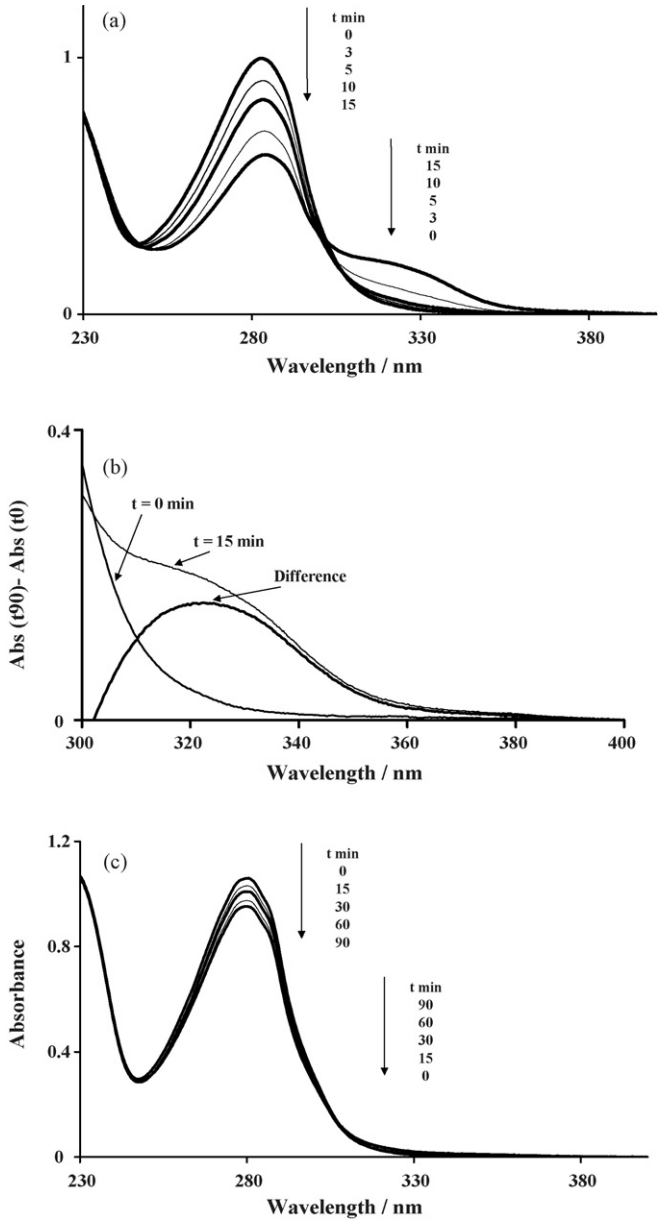


Fig. 7. Absorption spectra of THC9 and THC5 after irradiation with low medium pressure mercury lamp ($\lambda \approx 254$ nm) in non-degassed ethanol solution (concentration $\approx 10^{-4}$ mol L^{-1}, path length 1 cm, temperature $\approx 25\,^{\circ}C$) for different irradiation times: (a) THC9; (b) difference spectra $t_{90} - t_0$ of curves (a); (c) THC5.

surprising, if the formation of the phenoxy radical is the first step of the reaction in both mechanisms, photooxidation and hydrogen transfer to DPPH. It is known that the reducing power of molecules is enhanced when they are in their electronic excited states.

Some reactivity of the non-phenolic tetrahydrocurcuminoids THC1, THC4, THC5 and THC6 remains. For these molecules the presence of a carbonyl group in the chain between the two benzene rings likely induces a cleavage of the chain in α of the carbonyl functional group (Norrish type I reaction). This reaction probably occurs for the phenolic derivatives, but it is not the most important.

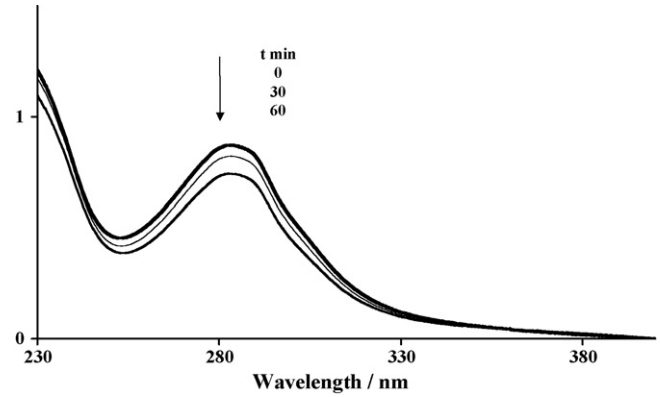


Fig. 8. Absorption spectra of THC2 incorporated in carboxymethylcellulose film after irradiation with low medium pressure mercury lamp ($\lambda \approx 254$ nm) (concentration $\approx 2\%$ (w/w), temperature $\approx 25\,^{\circ}C$) for different irradiation times.

2.2.2. *Carboxymethylcellulose films*

The most photoreactive phenolic THCs were incorporated in a carboxymethylcellulose (CMC) matrix at a concentration near 2% (weight THC/weight CMC). The transparent films (thickness: 36 ± 3 μm) were irradiated in the same conditions as for the ethanol solutions, and the UV-absorption spectra recorded. It was observed that THC9, including catechol groups, was reactive with the matrix and transparent films were not obtained. Molecule THC5, which represents *O*-methylated THC2 or THC3, was also studied. An example of the photoreactivity of phenolic THCs in CMC matrix is given in Fig. 8 for THC2. The relative reactivity quantum yields of the THCs in the films, corresponding to: $(Abs_{t=0} - Abs_{t=3600\,s}) \times 10^5/(Abs_{t=0} \times 3600)$, are reported in Table 4.

In CMC matrix, the compounds are less reactive than in ethanol solution as it can be seen by comparing Figs. 6a and 8 for THC2, because irradiations were conducted under similar light intensity with comparative absorbance of the THCs. The diffusion and lifetime of the reactive intermediates are different in solid state and fluid solution; this should affect their reactivities. As we have observed in solution, some photoreactivity remains in the solid state for THC5, which is representative of

Table 3
Reactivity quantum yields of THCs and antiradical power efficiency (ARP) measured by reaction of the THCs with the DPPH radical [21]

Compounds	Reactivity quantum yield (Φ_R)[a]	Antiradical power (ARP) [21]	Relative ARP
THC1	0.47×10^{-3}	<0.6[b]	–
THC2	1.34×10^{-3}	9.1	1.9
THC3	1.21×10^{-3}	5.3	1.1
THC4	0.23×10^{-3}	<0.6[b]	–
THC5	0.26×10^{-3}	<0.6[b]	–
THC6	0.28×10^{-3}	<0.6[b]	–
THC7	2.56×10^{-3}	11.0	2.3
THC8	0.44×10^{-3}	<0.6[b]	–
THC9	9.44×10^{-3}	15.9	3.3
THC10	1.0×10^{-3}	4.8	1

[a] ±10%.
[b] Value estimated by comparing the ARP of THC2 using EC_{50} and EC_{10}, respectively [21].

Table 4
Relative reactivity quantum yields of THCs incorporated in carboxymethylcellulose films

Compounds	Relative reactivity quantum yields[a] in the CMC films	Relative reactivity quantum yields in ethanol solution[b]
THC2	4.06	4.69
THC3	2.67	4.23
THC5	0.91	0.91
THC7	5.03	8.96
THC8	1.64	1.54
THC10	3.23	3.50

[a] ±10%.
[b] The figures are calculated to have the same values for THC5.

the non-phenolic tetrahydrocurcuminoids. For these molecules, again, the presence of a carbonyl group in the chain between the two benzene rings, likely induces a Norrish type I reaction. Comparison of the figures in Table 3 indicates that tetrahydroisocurcumin (THC3) and THC7 are relatively less reactive in the solid state than in ethanol solution, whereas the other studied THCs present comparative values. The meta position of the phenol for THC3 and the steric hindrance around the phenolic group for THC7 appear to be important factors in the solid state. The absence of the growing band centered at 320 nm, when irradiations are performed for THC2, THC3 and THC7 in CMC films, is another interesting difference with ethanol solutions. This band was assigned to the formation of quinonoid species such as *ortho*-quinones and/or quinone methides. The reactivity of quinonoids structures with cellulosic matrices, as already reported [34], might account for this observation.

2.2.3. Photoproducts

The THCs display two phenolic reactive centers, so their reactivity appears quite complex. To approach the nature of the photoproducts, we have comparatively studied 4-*n*-propylguaiacol (4PG) in methanol solution, this solvent having a lower boiling point than ethanol and so it is much easier to evaporate without altering unstable photoproducts.

Fig. 9a shows the UV-absorption spectra of 4PG in methanol solution after irradiation for 0, 15, 30, 45 and 60 min using low medium pressure Hg lamp. It shows a decrease of the band situated at 280 nm and the apparition of a new band centered at 310 nm. It looks like Fig. 6a related to THC2, the main difference being the increase of the absorption at 254 nm when 4PG is irradiated. The difference curve ($Abs(t_{60}) - Abs(t_0)$) shown in Fig. 9b confirms the similarity of the photochemical behavior of 4PG and THC2.

The disappearance quantum yield of 4PG in undegassed methanol was estimated by comparison with THC2 equal to 0.013, one order of magnitude higher. The influence of the presence of ground state oxygen on reactivity of 4PG in methanol solution (concentration $\approx 4 \times 10^{-4}$ mol $L^{-1)}$ was estimated by bubbling argon or oxygen gas for 20 min and irradiating the solution for 30 min. The relative conversion rates were 0.34/1/1.44 for Ar bubbling, undegassed and O_2 bubbling solutions, respectively. They are 0.31/1/1.06 for THC2 in ethanol solution

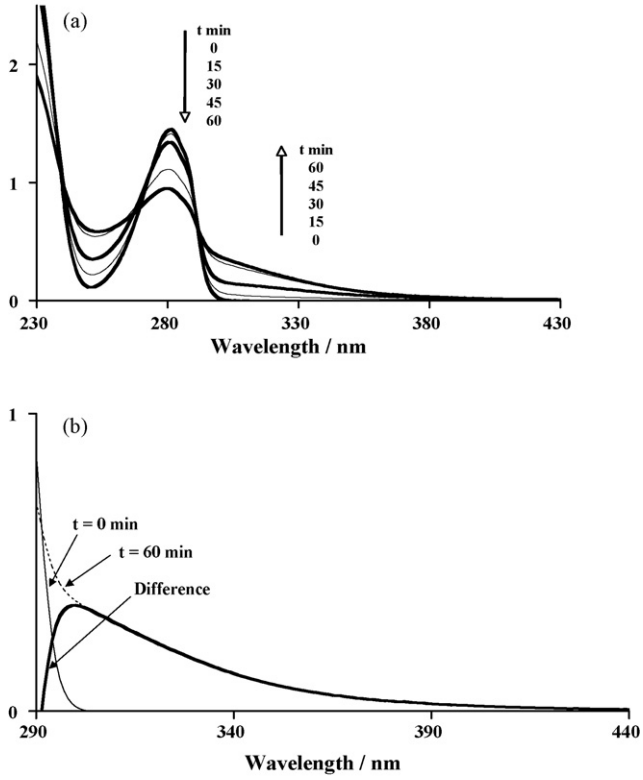


Fig. 9. (a) Absorption spectra of 4-*n*-propylguaiacol, after irradiation with a low medium pressure Hg lamp (λ ≈ 254 nmin non-degassed methanol solution, concentration ≈ 4×10^{-4} mol L^{-1}, path length 1 cm, temperature ≈ 25 °C); (b) difference spectra $t_{60} - t_0$ of curves (a).

(concentration ≈ 10^{-4} mol L^{-1}) after 30 min irradiation. These results indicate that photooxidation of phenol is occurring both for 4PG and THC2. The primary photoproducts are phenoxy radicals generally formed by an electron transfer from the phenol or phenolate anion to oxygen or other oxidant, to generate a cation radical, followed by its deprotonation [37]. Such mechanism is involved in lignin oxidation of lignocellulosics fibers [38] where phenoxy radicals play a central role. The phenoxy radical can evoluate to generate *ortho*-quinone by demethoxylation or dismutate to give the ketonic isomer of 4PG and the corresponding quinone methide, QMPG. The electronic absorption

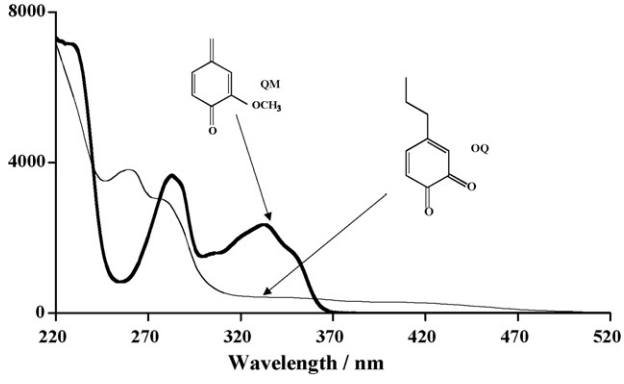
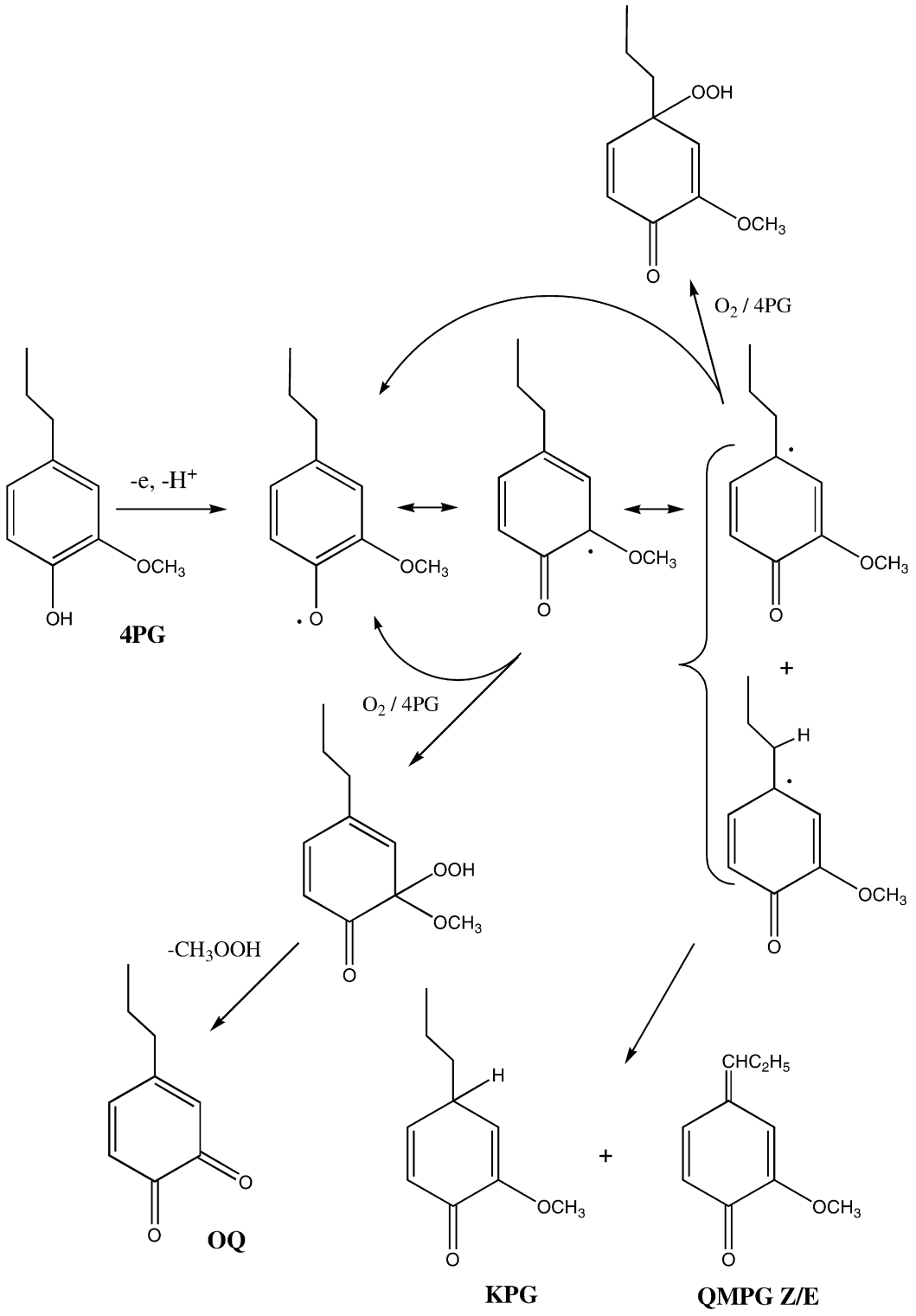


Fig. 10. Absorption spectra of 4-propyl-*ortho*-quinone(OQ) and quinone methide (QM) in methanol solution.



Scheme 2. Proposed photoreactivity of 4-propylguaiacol (4PG) in diluted undegassed alcoholic solution when irradiated with low pressure Hg lamp. It mimics the photoreactivity of tetrahydrocurcuminoids.

spectra of *ortho*-quinone OQ, obtained by oxidation of 4PG with sodium periodate [39], and of quinone methide QM, prepared by dehydrobromination of 4-bromethylguaiacol, are presented in Fig. 10. Compound QM, which does not present stereoisomerism, is easier to prepare in pure form from commercial 4-hydroxymethylguaiacol [40], compared to QMPG obtained, among other compounds, as E/Z isomers by silver oxide oxidation of 4PG [41,42]. Comparison of Figs. 9b and 10 shows that photoirradiation of 4PG in undegassed methanol gives quinone methide structures (QMPG) with some *ortho*-quinone (OQ). The photogeneration of quinone methides [43] and *ortho*-quinones [44] from phenols has been already observed. It was also shown

that they are photobleached [44,45]; this will explain the wavelength effect already observed when medium Hg lamp is used.

HPLC analysis on C18 silica reverse phase of the irradiated solution (10^{-4} mol L^{-1}, low pressure Hg lamp) after concentration of methanol at 20 °C under vacuum, shows the presence of the two isomers of QMPG but does not allow the detection of OQ which presents the same retention time as 4PG. Analysis by GC–MS shows the presence of two close peaks with the same mass spectrum, one due to the starting compound 4PG, the other one might be the ketonic isomer of 4PG (KPG). It has not been possible to identify the hydroperoxide formed by oxygen addition to the carbon 1 centered radical followed by hydrogen abstraction on 4PG, as it was observed on the photochemistry of other phenols [46,47]. Scheme 2 summarizes the photoreactivity of 4PG. The photochemical behavior of 4PG might give some indication on the photoreactivity of tetrahydrocurcuminoids.

3. Conclusion

The photophysical and photochemical properties of 10 tetrahydrocurcuminoids bearing various hydroxy and methoxy groups on their benzene subunits have been studied for the first time. They display very low fluorescence in ethanol solution at room temperature with a quantum yields between 0.9 and 13×10^{-3}. Their phosphorescence emission in ethanol glass at 77 K is more intense with quantum yields between 0.025 and 0.5. These figures depend on the balance between the $n\pi^*$ and $\pi\pi^*$ nature of their singlet and triplet excited states. The photochemical behavior of THCs was found closely related to their antioxidant properties. Their disappearance quantum yields measured in solution, between 5×10^{-4} and 10^{-2}, follows the same trend as their antiradical power efficiency. In general THCs are much more reactive, if they include a phenol group in meta or para of the linking chain and a phenol or methoxy group as neighbor. The characteristic UV absorption of the phenolic derivatives, irradiated in solution with the low pressure Hg lamp, emitting light at $\lambda = 254$ nm, and molecular analysis of the photoproducts of 4-*n*-propylguaiacol, chosen as simplest model of phenolic THCs, indicate the formation of quinone methides and in a less extent of *ortho*-quinone, obtained by demethoxylation. The residual reactivity of the non-phenolic THCs is probably due to Norrish type I cleavage of the linking chain of the bichromophores. The irradiation of the THCs in CMC films follows the same conclusions as the solution studies, except that quinone methides and *ortho*-quinone are not detected, probably because they are photobleached in the cellulosic matrix. The knowledge obtained in this study probably will help to better understand the complex mechanism of photodegradation of natural polymers such as lignin and also provide better understanding to promote the use of THCs in various applications where it is necessary to develop antioxidant properties, even in presence of solar light.

4. Experimental

The syntheses of the tetrahydrocurcuminoids THC1–THC10 have been reported previously [21]. The solvents (spectroscopic grade) were obtained from Aldrich and were used as received.

UV-absorption spectra were recorded on a Shimadzu 2501 PC spectrometer. Fluorescence spectra were measured with a Hitachi F4500 apparatus and a SPEX Fluorolog fluorimeter at room temperature (≈25 °C); the exciting wavelength was set at 280 nm and the slits on the excitation and emission beams were fixed at 2.5 nm bandwidth. The fluorescence emission spectra recorded at room temperature were corrected for instrumental response. The fluorescence quantum yields of THCs in ethanol solution were determined by reference to 9,10-diphenylanthracene in cyclohexane with appropriate correction for difference of refractive index of both solvents [26]. Absorbance of solutions was adjusted near 0.1 to avoid re-absorption of the emitted light. In these conditions, the fraction of absorbed light is very close to the absorbance. Areas under the fluorescence curve were calculated using the software of the fluorimeter. The emission spectra were corrected of the solvent Raman emission contribution. The solutions were degassed by argon bubbling before measurements. Phosphorescence measurements were made using the phosphorescence accessories of the Hitachi F4500 fluorimeter including appropriate quartz Dewar and quartz tube of 5 mm internal diameter. The solutions were frozen slowly in liquid nitrogen until they form a glass. The solutions used for fluorescence were also employed for phosphorescence, so the absorbance values taken to calculate fluorescence quantum yields were also used for phosphorescence quantum yield determinations. The phosphorescence spectra shown in Fig. 4 were obtained using the phosphorescence mode of the spectrometer, but for phosphorescence quantum yield determination, fluorescence mode was used to obtain the total spectrum, as shown for THC2 in Fig. 5. Only the area under the phosphorescence part, determined by the measure in the phosphorescence mode of the spectrometer (see Fig. 4), was considered. Naphthalene was used as standard [27,28].

Fluorescence decay lifetimes were measured by single photon counting Applied Photophysics apparatus as already depicted [48,49]. The fluorescence decays ($\lambda_{em} = 320$ nm) were analyzed using the DECAN program, kindly given by Pr. F.C. De Schryver (University of Leuven, Belgium), and fitted by the equation $I_F = I_F^0[A_1 \exp(t/\tau_1) + A_2 \exp(t/\tau_2)]$. The quality of the fit was estimated by inspection of the residues and by the values of χ^2 (between 1.02 and 1.1) and those of the Durbin–Watson test (between 1.85 and 1.95) [29].

Photodegradation of THCs in ethanol solutions and CMC films (solid state, see further) were monitored by UV-absorption spectroscopy at different times. The solutions were irradiated in 10 mm quartz cuvettes and the films. The irradiations were performed for all THCs using the 254 nm emission, given by low pressure mercury lamp (Vilbert-Lourmat T-8C for TLC detection), or for THC2 in ethanol solution, light with wavelengths longer than 300 nm given by a setup, which includes two parallel medium pressure mercury lamps (400 W), a fan directed towards the sample to maintain temperature at about 30 °C and a Pyrex container surrounding the sample cell, to eliminate wavelengths below 300 nm. The quartz cell was situated

at 6 cm from the low pressure mercury lamp. For the disappearance quantum yields in ethanol solutions, THCs concentration (near 10^{-4} mol L^{-1}) before and after irradiation was accurately determined using the molar extinction coefficient maximum of the 280 nm band in alcoholic solutions [21]. The photon flux was estimated by measuring, in the same conditions, the disappearance of an ethanol solution of anthracene (10^{-3} mol L^{-1}) degassed by argon bubbling. The conversion rate limited to 10%, as for THCs, was transformed in photodimerization rate by dividing by two. An intensity of 3.9×10^{15} photons cm^{-2} s^{-1} was obtained, using a value of $\Phi_{dim} = 10^{-2}$ at 10^{-3} mol L^{-1} [35,36] for photodimerization of anthracene; this corresponds to the high intensity domain considered by Charlton et al. [35].

Carboxymethylcellulose (CMC) films were prepared by mixing at about 5 mg, accurately weighted and dissolved in tetrahydrofuran (4 mL), with 15 g of a solution of CMC (1.5 g) in water (75 mL). After degassing by sonication (30 s), the CMC solution was placed in a Pyrex cylindrical container of 6.5 cm internal diameter, and placed in an aerated cabin for 48 h at room temperature. The transparent films obtained were easily removable from the Pyrex glass container. Their thickness was evaluated with a Carl-Zeiss microscope and found equal to 36 ± 3 μm. The films were cut in rectangular pieces (4 cm × 2 cm) and placed in a black cardboard mask presenting a rectangular open window (3 cm × 1.5 cm) which correspond to the irradiated surface and to the zone of absorbance measurements. The absorbance of films was normalized to zero at 400 nm.

4-Propylguaiacol (4PG), obtained by hydrogenation of eugenol [21], in undegassed methanol solution ($\approx 4 \times 10^{-4}$ mol L^{-1}) was irradiated in a quartz cuvette (1 cm path length) by the low pressure Hg lamp for 45 min. The solution is concentrated at 25 °C under vacuum and analyzed either by GC–MS or HPLC. GC–MS analysis was performed with a Finnigan Trace mass spectrometer interfaced with a Finnigan Trace GC Ultra gas apparatus (line transfer temperature, 250 °C) equipped with a PTV injector (splitless mode) using Helium as carrier gas. A fused silica capillary RTX-5MS column, 15 m, Ø 0.25 mm i.d., film thickness 0.25 μm was selected. The oven temperature was programmed from 40 °C (initial hold time of 1 min) to 320 °C at a rate of 15 °C min^{-1}; this final temperature was maintained for 15 min. The electron energy was fixed at 70 eV. The analytical HPLC used a Thermo Separation Product line including: a pump type SP1000, an automatic injector AS 3000 and a UV detector AS 2000 and a reverse phase column type Lichrospher 100 Å RP18 5 μm (250 mm × 4.6 mm) using a mixture of methanol/water (2/8, v/v) as eluent. As probable photoproducts, the *ortho*-quinone (OQ) was prepared according to reference [39] and the Z/E quinone methides (QMPG) were prepared by oxidation by Ag_2O of 4PG in dichloromethane at room temperature [41] followed by rapid filtration over silica gel and evaporation of the reaction mixture. A pure quinone methide compound (QM) has been prepared following the procedure described by Brunow et al. [40] starting from the corresponding benzyl alcohol.

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References

[1] H.H. Toennesen, J. Karlsen, J. Chromatogr. 259 (1983) 367–371.
[2] V.S. Govindarajan, CRC Crit. Rev. Food Sci. 12 (1979) 199–301.
[3] H.H. Toennesen, Thesis, University of Oslo, Norway, 1986.
[4] M. Ali, A. Bagati, J. Gupta, Indian J. Chem. 34B (1995) 884–888.
[5] Z. Sui, R. Salto, J. Li, C. Craik, P.R.O. De Montellano, Bioorg. Med. Chem. 1 (1993) 415–422.
[6] B.B. Aggarwal, A. Kumar, M.S. Aggarwal, S. Shishodia, in: D. Bagchi, H.G. Preuss (Eds.), Phytopharmaceuticals in Cancer Chemoprotection, CRC Press, Boca Raton, 2005, pp. 349–387.
[7] E. Skrzypczak-Jankun, N.P. McCabe, S.H. Selman, J. Jankun, Int. J. Mol. Med. 6 (2000) 521–526.
[8] M.T. Huang, T. Lysz, T. Ferraro, T.F. Abidi, J.D. Laskin, A.H. Conney, Cancer Res. 51 (1991) 813–819.
[9] S.C. Sharma, H. Mukhtar, S.K. Sharma, C.R. Krishna Murti, Biochem. Pharmacol. 21 (1972) 1210–1214.
[10] O.P. Sharma, Biochem. Pharmacol. 25 (1976) 1811–1812.
[11] P. Scartezzini, E. Speroni, J. Ethnopharmacol. 71 (71) (2000) 23–43.
[12] M. Majeed, V. Badmaev, U. Shimakumar, R. Rajendran, Curcuminoids: Antioxidant Phytonutrients, Nutriscience Publishers Inc., Piscataway, 1990.
[13] M.H. Pan, T.M. Huang, J.K. Lin, Drug Metab. Dispos. 27 (1999) 486–494.
[14] T. Osawa, Y. Sugiyama, M. Inayoshi, S. Kawakishi, Biosci. Biotechnol. Biochem. 59 (1995) 1609–1612.
[15] Y. Sugiyama, S. Kawakishi, T. Osawa, Biochem. Pharmacol. 52 (1996) 519–525.
[16] Y. Nakamura, Y. Ohto, A. Murakami, T. Osawa, H. Ohigashi, Jpn. J. Cancer Res. 89 (1998) 361–370.
[17] J.M. Kim, S. Araki, D.J. Kim, C.B. Park, N. Takasuka, H. Baba-Toriyama, T. Ota, Z. Nir, F. Khachika, N. Shimidzu, Y. Tanaka, T. Ozawa, T. Uraji, M. Murakoshi, H. Nishino, H. Tsuda, Carcinogenesis 19 (1998) 81–85.
[18] A. Sundaryono, A. Nourmamode, C. Gardrat, S. Grelier, O. Despeyroux, A. Castellan, Holz Roh Werkst. 61 (2003) 377–381.
[19] A. Sundaryono, A. Nourmamode, C. Gardrat, S. Grelier, G. Bravic, D. Chasseau, A. Castellan, Photochem. Photobiol. Sci. 2 (2003) 914–920.
[20] K.I. Priyadarsini, D.K. Maity, G.H. Naik, M.S. Kumar, M.K. Unnikrishnan, J.G. Satav, H. Mohan, Free Radical Biol. Med. 35 (2003) 475–484.
[21] E. Portes, C. Gardrat, A. Castellan, submitted for publication.
[22] A. Castellan, J.P. Desvergne, J.P. Bideau, G. Bravic, C. Courseille, Mol. Cryst. Liq. Cryst. 93 (1983) 103–112.
[23] R.M. Silverstein, G.C. Bassler, T.C. Morill, Spectrometric Identification of Organic Compounds, J. Wiley and Sons, New York, 1991.
[24] A. Castellan, J. Michl, J. Am. Chem. Soc. l00 (1978) 6824–6827.
[25] C. Reichardt, Angew. Chem. Int. Ed. 18 (1979) 98–110.
[26] S. Hamal, F. Hirayama, J. Phys. Chem. 87 (1983) 83–89.
[27] J.B. Birks, Photophysics of Aromatic Molecules, Wiley Interscience, New York, 1970, p. 126.
[28] H. Aagren, B.F. Minaev, S. Knuts, J. Phys. Chem. 98 (1994) 3943–3949.
[29] D.V. O'Connor, D. Phillips, Time-Correlated Single Photon Counting, Academic Press, London, 1984.
[30] N.J. Turro, Modern Molecular Photochemistry, University Science Books, Sausalito, CA, USA, 1991.
[31] B. Valeur, Molecular Fluorescence: Principle and Applications, Wiley-VCH, New York, 2001.

[32] R. Ruggiero, A.E.H. Machado, C. Gardrat, W. Hoareau, S. Grelier, B. Siegmund, A. Castellan, J. Photochem. Photobiol. A: Chem. 173 (2005) 150–155.
[33] A. Castellan, J. Kolc, J. Michl, J. Am. Chem. Soc. l00 (1978) 6687–6692.
[34] A. Castellan, A. Nourmamode, C. Jaeger, I. Forsskåhl, in: C. Heitner, J.C. Scaiano (Eds.), Photochemistry of Lignocellulosic Materials, ACS Symposium Series, vol. 531, 1993, pp. 60–76.
[35] J.L. Charlton, R. Dabestani, J. Saltiel, J. Am. Chem. Soc. 105 (1983) 3473–3476.
[36] A. Castellan, C. R. Acad. Sci. 28lC (1975) 221–224.
[37] H.I. Joschek, S.I. Miller, J. Am. Chem. Soc. 88 (1966) 3273–3281.
[38] A. Castellan, N. Colombo, A. Nourmamode, J.H. Zhu, D. Lachenal, R.S. Davidson, L. Dunn, J. Wood Chem. Technol. 10 (1990) 461–493.
[39] W.G. Trindade, W. Hoareau, I.A.T. Razera, R. Ruggiero, E. Frollini, A. Castellan, Macromol. Mater. Eng. 289 (2004) 728–736.
[40] G. Brunow, J. Sipilä, T. Mäkelä, Holzforschung 43 (1989) 55–59.
[41] S.M. Schevchenko, A.G. Anushkinskii, V.A. Gindin, M.Ya. Zarubin, Zh. Org. Khim. 26 (1990) 1069–1075.
[42] S.M. Schevchenko, S.G. Semenov, A.G. Anushkinskii, A.V. Pranovich, E.I. Evstigneev, Zh. Org. Khim. 26 (1990) 1839–1848.
[43] L. Diao, C. Yang, P. Wan, J. Am. Chem. Soc. 117 (1995) 5369–5370.
[44] D.S. Argyropoulos, C. Heitner, J.A. Schmidt, Res. Chem. Intermed. 21 (1995) 263–274.
[45] J. Dolenc, B. Sket, M. Strlic, Tetrahedron Lett. 43 (2002) 5669–5671.
[46] T. Matsuura, N. Yoshimura, A. Nishinaga, I. Saito, Tetrahedron Lett. 21 (1969) 1669–1671.
[47] M. Prein, M. Maurer, E.M. Peters, K. Peters, H.G. Von Schnering, W. Adam, Chem. Eur. J. 1 (1995) 89–94.
[48] A. Castellan, J.P. Desvergne, R. Lesclaux, J.C. Soulignac, Chem. Phys. Lett. 106 (1984) 117–123.
[49] J.P. Desvergne, N. Bitit, A. Castellan, M. Webb, H. Bouas-Laurent, J. Chem. Soc., Perkin Trans. II (1988) 1885–1894.