Photodegradation of Nifedipine under Aerobic Conditions: Evidence of Formation of Singlet Oxygen and Radical Intermediate

To the Editor:

The nitrophenyldihydropyridine derivative nifedipine (1) is a coronary vasodilator[1–3] and calcium antagonist[4] that undergoes photochemical oxidation when exposed to light.[5] The possibility of the photooxidation of 1, as well as the kinetics of this process,[6] has been pointed out by several investigators.[7–9] Nifedipine undergoes rapid photochemical oxidation to the corresponding nitrophenylpyridine when exposed to light. This transformation is accompanied by a remarkable weakening of the pharmacological activity.[10] This drug produces photosensitivity reactions such as severe blistering and skin eruption.[11] Furthermore, 1 can initiate adverse light-induced biological effects in the skin.[12] In this context, nothing is as yet known about the relation between the photochemistry and phototoxicity of 1. We examined the photolysis of 1 under various conditions, with the main goals of establishing the role of oxygen in these photoprocesses and the mechanism of the reaction.

Irradiation of a methanol solution of 1 under oxygen atmosphere yields 2 as the main photoproduct and also produces singlet oxygen, as evidenced by trapping of oxygen with 2,5-dimethylfuran (Scheme I).

Nifedipine (1; Adalat, BAY 1040, Bayer) was irradiated for 8 h in methanol (0.500 g, 1.44 mmol in 50 mL) at 20 °C by means of a Pyrex immersion-well photoreactor (Applied Photophysics parts no. 3230 + 3307), equipped with a Philips HPL-N 250-W medium-pressure Hg lamp, under oxygen atmosphere. The course of the reaction was followed by UV–visible spectrophotometry with a Perkin-Elmer 559 A spectrophotometer. The result is shown, for a methanolic solution (6.0×10^{-5} M) of 1, in Figure 1.

After irradiation, the solvent was evaporated at reduced pressure, and the residue was purified by column chromatography (silica gel). Elution was carried out with a mixture of hexane:ethyl acetate (5:1, v/v). The photoproduct 2 was isolated (0.364 g, 1.06 mmol; mp, 78–80 °C) and analyzed by ^{1}H NMR spectrometry (Bruker Aspect 3000, 300 MHz), IR spectrometry (Nicolet DX V5.07), and MS (Carlo Erba/Kratos MS25RFA): IR (KBr) ν: 2900, 1728, 1556, 1489, 1436, 1416, 1370, 1330, 1297, 1244, 1157, 1111, and 1044 cm^{-1}; ^{1}H NMR (C_3D_6O, 300 MHz): δ 2.61 (s, 6H, -CH_3), 3.39 (s, 6H, -$OOCH_3$), 6.55 (m, 1H, aromatic-H), 7.58 (m, 2H, aromatic-H), 7.89 (m, 1H, aromatic-H); MS (70 ev): *m/z* (%) 344 (4, M^+), 313 (6, M^+-OCH_3), 298 (20, M^+-NO_2), 283 (40, M^+-NO_2-CH_3), 267 (100, M^+-NO_2-OCH_3), 253 (40), 223 (20), 152 (40), 127 (25), 77 (20), 63 (20), 59 (50), 44 (30), 39 (25). Compound 2 was isolated by Mendenwald et al.[13] and Schlossmann[14] as one of the principal metabolites of 1, and the spectroscopic data[13] for this compound were virtually the same as those reported here. Nitroso monomer and dimer products, previously obtained by Majeed et al.[6] and Goerlitzer and Buss,[15] were isolated by MS in only trace amounts under our irradiation conditions. When the irradiation of 1 was repeated under the same experimental conditions, but under a N_2 atmosphere, the same compound was obtained as the main photoproduct (i.e., 2).

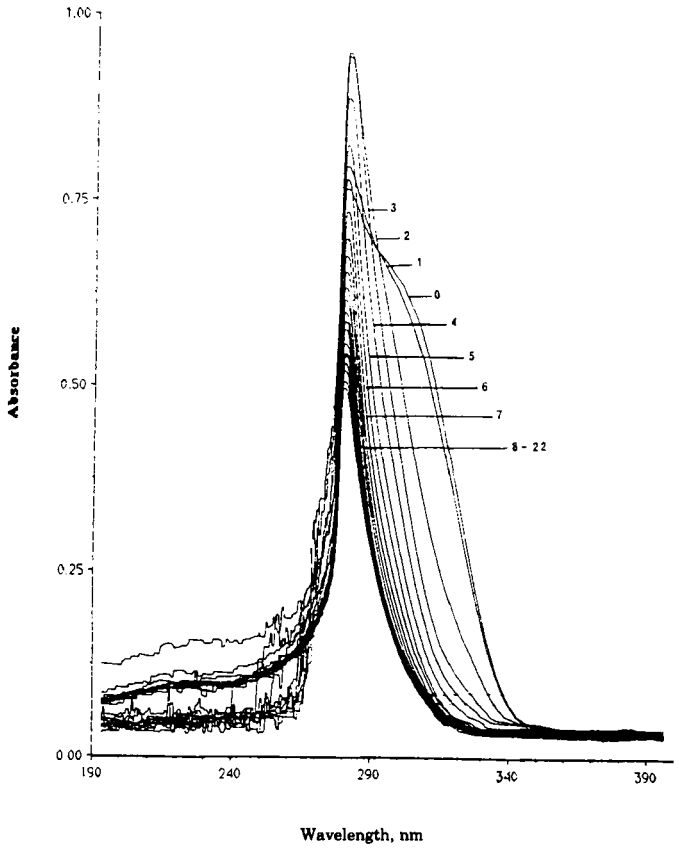
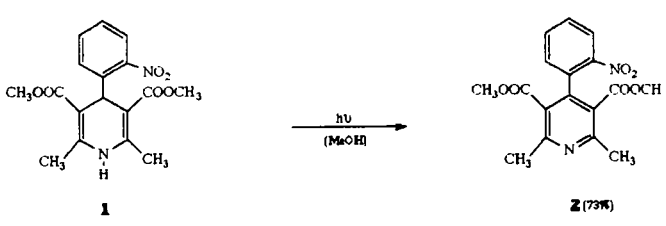


Figure 1—UV–visible spectra of 1 at regular time intervals of 8 s of irradiation.

In a separate experiment, 1 was found to be capable of producing singlet oxygen when irradiated in the presence of molecular oxygen. This fact can be verified by trapping the excited species by means of 2,5-dimethylfuran, which is routinely used as an $^1O_2$ scavenger.[16–18] Trapping of singlet oxygen in this manner leads to the formation of hexene-2,5-dione (20%), *cis*- and *trans*-3-oxo-1-butenyl acetate (10 and 40%, respectively), and 2-methoxy-5-hydroperoxide-2,5-dimethylfuran (30%),[19] as detected by GC–MS (Carlo Erba/Kratos MS25RFA instrument equipped with a 25-m capillary column of cross-linked 5% phenylmethylsilicone). A control experiment was performed in the presence of 2,5-dimethylfuran under N_2 atmosphere, and no



Scheme I

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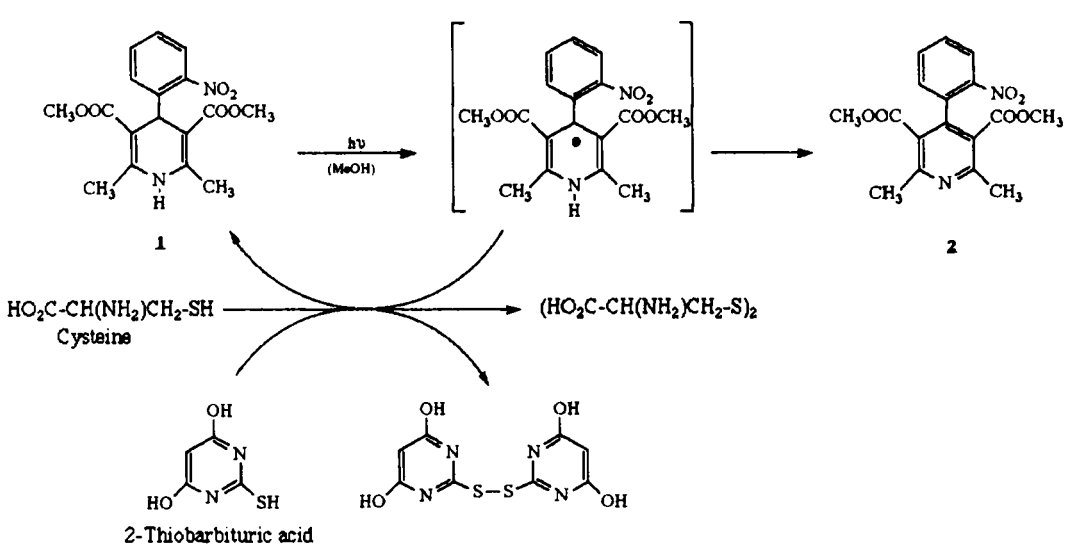

Scheme II—Proposed mechanism for the photodegradation of 1 in oxygenated media.

formation of the corresponding products was detected. When the experiment was carried out in the presence of 2,5-dimethylfuran, molecular oxygen, and sodium azide (a singlet-oxygen quencher), the formation of trapping products from $^1O_2$ with 2,5-dimethylfuran was not detected either. This result shows that not all of the $^1O_2$ produced in the reaction is physically and chemically quenched by 1, as was shown by Gurinovich and co-workers.[20]

As shown in Figure 1, the abnormal enhancement of the absorbance during the photolysis of 1 was greatest in the first 24 s (absorbances 1, 2, and 3 in Figure 1). Subsequently, diminution of this band demonstrated normal photodegradation. This fact indicated that an unstable intermediate was present during this process. The course of this reaction is shown in Scheme II. The formation of photoproduct 2 is compatible with an initial excitation of 1 after light absorption, followed by a photooxidation. The formation of 2 probably involves reaction of the radical intermediate. This intermediate could be detected when the irradiation was carried out in the presence of 2-thiobarbituric acid and cysteine, which was used as a radical scavenger (it has an effect similar to that of glutathione).[17] An indication of free radical formation was given by the capacity of 1 to induce dimerization of 2-thiobarbituric acid under irradiation, as was detected by UV–visible spectrophotometry at 532 nm, and of cysteine, detected by MS of the precipitated cystine product. The possibility of involvement of free radicals was also investigated by photopolymerization experiments.[21] Indication of the free radical formation was given by the capacity of 1 in deaerated methanolic solution to induce polymerization of acrylamide (0.125 M) under irradiation.

From these data it may be inferred that the phototoxicity mechanism for 1 most probably involves reactions of a free radical intermediate, stable photoproducts (2), or singlet oxygen with cellular components.

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Received February 4, 1991.
Accepted for publication June 15, 1991.