Synthesis of Furo[3,2-*g*][1,4]Benzoxazin-3-ones, New Psoralen Isosters

Adriana Chilin,*[a] Alessia Confente,[a] Giovanni Pastorini,[b] and Adriano Guiotto[a]

Keywords: Antitumour agents / Nitrogen heterocycles / Ring contraction / Aromatic nitration

Furobenzoxazin-3-one, a new tricyclic nucleus, was synthesised in two different and straightforward routes: the first route consisted of condensing a furan ring onto a preconstituted 1,4-benzoxazinone nucleus, and the other in condensing a 1,4-oxazine ring onto the appropriate benzofuran system obtained from coumarin by ring contraction.

Introduction

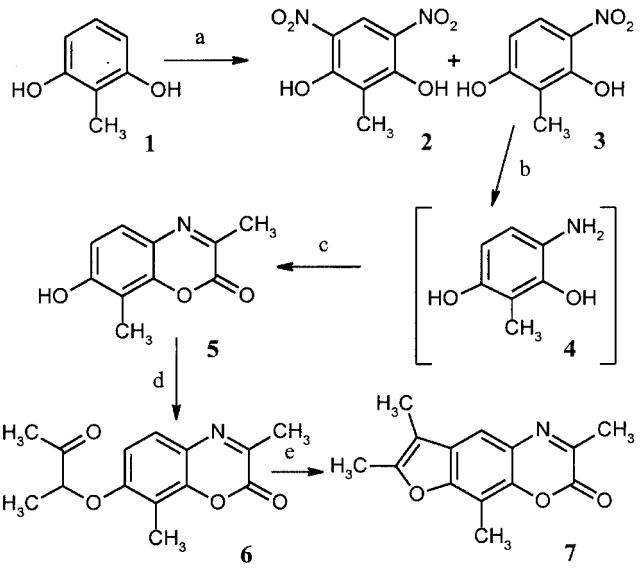
Psoralens are well-known natural and/or synthetic drugs currently used in PUVA (Psoralen plus UVA) therapy for various skin diseases.[1] Whilst searching for new isosters of psoralens in order to retain the high antiproliferative activity of the parent compounds but hoping to reduce the severe side-effects,[2−4] we synthesised a number of tricyclic compounds in which a carbon or an oxygen atom of the furocoumarin nucleus was substituted by a nitrogen atom.[5−7]

Both the photochemical behaviour of these psoralen isosters in terms of their ability to photobind to DNA thymine, and their photobiological properties, are deeply modified.[5−8] Accordingly, we planned the synthesis of a new tricyclic system, furo[3,2-*g*][1,4]benzoxazin-3-one, in which the carbon atom at the 4-position of psoralen was substituted by a nitrogen atom.

Results and Discussion

To build this new tricyclic system, we tried two synthetic strategies, one consisting of condensing a furan ring onto a preconstituted 1,4-benzoxazinone, and the other in condensing a 1,4-oxazine ring onto an appropriate benzofuran.

Following the first strategy, the synthesis started from 2-methylresorcinol, because the presence of the methyl group univocally afforded the desired product with a linear psoralen-like geometry. Thus, as shown in Scheme 1, resorcinol **1** was directly nitrated under mild conditions to yield compounds **2** and **3**: *n*-pentyl nitrite in ether[9] gave the mononitrated derivative **3** in satisfactory yield (22%), along with a small amount of dinitro derivative **2** (5%), and many unidentified degradation products, whereas classic nitrating agents (like nitric acid or potassium nitrate in various conditions) only gave the dinitrated product, and other reagents, such as nitronium tetrafluoroborate, did not give better yields than the reaction with *n*-pentyl nitrite. The nitro derivative **3** was reduced by catalytic hydrogenation to the corresponding amino derivative **4**, which was directly condensed with methyl pyruvate[10] to give 1,4-benzoxazin-3-one (**5**). All attempts to isolate **4** failed, owing to rapid oxidative degradation, with darkening of the reaction product. Benzoxazinone **5** was then condensed with 3-chloro-2-butanone, and the resulting ether **6** was cyclized in an acidic

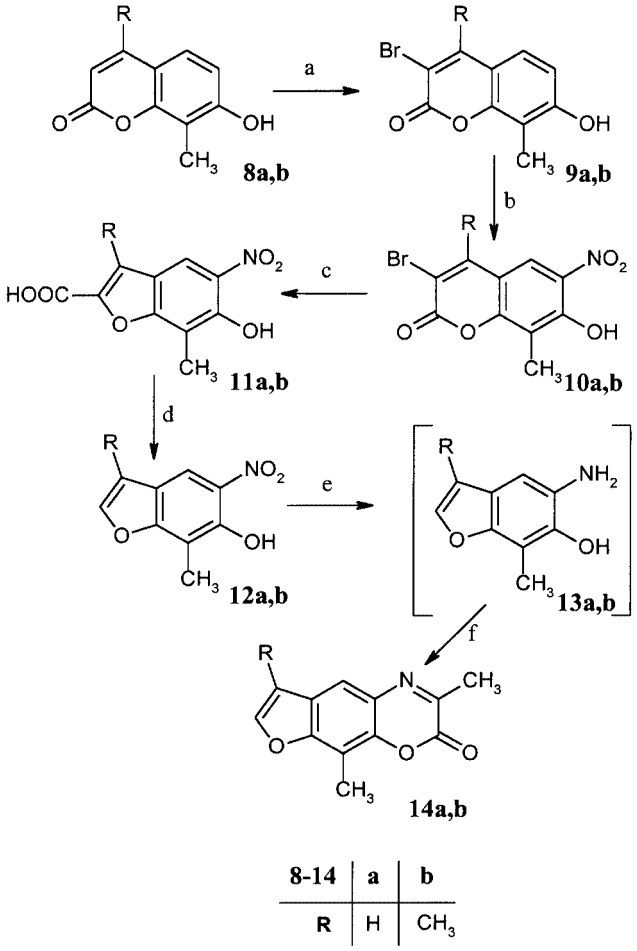


Scheme 1. Reagents and conditions: (a) *n*-pentyl nitrite, diethyl ether, room temp., 1 h, 5% (**2**) and 22% (**3**); (b) Pd/C 10%, H_2, abs. EtOH, room temp., 24 h; (c) methyl pyruvate, room temp., 5 h, 87% (b + c); (d) 3-chlorobutan-2-one, anhydrous K_2CO_3, acetone, reflux, 7 h, 54%; (e) conc. H_2SO_4, room temp., 3 h, 28%

[a] Dipartimento di Scienze Farmaceutiche, Università degli Studi di Padova,
Via Marzolo 5, 35131 Padova, Italy
Fax: (internat.) + 39-049/827-5366
E-mail: adriana.chilin@unipd.it
[b] Istituto di Chimica Biomolecolare del CNR,
Via Marzolo 3, 35131 Padova, Italy

medium[7] to yield 2,5,7,8-tetramethyl-3*H*-furo[3,2-*g*][1,4]-benzoxazin-3-one (**7**).

In the alternative route, we first built the benzofuran nucleus and then the azalactonic ring. In this case, a nitration reaction introducing the nitrogen atom into the final appropriate position was carried out on bromocoumarins **9**, and the resulting nitrocoumarins were submitted to Perkin's ring contraction[11,12] to afford the desired 5-nitrobenzofurans. This route was required because benzofurans are preferentially nitrated on the unsubstituted or partially substituted furan ring. Therefore, as shown in Scheme 2, the 7-hydroxy-8-methylcoumarins **8** were reacted with bromine to yield the corresponding 3-bromo derivatives **9**, which were submitted first to nitration and then to ring contraction in a strongly alkaline medium. Compounds **11** were then decarboxylated, yielding nitrobenzofurans **12**, which were reduced by catalytic hydrogenation to the corresponding aminobenzofurans **13** and directly condensed with methyl pyruvate to give 2,5-dimethyl-3*H*-furo[3,2-*g*][1,4]benzoxazin-3-one (**14a**) and 2,5,8-trimethyl-3*H*-furo[3,2-*g*][1,4]benzoxazin-3-one (**14b**).



Scheme 2. Reagents and conditions: (a) Br_2, glacial AcOH, 60 °C, 30 min, 48 and 60%; (b) HNO_3, H_2SO_4, 0 °C, 95%; (c) KOH, di(-ethylene glycol) ethyl ether, 98%; (d) quinoline, Cu, reflux, 30 min, 25%; (e) Pd/C 10%, H_2, abs. EtOH, room temp., 2 h; (f) methyl pyruvate, room temp., 12 h, 50% (e + f)

This second synthetic pathway yielded the 4-azapsoralen nucleus, although more steps were required than in the first strategy, but with a nearly identical overall yield. In addition, the second method yielded derivatives which were unsubstituted or monomethylated on the furan ring.

In conclusion, a new tricyclic system was synthesised in two different straightforward ways, with similar yields and with different degrees of substitution in the molecule.

Experimental Section

Analytical TLC was performed on precoated 60 F_{254} silica gel plates (0.2 mm; Merck) eluting with a $CHCl_3$/MeOH mixture (9:1). Column chromatography was performed using silica gel 60 (0.063−0.100 mm; Merck), eluting with $CHCl_3$. Melting points were determined on a Gallenkamp MFB-595-010M melting point apparatus and are uncorrected. IR spectra were recorded on a Perkin−Elmer 1600 FT-IR spectrometer. ^1H NMR and ^{13}C NMR spectra were recorded at 300.13 MHz and 75.47 MHz, respectively, on a Bruker AMX300 spectrometer with TMS as internal standard. HR-ESI-TOF mass spectra of the final products were recorded on a PE Biosystems Mariner 5220 spectrometer, using desipramine hydrochloride and dansyl-Gly-Trp as internal standards. Microanalyses were performed by the Microanalytical Laboratory of the Department of Pharmaceutical Sciences of University of Padova and were within ±0.3%. All reagents and solvents were of commercial quality and were used without purification.

The starting materials **8a**[13] and **8b**[14] were prepared according to literature methods.

1,3-Dihydroxy-2-methyl-4-nitrobenzene (3): *n*-Pentyl nitrite[15] (0.8 mL, 9.6 mmol) was added to a solution of **1** (1.0 g, 8.0 mmol) in diethyl ether (40 mL) and the mixture was kept at room temperature for 1 h. The solvent was evaporated under reduced pressure and the residue was purified by column chromatography to give **2** (0.05 g, 5%), followed by the desired product **3** (0.30 g, 22%).
2: M.p. 133 °C. ^1H NMR ($CDCl_3$): δ = 2.13 (s, 3 H, 2-Me), 7.74 (s, 1 H, 5-H), 11.21 (s, 2 H, OH) ppm. $C_7H_6N_2O_6$ (214.14): calcd. C 39.26, H 2.82, N 13.08; found C 39.24, H 2.91, N 13.05.
3: M.p. 138 °C. ^1H NMR ($CDCl_3$): δ = 2.12 (s, 3 H, 2-Me), 6.37 (d, *J* = 9.3 Hz, 1 H, 6-H), 7.85 (d, *J* = 9.3 Hz, 1 H, 5-H), 11.29 (s, 2 H, OH) ppm. $C_7H_7NO_4$ (169.14): calcd. C 49.71, H 4.17, N 8.28; found C 49.64, H 4.11, N 8.26.

7-Hydroxy-3,8-dimethyl-1,4-benzoxazin-2-one (5): A catalytic amount of Pd/C 10% was added to a solution of **3** (0.20 g, 1.2 mmol) in absolute EtOH (25 mL) and the mixture was kept at room temperature under a slight overpressure of H_2. The mixture was stirred until **3** disappeared (24 h, TLC) and the catalyst was then filtered off. Methyl pyruvate (0.24 g, 0.21 mL, 2.3 mmol) was added to the filtrate and the mixture was stirred at room temperature for a further 5 h. The solvent was evaporated under reduced pressure and the residue was crystallised from MeOH to give **5** (0.18 g, total 87%); m.p. 245 °C. ^1H NMR (CD_3OD): δ = 2.12 (s, 3 H, 8-Me), 2.33 (s, 3 H, 3-Me), 6.71 (d, *J* = 8.7 Hz, 1 H, 6-H), 7.23 (d, *J* = 8.7 Hz, 1 H, 5-H) ppm. $C_{10}H_9NO_3$ (191.19): calcd. C 62.82 H 4.74 N 7.33; found C 62.85, H 4.70, N 7.36.

3,8-Dimethyl-7-(1-methyl-2-oxopropoxy)-2*H*-1,4-benzoxazin-2-one (6): A mixture of **5** (0.19 g, 1.0 mmol), 3-chlorobutan-2-one (0.16 g, 0.15 mL, 1.5 mmol), anhydrous K_2CO_3 (0.50 g) and acetone (25 mL) was refluxed until **5** disappeared (7 h, TLC). After cooling,

the solid was filtered off and washed with acetone. The solvent was evaporated under reduced pressure from the combined filtrate and washings and the residue was purified by column chromatography to give **6** (0.14 g, 54%) as a gum. ^{1}H NMR (CD_3OD): δ = 1.47 (d, *J* = 6.8 Hz, 3 H, 1′-H), 2.14 (s, 3 H, 8-Me), 2.24 (s, 3 H, 4′-H), 2.37 (s, 3 H, 3-Me), 4.90 (q, *J* = 6.8 Hz, 1 H, 2′-H), 6.75 (d, *J* = 8.9 Hz, 1 H, 6-H), 7.38 (d, *J* = 8.9 Hz, 1 H, 5-H) ppm. $C_{14}H_{15}NO_4$ (261.28): calcd. C 64.36, H 5.79, N 5.36; found C 64.39, H 5.81, N 5.35.

2,5,7,8-Tetramethyl-3*H*-furo[3,2-*g*][1,4]benzoxazin-3-one (7): A solution of **6** (0.14 g, 0.54 mmol) in conc. H_2SO_4 (5 mL) was stirred at room temperature until the starting product had disappeared (3 h, TLC). The mixture was poured onto ice and H_2O (50 mL), neutralised with 10% $NaHCO_3$ and extracted with EtOAc (3 × 25 mL). The dried (Na_2SO_4) organic phase was concentrated under reduced pressure. The residue was purified by column chromatography and crystallised from MeOH to give **7** (0.04 g, 28%); m.p. 204 °C. IR (KBr): $\tilde{\nu}$ = 2915, 1730, 1635, 1575, 1430, 1160, 1120, 955, 910, 760 cm^{-1}. ^{1}H NMR ($CDCl_3$): δ = 2.10 (s, 3 H, 5-Me), 2.34 (s, 3 H, 2-Me), 2.46 (s, 3 H, 7-Me or 8-Me), 2.49 (s, 3 H, 7-Me or 8-Me), 7.47 (s, 1 H, 9-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 154.30 (C-3), 153.62 (C-5a), 152.98 (C-4a), 151.69 (C-7), 142.60 (C-2), 128.04 (C-8a and C-9a), 115.20 (C-9), 110.51 (C-5), 108.66 (C-8), 21.47 (2-Me), 12.37 (7-Me), 8.36 (8-Me), 8.26 (5-Me) ppm. HRMS: *m*/*z* = 244.0972 (calcd. for $C_{14}H_{14}NO_3$ 244.0968). $C_{14}H_{13}NO_3$ (243.26): calcd. C 69.12, H 5.39, N 5.76; found C 69.12, H 5.43, N 5.75.

General Procedure for the Synthesis of 3-Bromo-7-hydroxy-8-methylcoumarins (9): A solution of Br_2 (6.4 g, 40.0 mmol) in glacial AcOH (25 mL) was added dropwise at 60 °C to a solution of **8** (40.0 mmol) in glacial AcOH (150 mL), and the mixture was stirred for an additional 15 min. After cooling, the obtained precipitate was collected and crystallised from EtOH.

3-Bromo-7-hydroxy-8-methylcoumarin (9a): White needles (5.0 g, 48%); m.p. 220 °C. ^{1}H NMR ([D$_6$]acetone): δ = 2.27 (s, 3 H, 8-Me), 6.94 (d, *J* = 8.6 Hz, 1 H, 6-H), 7.39 (d, *J* = 8.6 Hz, 1 H, 5-H), 8.30 (s, 1 H, H-4), 9.34 (s, 1 H, OH) ppm. $C_{10}H_7BrO_3$ (255.07): calcd. C 47.09, H 2.76, Br 31.33; found C 47.12, H 2.79, Br 31.36.

3-Bromo-7-hydroxy-4,8-dimethylcoumarin (9b): White needles (6.4 g, 60%); m.p. 270 °C. ^{1}H NMR ([D$_6$]DMSO): δ = 2.25 (s, 3 H, 8-Me), 2.62 (s, 3 H, 4-Me), 6.99 (d, *J* = 8.9 Hz, 1 H, 6-H), 7.61 (d, *J* = 8.9 Hz, 1 H, 5-H), 10.59 (s, 1 H, OH) ppm. $C_{11}H_9BrO_3$ (269.10): calcd. C 49.10, H 3.37, Br 29.69; found C 49.11, H 3.47, Br 29.65.

General Procedure for the Synthesis of 3-Bromo-7-hydroxy-8-methyl-6-nitrocoumarins (10): A mixture of HNO_3 (1.4 mL) and H_2SO_4 (1.5 mL) was added dropwise at 0 °C to a solution of **9** (20.0 mmol) in conc. H_2SO_4 (20 mL). The mixture was poured onto ice and H_2O (200 mL) and the precipitate was collected and washed with H_2O. The solid was crystallised from MeOH.

3-Bromo-7-hydroxy-8-methyl-6-nitrocoumarin (10a): Pale yellow crystals (5.7 g, 95%); m.p. 194 °C. ^{1}H NMR ($CDCl_3$): δ = 2.40 (s, 3 H, 8-Me), 8.04 (s, 1 H, 4-H), 8.18 (s, 1 H, 5-H), 11.21 (s, 1 H, OH) ppm. $C_{10}H_6BrNO_5$ (300.07): calcd. C 40.03, H 2.02, Br 26.63, N 4.67; found C 40.01, H 2.00, Br 26.59, N 4.72.

3-Bromo-7-hydroxy-4,8-dimethyl-6-nitrocoumarin (10b): Yellow crystals (5.9 g, 95%); m.p. 265 °C. ^{1}H NMR ($CDCl_3$): δ = 2.41 (s, 3 H, 8-Me), 2.64 (s, 3 H, 4-Me), 8.36 (s, 1 H, 5-H), 11.17 (s, 1 H, OH) ppm. $C_{11}H_8BrNO_5$ (314.10): calcd. C 42.16, H 2.56, Br 25.44, N 4.46; found C 42.08, H 2.60, Br 25.45, N 4.46.

General Procedure for the Synthesis of 6-Hydroxy-7-methyl-5-nitrobenzofuran-2-carboxylic Acids (11): Compound **10** (16.0 mmol) was added portionwise to a solution of KOH (13.5 g, 24.0 mmol) in di(ethylene glycol) ethyl ether (100 mL) heated at 100 °C. After cooling, the mixture was diluted with H_2O (100 mL) and neutralised with 1 N HCl. The obtained precipitate was collected and crystallised from glacial AcOH.

6-Hydroxy-7-methyl-5-nitrobenzofuran-2-carboxylic Acid (11a): Colourless needles (3.7 g, 98%); m.p. 294 °C. ^{1}H NMR ([D$_6$]DMSO): δ = 2.50 (s, 3 H, 7-Me), 7.79 (s, 1 H, 3-H), 8.46 (s, 1 H, 4-H), 10.74 (s, 1 H, OH) ppm. $C_{10}H_7NO_6$ (237.17): calcd. C 50.64, H 2.97, N 5.91; found C 50.65, H 2.99, N 5.99.

6-Hydroxy-3,7-dimethyl-5-nitrobenzofuran-2-carboxylic Acid (11b): Colourless needles (3.9 g, 98%); m.p. 300 °C. ^{1}H NMR (CD_3OD): δ = 2.43 (s, 3 H, 7-Me), 2.55 (s, 3 H, 3-Me), 8.39 (s, 1 H, 4-H) ppm. $C_{11}H_9NO_6$ (251.20): calcd. C 52.59, H 3.61, N 5.58; found C 52.61, H 3.67, N 5.54.

General Procedure for the Synthesis of 6-Hydroxy-7-methyl-5-nitrobenzofurans (12): A mixture of **11** (15.0 mmol), quinoline (20 mL) and Cu powder (1.0 g) was refluxed for 30 min. After cooling, the mixture was diluted with EtOAc (50 mL), the solid was filtered off and the filtrate was extracted with 1 N HCl (4 × 50 mL). The dried (Na_2SO_4) organic layer was concentrated under reduced pressure and the residue purified by column chromatography.

6-Hydroxy-7-methyl-5-nitrobenzofuran (12a): Off-white powder (0.72 g, 25%); m.p. 70 °C. ^{1}H NMR ($CDCl_3$): δ = 2.25 (s, 3 H, 7-Me), 6.56 (d, *J* = 2.3 Hz, 1 H, 3-H), 7.47 (d, *J* = 2.3 Hz, 1 H, 2-H), 8.00 (s, 1 H, 4-H), 10.75 (s, 1 H, OH) ppm. $C_9H_7NO_4$ (193.16): calcd. C 55.96, H 3.65, N 7.25; found C 56.00, H 3.66, N 7.22.

6-Hydroxy-3,7-dimethyl-5-nitrobenzofuran (12b): Off-white powder (0.77 g, 25%); m.p. 105 °C. ^{1}H NMR ($CDCl_3$): δ = 2.23 (d, *J* = 1.2 Hz, 3 H, 3-Me), 2.42 (s, 3 H, 7-Me), 7.42 (q, *J* = 1.2 Hz, 1 H, 2-H), 8.16 (s, 1 H, 4-H), 10.98 (s, 1 H, OH) ppm. $C_{10}H_9NO_4$ (207.19): calcd. C 57.97, H 4.38, N 6.76; found C 57.92, H 4.42, N 6.76.

General Procedure for the Synthesis of Methyl-3*H*-furo[3,2-*g*]-[1,4]benzoxazin-3-one (14): A catalytic amount of Pd/C 10% was added to a solution of **12** (3.5 mmol) in absolute EtOH (50 mL) and the mixture was kept at room temperature under a slight overpressure of H_2. After stirring for 2 h, the catalyst was filtered off and methyl pyruvate (0.72 g, 0.64 mL, 7.0 mmol) was added to the filtrate. The mixture was stirred at room temperature for 12 h and then the solvent was evaporated under reduced pressure. The residue was purified by column chromatography and crystallised from MeOH.

2,5-Dimethyl-3*H*-furo[3,2-*g*][1,4]benzoxazin-3-one (14a): Pale yellow needles (37 mg, 50%); m.p. 184 °C. IR (KBr): $\tilde{\nu}$ = 3125, 2925, 1720, 1620, 1575, 1340, 1140, 1100, 1010, 870, 760 cm^{-1}. ^{1}H NMR ($CDCl_3$): δ = 2.52 (br. s, 6 H, 2-Me and 5-Me), 6.79 (d, *J* = 2.2 Hz, 1 H, 8-H), 7.64 (d, *J* = 2.2 Hz, 1 H, 7-H), 7.69 (s, 1 H, 9-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 154.65 (C-3), 153.97 (C-5a), 152.43 (C-4a), 147.07 (C-7), 143.12 (C-2), 128.47 (C-9a), 124.67 (C-8a), 118.02 (C-9), 109.66 (C-5), 107.45 (C-8), 21.51 (2-Me), 8.52 (5-Me) ppm. HRMS: *m*/*z* = 216.0664 (calcd. for $C_{12}H_{10}NO_3$ 216.0655). $C_{12}H_9NO_3$ (215.21): calcd. C 66.97, H 4.21, N 6.51; found C 66.99, H 4.23, N 6.49.

2,5,8-Trimethyl-3*H*-furo[3,2-*g*][1,4]benzoxazin-3-one (14b): Pale yellow needles (40 mg, 50%); m.p. 182 °C. IR (KBr): $\tilde{\nu}$ = 3100, 2925, 1725, 1620, 1575, 1350, 1130, 1070, 860, 760 cm^{-1}. ^{1}H NMR ($CDCl_3$): δ = 2.26 (d, *J* = 1.2 Hz, 3 H, 8-Me), 2.54 (s, 3 H, 2-Me or 5-Me), 2.56 (s, 3 H, 2-Me or 5-Me), 7.48 (q, *J* = 1.2 Hz, 1 H, 7-H), 7.65 (s, 1 H, 9-H) ppm. ^{13}C NMR ($CDCl_3$): δ = 154.95 (C-3), 154.08 (C-5a), 152.13 (C-4a), 143.39 (C-7), 143.18 (C-2), 128.17 (C-9a), 126.52 (C-8a), 116.59 (C-8), 116.35 (C-9), 109.52 (C-5),

21.51 (2-Me), 8.41 (8-Me), 8.24 (5-Me) ppm. HRMS: *m/z* = 230.0826 (calcd. for $C_{13}H_{12}NO_3$ 230.0813). $C_{13}H_{11}NO_3$ (229.24): calcd. C 68.11, H 4.84, N 6.11; found C 68.09, H 4.82, N 6.15.

[1] J. A. Parrish, R. S. Stern, M. A. Pathak, T. B. Fitzpatrick, in *The Science of Photomedicine* (Ed.: J. D. Regan and J. A. Parrish), Plenum, New York, **1982**, p. 595−624.
[2] D. J. Kirkland, K. L. Creed, P. Mannisto, *Mutation Res.* **1983**, *116*, 73−82.
[3] G. J. Hook, J. A. Heddle, R. R. Marshall, *Cytogenet. Cell Genet.* **1983**, *35*, 100−103.
[4] R. S. Stern, R. Lange, *J. Invest. Dermatol.* **1988**, *91*, 120−124.
[5] O. Gia, S. Mobilio, A. Chilin, P. Rodighiero, M. Palumbo, *J. Photochem. Photobiol. B: Biol.* **1988**, *2*, 435−442.
[6] A. Chilin, P. Rodighiero, G. Pastorini, A. Guiotto, *Gazz. Chim. Ital.* **1988**, *118*, 513−516.
[7] P. Rodighiero, A. Guiotto, A. Chilin, F. Bordin, F. Baccichetti, F. Carlassare, D. Vedaldi, S. Caffieri, A. Pozzan, F. Dall′Acqua, *J. Med. Chem.* **1996**, *39*, 1293−1302.
[8] D. Vedaldi, F. Dall′Acqua, S. Caffieri, F. Baccichetti, F. Carlassare, F. Bordin, A. Chilin, A. Guiotto, *Photochem. Photobiol.* **1991**, *53*, 143−148.
[9] T. Ajello, G. Sigillò, *Gazz. Chim. Ital.* **1939**, *69*, 57−71.
[10] E. Biekert, D. Hoffmann, F. J. Meyer, *Chem. Ber.* **1961**, *94*, 1664−1670.
[11] W. H. Perkin, *J. Chem. Soc.* **1870**, *8*, 368−371.
[12] W. H. Perkin, *J. Chem. Soc.* **1871**, *9*, 37−52.
[13] K. D. Kaufman, D. J. Erb, T. M. Blok, R. W. Carlson, D. J. Knoechel, *J. Heterocycl. Chem.* **1982**, *19*, 1051−1056.
[14] S. Marciani, A. Guiotto, P. Rodighiero, O. Gia, G. Rodighiero, *Il Farmaco Ed. Sci.* **1979**, *34*, 234−247.
[15] P. S. J. Canning, K. McCrudden, H. Maskill, B. Sexton, *J. Chem. Soc., Perkin Trans. 2* **1999**, 2735−2740.

Received February 4, 2002
[O02062]