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Phototransformation of the drug rivastigmine: Photoinduced cleavage of benzyl-nitrogen sigma bond

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ABSTRACT

The photochemical behaviour of rivastigmine has been investigated under UV-B irradiation by HPLC and NMR. Kinetic data have been obtained irradiating aqueous solutions (5×10^{-5} M). For mechanistic purposes and products studies irradiation has also been carried out on concentrated solutions (1×10^{-3} M) in water/acetonitrile, acetonitrile, methanol, methanol/acetonitrile, in the presence and absence of oxygen. Photoproducts, isolated by chromatographic processes, have been identified by spectroscopic means. Photochemical cleavage of the benzyl-nitrogen bond occurs and leads to radical-derived and, mainly, ion-derived products. Mechanistic pathways have been hypothesised on the basis of both steady-state irradiation and laser flash photolysis experiments.

1. Introduction

The behaviour of a substance under irradiation is a concern of increasing attention in various scientific fields from chemistry to biochemistry, from engineering to environmental sciences. Photochemical transformations may be usefully employed in organic synthesis, often avoiding drastic conditions [1], or may represent an environmental problem [2]. Indeed a substance introduced into the environment can convert to products that may be persistent or noxious, sometimes even more than the parent compound. Sunlight combined with oxygen, and/or natural or synthetic sensitizers, is one of the main factors for these transformations. In this context, our researches are addressed to investigate the photochemistry of bioactive compounds, especially drugs [3] that represent emerging environmental contaminants [4,5]. The interest for drugs is also due to the photobiological risk associated with some of them [6]. Given the complexity and heterogeneity of this broad class of compounds, it is often difficult to predict or rationalize the photochemical behaviour of drugs. Moreover the knowledge of factors (substituents, solvents, light) that affect the photochemistry of organic compounds are generally limited to relatively simple molecules, in organic solvents and irradiation at 254 nm. Hence, extension of photochemical investigation to bioactive compounds is useful to evaluate their photobiological risk and also to confirm known photochemical processes or, sometime, to evidence new photoinduced routes [3,7]. Here we analyse the photochemical behaviour of rivastigmine, (*S*)-3-[(1-dimethylamino)ethyl]phenyl N-ethyl-N-methyl carbamate, an inhibitor of acetylcholinesterase that is used for the treatment of mild to moderate Alzheimer's disease in adults [8]. Introduced in EU in 1998 and in USA in 2000, rivastigmine is relatively a new drug, and attention has been generally focused to analytical methods for its detection [9] or determination of optical purity [10].

Previous studies have examined the behaviour of rivastigmine hydrogen tartrate bulk drug under forced degradation conditions [0.5 N hydrochloric acid, 0.5 N sodium hydroxide, 3% hydrogen peroxide, heat (60 °C), and UV light (254 nm)] [11]. Rivastigmine degradation was observed only under basic conditions yielding, after 48 h, trace amount of (*S*)-3-(1-dimethylaminoethyl) phenol, which is also the major human metabolite known as NAP 226-90 [11].

The aim of this work is to explore the behaviour of free rivastigmine with particular attention to mimic environmental conditions (water, $\lambda > 300$ nm). Various steady state irradiation and laser flash photolysis experiments have been carried out in order to understand the mechanisms leading to its photodegradation.

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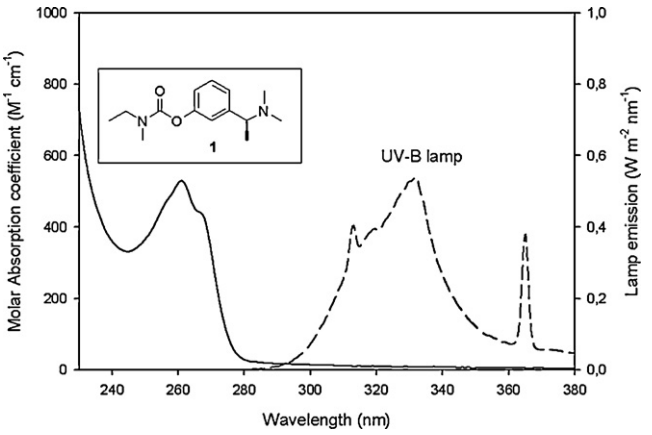

Fig. 1. Molar absorption coefficient of rivastigmine (**1**) aqueous solution (solid line) and emission spectrum of radiation reaching the solution inside the pyrex reactor (dashed line).

2. Materials and methods

2.1. Chemicals

Rivastigmine tartrate was purchased by Kemprotec, analytical standard grade (99%). *p*-Nitroanisole (PNA) and 9,10-anthraquinone were supplied by Aldrich and used without further purification. Milli-Q water (Millipore) was used to prepare aqueous solutions. Acetonitrile and methanol were of HPLC grade (Sigma-Aldrich). Rivastigmine (**1**, Fig. 1) was obtained by dissolving the salt in $NaHCO_3$ (sat.) and extracting with ethyl acetate.

2.2. Apparatus

HPLC experiments were carried out on an Agilent 1100 HPLC system, equipped with an UV detector, using a RP-18 column (Gemini, 5 μm, 110 A, 250 mm × 4.6 mm). The flow was set to 0.8 mL/min. The detector lamp was set at 254 nm. The column was equilibrated with a mixture of A (H_2O containing 1% formic acid)–B (acetonitrile) 9:1 (v/v). The run followed this programme: isocratic B 10% from the start to 7 min, an increase of B up to 90% from 7 to 12 min, isocratic B 90% for 11 min, return to B 10% in 2 min.

Analytical and preparative TLC were made on Kieselgel 60 F_{254} plates with 0.2 mm and 0.5 or 1 mm layer thickness, respectively (Merck).

NMR spectra were recorded on a Varian Inova-500 instrument operating at 499.6 and 125.6 MHz for ^1H and ^{13}C, respectively, and referenced with deuterated solvents ($CDCl_3$).

Electronic impact mass spectra (EI-MS) were obtained with a GC–MS QP5050A (Shimadzu) equipped with a 70 eV EI detector.

IR spectra were recorded on a Jasco FT/IR-430 instrument equipped with single reflection ATR using $CHCl_3$ as solvent.

UV–Vis spectra were recorded with a Varian Cary 300 UV–Vis spectrophotometer.

A Varian Cary Eclipse fluorescence spectrofluorimeter was used, adopting a 5 nm bandpass on both excitation and emission.

2.3. Steady-state irradiation experiments

2.3.1. Irradiation in water

Rivastigmine water solutions (5×10^{-5} M) were irradiated in a photochemical setup (system I). System I consisted of a thermostated pyrex cylindrical reactor (total volume 100 mL) surrounded by six Duke GL20E 20 W lamps. The temperature was held at 293 ± 2 K. Emission spectrum reaching solution (Fig. 1) was measured with an Ocean Optics SD 2000 CCD spectrophotometer (calibrated using a DH-2000-CAL Deuterium Tungsten Halogen reference lamp) and normalized to the actinometry results using PNA/pyridine actinometer [12]. The incident photon flux in solution was 6.10×10^{19} photons m^{-2} s^{-1}.

2.3.1.1. Phototransformation kinetics and quantum yield calculation. Rivastigmine solutions (5×10^{-5} M) were irradiated as above. At fixed time intervals an aliquot (200 μL) was withdrawn and analysed by HPLC. The time evolution of rivastigmine could be fitted with a pseudo-first order equation $C_0 = C_t \exp(-kt)$ where C_0 was the initial rivastigmine concentration, C_t the concentration at time t and k the pseudo-first order degradation rate constant.

The kinetic constant calculated is $k = (1.08 \pm 0.02) \times 10^{-6}$ s^{-1} and the half-life time is $t_{1/2}$ = 177.5 h. Assuming rivastigmine as the only absorbing species present in water, the polychromatic quantum yield degradation (ϕ) of the drug was calculated, in the overlap range 288–320 nm with lamp emission (see Fig. 1), as follows:

$$\phi = \frac{R_{riv}}{I_a}$$

where R_{riv} is the rivastigmine degradation rate (M s^{-1}) and I_a is the absorbed photon flux per unit of surface and unit of time. The latter was calculated from:

$$I_a = \int_{\lambda_1}^{\lambda_2} I_0(\lambda)(1 - 10^{-\varepsilon(\lambda)d[Riv]})$$

where I_0 is the incident photon flux, ε the molar absorption coefficient of rivastigmine, d the optical path length inside the cells and [*Riv*] the initial rivastigmine concentration.

Polychromatic quantum yield ϕ_{riv} is 2.61×10^{-3}.

2.3.2. Sunlight irradiation of rivastigmine

A solution of rivastigmine in water (1×10^{-5} M) was exposed to sunlight in a closed pyrex tube in September–October 2010 in Naples and analysed by HPLC after 6, 12 and 20 days.

2.3.3. Irradiation experiments for mechanistic and preparative purposes

Irradiations were performed in quartz tubes (20 cm × 1 cm, 25 mL) by means of a Helios Italquartz photoreactor (system II) equipped with six 15 W UV-B lamps with a maximal output at ca. 310 nm (1 mW cm^{-2}) or four 15 W UV-A lamps with a maximal output at ca. 366 nm (1 mW cm^{-2}).

2.3.3.1. Irradiation in different solvents. Four 1×10^{-3} M solutions of rivastigmine in H_2O/CH_3CN (9:1, v/v), CH_3CN, CH_3OH, CH_3OH/CH_3CN (9:1, v/v) were prepared by dissolving 5 mg in 20 mL. Each solution was irradiated in open quartz tubes with UV-B lamps and analysed by HPLC and ^1H NMR at selected times. Similar irradiation experiments were performed as above in closed quartz tubes after saturating with argon.

A further series of solutions treated as above were irradiated for 1 h. Then, the solvents were evaporated and each residue was carefully analysed by ^1H NMR. The photoproducts in each mixture were identified by comparing the NMR signals with those of standard compounds, which were isolated and characterized by performing preparative photochemical experiments (see Section 2.3.3.3, Fig. 2, Table 2 and Supporting Content).

All the experiments were performed in triplicate.

Samples of all solutions were kept in the dark and analysed as above showing no degradation after 48 h.

2.3.3.2. Photoproducts isolation. A solution of rivastigmine (32 mg) was dissolved in 128 mL of Milli-Q H_2O/CH_3CN (9:1, v/v, 1×10^{-3} M) and irradiated in open tubes with UV-B lamps for

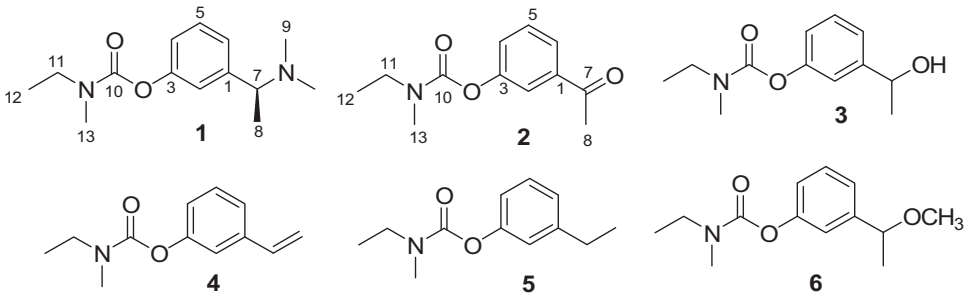

Fig. 2. Rivastigmine and its photoproducts.

5 h. Then, the solvents were evaporated under vacuum and the residue (20 mg) was separated by preparative TLC. Elution with CH_2Cl_2/AcOEt (8:2, v/v) gave a mixture of **4** and **5** (<1 mg), ketone **2** (4 mg), alcohol **3** (7 mg), an intractable material (5 mg) and rivastigmine (2 mg) at decreasing Rfs (Fig. 2).

Rivastigmine (15 mg) dissolved in 60 mL of acetonitrile (1×10^{-3} M) was exposed in open tubes to UV-B lamps for 1 h. After evaporation of the solvent, preparative TLC of the residue as above gave ketone **2** (10 mg) and the drug (1 mg).

A solution of rivastigmine (15 mg) in methanol (60 mL, 1×10^{-3} M) was saturated with argon and irradiated in closed tubes to UV-B lamps for 5 h. After evaporation of the solvent, preparative TLC of the residue as above gave a fraction (1 mg) composed of **4** and **5** (in ca. 1:1 molar ratio), ether **6** (3 mg) and rivastigmine (9 mg) (Fig. 2).

Products **2**, **3** and **6** were fully characterized. Alkene **4** and alkane **5** were recovered as a 1:1 mixture in small amounts, therefore only ^{1}H NMR data could be obtained and refer to this mixture.

*2.3.3.3. Spectroscopic data of photoproducts **2–6**.* 3-Acetylphenyl N-ethyl-N-methyl carbamate (**2**): UV λ_{max} (CH_3OH) nm: 282 (log ε 3.2); EI-MS *m/z* 221, 177, 86, 58; ν_{max} ($CHCl_3$) 1717, 1693, 1607, 1406 and 1168 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 1.

3-(1-Hydroxyethyl)phenyl N-ethyl-N-methyl carbamate (**3**): UV λ_{max} (CH_3OH) nm 252 (log ε 2.9); EI-MS *m/z* 223, 86, 77, 58; ν_{max} ($CHCl_3$) 3626, 3501, 1716, 1600, 1172 and 1014 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 1.

3-Vinylphenyl N-ethyl-N-methyl carbamate (**4**) and 3-ethylphenyl N-ethyl-N-methyl carbamate (**5**): (1:1 mixture); selected ^{1}H NMR signals for alkene **4**: δ_H ($CDCl_3$) 7.30 (1H, t, *J* = 7.8 Hz, H-5), 7.22 (1H, d, *J* = 7.8 Hz, H-6), 7.16 (1H, brs, H-2), 6.68 (1H, dd, *J* = 17.6, 11.0 Hz, H-7), 5.74 and 5.26 (2H, d, *J* = 17.6 Hz and *J* = 11.0 Hz, H-8); selected ^{1}H NMR signals for alkane **5**: δ 7.25 (1H, t, *J* = 8.0 Hz, H-5), 7.02 (1H, d, *J* = 8.0 Hz, H-6), 6.95 (1H, brs, H-2), 2.65 (2H, q, *J* = 7.0 Hz, H-7), 1.23 (3H, t, *J* = 8.0 Hz, H-8); overlapping ^{1}H NMR signals: δ_H 6.92 (2H, brs, H-4), 3.47 and 3.40 (4H, 2q, *J* = 6.0 Hz, H-11), 3.06 and 2.99 (6H, 2 s, H-13), 1.22 and 1.17 (6H, 2t, *J* = 6.0 Hz, H-12).

3-(1-methoxyethyl)phenyl N-ethyl-N-methyl carbamate (**6**): UV λ_{max} (CH_3OH) nm: 261 (log ε 2.7); EI-MS *m/z* 237, 236, 206, 86, 58; ν_{max} ($CHCl_3$) 1715, 1608, 1236 and 1168 cm^{-1}; ^{1}H and ^{13}C NMR, see Table 1.

2.3.3.4. Irradiation experiments with sensitizers. Three solutions of rivastigmine (5 mg) in 100 mL of H_2O/CH_3CN (9:1, v/v, 5×10^{-4} M) in the presence of 0.3, 0.5, 0.7 equiv. of ketone **2** were irradiated in closed quartz tubes with UV-B lamps and analysed by HPLC.

Two solutions of rivastigmine (5 mg) in 100 mL of H_2O/CH_3CN (1:1, v/v, 5×10^{-4} M) with and without 0.5 equiv. of 9,10-anthraquinone were irradiated in closed quartz tubes with UV-A lamps (centred at λ 360 nm) and analysed by HPLC.

2.4. Laser flash photolysis studies

For 266/355 nm excitation, experiments were carried out using the fourth/third harmonic of a Quanta Ray GCR 130-01 Nd:YAG laser system instrument, used in a right-angle geometry with respect to the monitoring light beam. The single pulses were ca. 9 ns in duration, with energy of ~60 mJ/pulse. Individual cuvette samples (*V* = 3 mL) were used for a maximum of four consecutive laser shots. The transient absorbance at the pre-selected wavelength was monitored by a detection system consisting of a pulsed xenon lamp (150 W), monochromator and a photomultiplier (1P28). A spectrometer control unit was used for synchronising the pulsed light source and programmable shutters with the laser output. The signal from the photomultiplier was digitised by a programmable digital oscilloscope (HP54522A). A 32 bits RISC-processor kinetic spectrometer workstation was used to analyse the digitised signal. The pseudo-first order decay constants of transient species were obtained by fitting the absorbance vs. time data with single or double exponential equations. The error was calculated as 3σ from the fit of the experimental data. All experiments were performed at ambient temperature (295 ± 2 K) in aerated and argon-saturated solutions.

3. Results and discussion

Firstly, the drug stability was checked in aqueous solution in the dark. It was stable after 48 h, even when tested in acidic (pH 4) and alkaline (pH 9) solutions. These pH ranges are milder than those previously employed [11] and are usually considered environmentally relevant [13].

The absorption spectrum of rivastigmine (**1**) in water shows a band centred at 260 nm with a shoulder at 270 nm and a weak tail extending to $\lambda > 290$ nm (Fig. 1). Overlap of this spectrum with UV-B lamp emission spectrum is reported in Fig. 1.

Kinetics experiments were made irradiating a dilute water solution of rivastigmine (5×10^{-5} M) in a pyrex photoreactor with UV-B light. Under these conditions the drug exhibited a half-life time of 177.5 h and a polychromatic quantum yield of 2.61×10^{-3}.

A 1×10^{-5} M solution of the drug in water was exposed to sunlight in quartz tubes in Naples in September–October 2010. Drug changes were monitored by HPLC. After 12 days rivastigmine decreased by approximately 50% and converted to ketone **2** and, in small amount, alcohol **3**. Ketone **2** was identified by comparing HPLC retention time with an authentic sample (see below) and by characteristic ^{1}H NMR signals identified in the mixture. Due to the low amount, alcohol **3** was detected only by ^{1}H NMR comparing its signals with those of an authentic sample (see below).

With the purpose of isolating the photoproducts more concentrated drug solutions (1×10^{-3} M) were used. Since rivastigmine (**1**) and its photoproducts are slightly soluble in water, acetonitrile was chosen as cosolvent to obtain clear solutions. In order to accelerate the photoreaction, irradiation of the 1×10^{-3} M solution

Table 1
^{1}H and ^{13}C NMR Data of compounds **1**, **2**, **3** and **6** ($CDCl_3$, 500 MHz).[a]

No.[b]	**1**		**2**		**3**		**6**	
	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C
1		145.4		138.4		147.4		145.1
2	7.05 brs	120.7*	7.70 brs	121.6	7.14 brs	118.8	7.06 brs	119.5
3		151.4		151.8		151.6		151.7
4	7.00 brd (7.8)	120.2*	7.34 brd (7.0)	126.7	7.01 brd (8.0)	120.7	7.04 brd (7.8)	120.8
5	7.27 t (7.8)	128.8	7.47 t (8.0)	129.4	7.32 t (8.5)	129.2	7.33 t (7.8)	129.2
6	7.10 d (7.8)	124.2	7.79 d (8.0)	125.0	7.18 d (8.5)	122.1	7.13 d (8.5)	122.8
7	3.26 q (6.7)	65.5		197.2	4.88 q (6.4)	70.0	4.29 q (6.5)	79.3
8	1.35 d (6.7)	19.9	2.60 s	19.9	1.48 d (6.4)	25.0	1.42 d (6.5)	23.8
9	2.19 s	43.0						
10		154.5; 154.3		154.5; 154.3		154.6; 154.4		154.6; 154.3
11	3.45 and 3.39 q (7.0)	44.0	3.49 and 3.41 q (7.0)	44.1	3.48 and 3.41 q (6.9)	44.1	3.48 and 3.41 q (6.9)	44.1
12	1.22 and 1.17 t (7.0)	13.2; 12.4	1.27 and 1.20 t (7.0)	13.2; 12.4	1.24 and 1.19 t (6.9)	13.2; 12.5	1.25 and 1.19 t (6.9)	13.2; 12.5
13	3.05 and 2.97 s	34.1; 33.7	3.09 and 3.00 s		3.07 and 2.99 s	34.2; 33.8	3.07 and 2.99 s	34.2; 33.8
OMe							3.23 s	56.6

[a] *J* values are in parentheses and are reported in Hz; chemical shifts are given in ppm; assignments were confirmed by COSY, HSQC, and HMBC experiments.
[b] Numbering of carbons of compounds **1** and **2** is reported in Fig. 2; it has been used also for compounds **3**–**6**.

(water/acetonitrile 9:1, v/v) was carried out in quartz tubes using UV-B lamps. Drug changes were monitored by HPLC and ^{1}H NMR confirming the trend observed in dilute conditions. Products **2** and **3** were isolated by TLC and spectroscopically characterized (Fig. 2, Table 1). Furthermore, chromatography gave a fraction consisting of two products in very small amounts that were successively identified as alkene **4** and alkane **5** (Fig. 2).

With the aim of verifying the role of water and oxygen in the drug photodegradation and for products studies, UV-B irradiation experiments were carried out in different solvents (acetonitrile, methanol, methanol/acetonitrile 9:1, v/v) and in the presence and absence of oxygen. Table 2 reports product distribution after 1 h irradiation. The product percentages were deduced by ^{1}H NMR by integration of characteristic signals.

All products isolated retain the carbamate moiety while they present a new function instead of $N(Me)_2$ function (Fig. 2). As shown in Table 2, in all solvents ketone derivative **2** is the main photoproduct, in water even under deareated conditions. Vinyl **4** and ethyl **5** derivatives are secondary products. Alcohol derivative **3** is present only in water while in methanol ether **6** is found. These observations were confirmed by irradiation experiments at longer times by both ^{1}H NMR and HPLC analyses.

Product structures indicate that, despite the presence in the drug of two photolabile functions, carbamate and benzylamine, only the latter is involved in the formation of the photoproducts. In particular, the primary photochemical event is the cleavage of benzyl-nitrogen sigma bond. This breakdown pattern is also observed in the mass spectrum of rivastigmine that exhibites a base peak at *m/z* 206 $[M-NMe_2]^+$ [14]. As reported for various benzylic compounds [15,16], the photochemical cleavage may occur

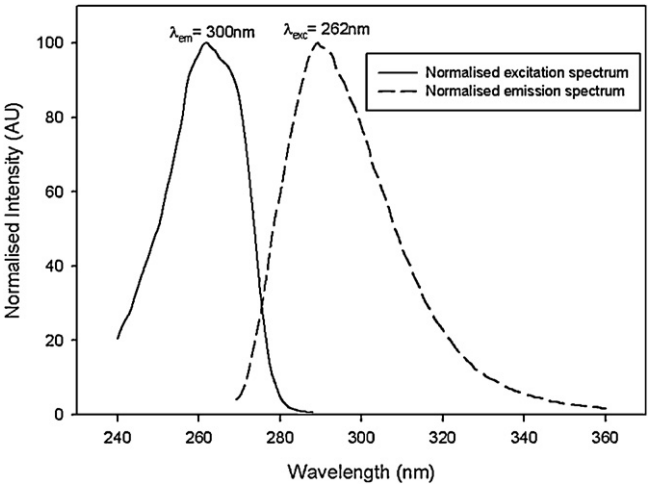

Fig. 3. Normalized fluorescence spectra (emission and excitation) of rivastigmine aqueous solution at 295 ± 2 K.

homolytically and/or heterolytically from the excited state S_1. In the case of rivastigmine, a value of 434 kJ mol^{-1} for the transition energy $S_0 \rightarrow S_1$ was estimated from the intersection between normalized emission and excitation spectra (Fig. 3). A plausible interpretation of the photochemical events from excited **1** is depicted in Fig. 4. If the cleavage occurs homolytically a radical pair **7** is formed, and by oxygen trapping it leads to ketone **2** via peroxidic species **8**. As minor route a disproportion occurs affording alkene **4** and alkane **5**. Radical pair **7** can also convert to ionic pair

Table 2
Product distribution after 1 h of UV-B irradiation of rivastigmine solutions (1×10^{-3} M) under different conditions.

Solvent	Condition	Yield (%)[a]					
		1	**2**	**3**	**4**	**5**	**6**
H_2O/CH_3CN[b]	In air	70	20	10	Trace	Trace	–
	Under argon	70	20	10	Trace	Trace	–
CH_3CN	In air	<5	95	–	–	–	–
	Under argon	90	–	–	5	5	–
CH_3OH	In air	<95	5	–	Trace	Trace	Trace
	Under argon	>95	–	–	Trace	Trace	<5
CH_3OH/CH_3CN[c]	In air	<90	10	–	Trace	Trace	–
	Under argon	<80	–	–	10	10	Trace

[a] By ^{1}H NMR.
[b] H_2O/CH_3CN 9:1 (v/v).
[c] CH_3OH/CH_3CN 9:1 (v/v).

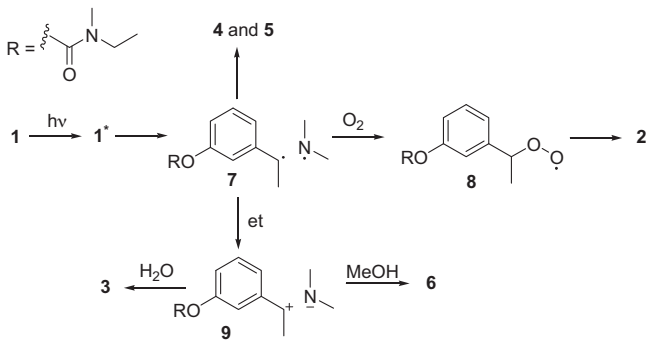

Fig. 4. Suggested phototransformation pathways.

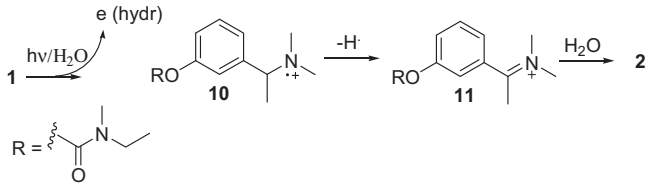

Fig. 5. Photoionization of rivastigmine in water.

via an electron-transfer with the formation of the well stabilized benzylic cation **9**. This process is supported by the presence of benzyl alcohol **3** in aqueous media and benzyl ether **6** in methanol deriving from the nucleophilic solvent trapping of the charged positive species [15]. The benzylic carbocation formation could be partly favoured by the presence of the oxygen-substituent in *meta*-position of the aromatic ring. In the photochemically excited state, *meta*-alkoxy groups are known to be particularly efficient in stabilization of a developing positive charge at a benzylic site [17].

Under aqueous conditions it is also possible that the drug undergoes a photoionization leading to radical cation **10** that could decay to **2** via reaction with oxygenated species (O_2, superoxide anion) or, in the absence of oxygen, via hydrolysis of iminium ion **11**, as reported in Fig. 5. This hypothesis accounts for the findings of ketone **2** in deaerated aqueous conditions.

It is well known that the photochemical single electron-transfer reaction (SET) of amines can be promoted by ketones [18,19]. Really, we observed that the photodegradation rate of rivastigmine increased (i) in time as well as ketone **2** was formed and (ii) in the presence of increasing concentrations of **2**, suggesting that ketone **2** itself could act as a photosensitizer. The tendency of the drug to give a radical cation via SET-promoted photochemical reaction (Fig. 6) was confirmed using a well-known electron-transfer sensitizer as 9,10-anthraquinone [20]. Irradiation at 360 nm of a water/acetonitrile solution of rivastigmine in the presence of 0.5 equiv. of this compound converted the drug to ketone **2** within 20 min while no trace was detected in the blank experiment.

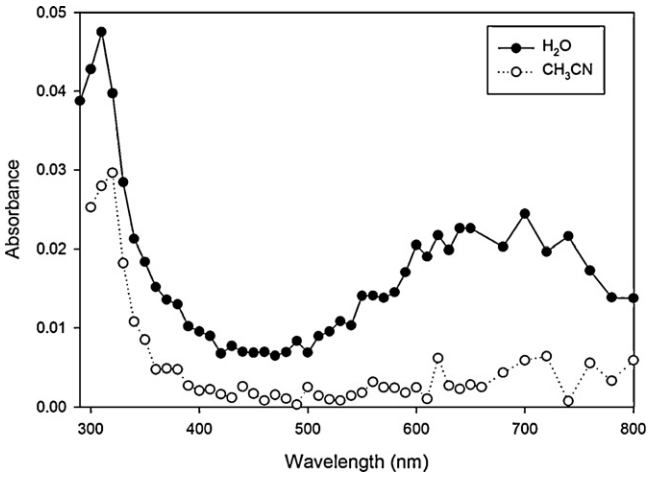

Fig. 7. Transient absorption spectra obtained after 266 nm excitation of 1.66 mM rivastigmine in water (filled circles) and acetonitrile (empty circles) solutions at 295 ± 2 K.

3.1. Laser flash photolysis data

In order to support our mechanistic hypotheses transient absorption spectroscopy experiments were performed. Fig. 7 shows the transient absorption spectra upon LFP excitation (266 nm) of rivastigmine (1.66 mM) in pure water and acetonitrile immediately after the laser pulse (0.2 μs) at 295 K in open cuvettes. For the absorption band centred at 310 nm, present in both cases, a fast decay of a first species (probably due to the triplet state) and formation and consequent decay of a long lived transient were observed. Observing the trace at 310 nm (data not shown) it is possible to see the parallel decay and growth from two unrelated transients having different rate constants. From the fit of the second species absorption vs. time decay we obtained a correlation between the pseudo-first order decay and the water/acetonitrile percentage ranging from $(3.24 \pm 0.01) \times 10^5$ (100% water) up to $(6.18 \pm 0.06) \times 10^6$ (100% acetonitrile). On the basis of literature data, the methyl-benzyl radicals typical absorption band is in the range 315–320 nm [21,22]. Thus, it is reasonable to attribute this signal to the formation of the resonance-stabilized benzylic radical **7**. Isolation of typical radical-derived products as alkene **4** and alkane **5** confirms this assignment. The second absorption band, centred at 700 nm and detected only in water, could be attributed to the presence of the solvated electron [23]. This supports the proposed mechanism reported in Fig. 5 although the absence of absorption band of radical cation **10**. In an attempt to evidence the formation of this intermediate we tried to selectively form it via electron transfer from rivastigmine to the triplet excited chloranyl at 355 nm [24] (data not shown). Under such conditions it was possible to discern only the band at 450 nm due to the chloranil radical anion, but no transient relative to radical cation **10**. It is probable that this species is not detectable owing to its low absorption and/or short life-time.

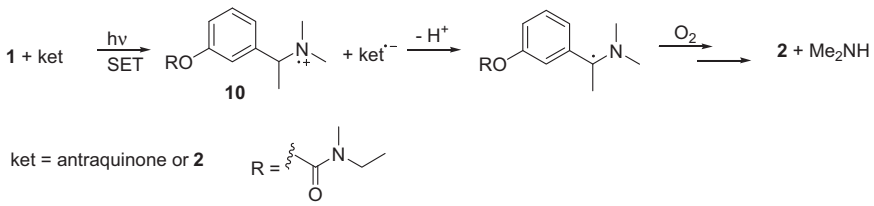

Fig. 6. Single electron-transfer (SET) of rivastigmine in the presence of aromatic ketones.

4. Conclusion

The work gives further information on the photochemical behaviour of benzylic amines confirming the tendency to undergo β-cleavage and to give an electrofugal group in the presence of suitable N-substituents [18]. Indeed, the photodegradation of rivastigmine in water involves the tertiary amino site and leads mainly to ion-derived products characterized by departure of Me_2N-group. It is reported that amines bearing N-substituents that are capable of stabilizing the formed aminium radicals have lower oxidation potentials as compared to those with N-electron withdrawing substituents as amides and carbamates [18]. This could account for the unreactivity of the carbamic-N function of rivastigmine as compared with the reactivity of the benzylaminic function [18].

The drug photodegradation is slow by direct irradiation with UV-B light or sunlight, but it is promoted by ketones or photoelectron-transfer sensitizers such as anthraquinone. It is noteworthy that the main photoproduct is a ketone, and it influences the photodegradation rate of the drug acting as photosensitizer. This poses a problem in a possible indirect photosensitive effect of rivastigmine.

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NMR spectra have been performed at Centro di Metodologie Chimico-Fisiche of University of Naples on a Varian Inova-500 instrument of Consortium INCA.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jphotochem.2012.04.015.

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