Synthesis of Benzoxazino- and Naphthoxazinoquinoline Derivatives as Possible Antitumor Agents

M. NASR[x], M. M. ABBASI, and I. NABIH

Abstract □ The synthesis of 2-methoxy-6-methyl-1-nitro-12*H*-quino[3,4-*b*][1,4]benzoxazine (III*a*), its 10-chloro derivative (III*b*), 2-methoxy-6-methyl-1-nitro-14*H*-naphth[1′,2′:5,6][1,4]oxazino[2,3-*c*]quinoline (IV*a*), and 2-methoxy-6-methyl-1-nitro[1,4]benzodioxino[2,3-*c*]quinoline (VI) is described. Compounds III*a*, III*b*, and IV*a* were condensed with benzaldehyde to give the corresponding styryl derivatives III*c*, III*d*, and IV*b* in an attempt to introduce new compounds that may have antitumor activity.

Keyphrases □ Benzoxazinoquinoline derivatives—synthesized as possible antitumor agents □ Naphthoxazinoquinoline derivatives—synthesized as possible antitumor agents □ Styrylquinoline derivatives—synthesis of benzoxazinoquinolines and naphthoxazinoquinolines □ Antitumor agents, potential—synthesis of benzoxazinoquinoline and naphthoxazinoquinoline derivatives

The preparation of benzoxazine derivatives *via* the Turpin (1) reaction of 2-aminophenol with picryl chloride has created much interest both from the synthetic and biological points of view (2–5). Crossley *et al.* (6) prepared a series of phenoxazine derivatives, and their differential tissue staining and tumor growth retardation activity were determined. Moreover, representatives of this class of compounds have exhibited antitubercular (7), sedative, antiepileptic, and tranquilizing (8) activities. Also, spasmolytic activity has been shown by phenoxazine carboxylic acid derivatives (9, 10).

In view of the fact that styrylquinolines have also been reported to exhibit antitumor activity (11), new agents that would structurally include systems of oxazine and benzoxazine fused with styrylquinolines might prove to be useful as antitumor agents. Therefore, the synthesis of some new benzoxazino- and naphthoxazinoquinoline derivatives *via* the Turpin reaction is reported here.

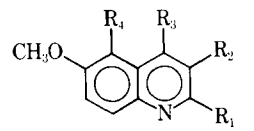
DISCUSSION

Synthesis was achieved by nitration of 4-hydroxy-6-methoxy-2-methylquinoline (I*a*) (12) to give the corresponding 3,5-dinitro derivative (I*b*) which, upon treatment with phosphorus oxychloride, yielded 4-chloro-3,5-dinitro-6-methoxy-2-methylquinoline (I*c*). Treatment of I*c* with benzaldehyde in the presence of fused zinc chloride gave 3,5-dinitro-4-hydroxy-6-methoxy-2-styrylquinoline (I*d*), which upon reaction with phosphorus oxychloride gave the 4-chloro derivative I*e*. When I*c* was allowed to condense with 2-aminophenol or 2-amino-4-chlorophenol, the product was either the 2-methoxy-6-methyl-1-nitro-12*H*-quino[3,4-*b*][1,4]benzoxazine (III*a*) or its 10-chloro derivative (III*b*).

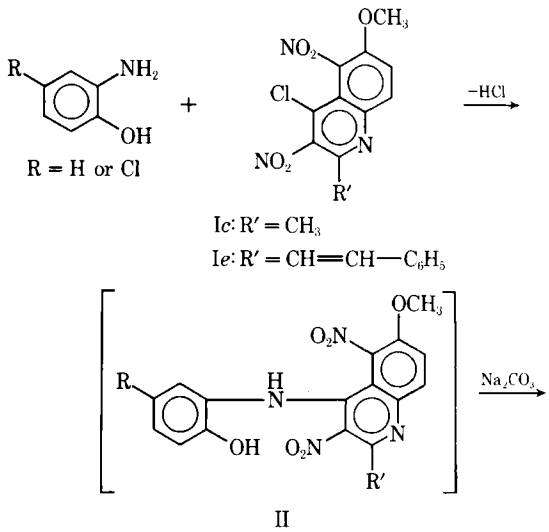
One can adequately explain the formation of III*a* and III*b* from the condensation of 2-aminophenols with I*c* and I*e* on the basis of initial formation of intermediate II *via* hydrochloric acid elimination (Scheme I). This, followed by intramolecular nucleophilic displacement of the nitro group by the phenoxide moiety under the influence of alkali, gave III*a* and III*b*.

Treatment of III*a* or III*b* with benzaldehyde in the presence of fused zinc chloride gave the styryl derivatives III*c* or III*d*. Compounds III*c* and III*d* were also obtained when the styryl derivative I*e* was allowed to condense with 2-aminophenol and 2-amino-4-chlorophenol, respectively. Similarly, reaction of I*c* with 2-amino-1-naphthol gave 2-methoxy-6-methyl-1-nitro-14*H*-naphth[1′,2′:5,6][1,4]oxazino[2,3-*c*]quinoline (IV*a*). The styryl derivative IV*b* was obtained when IV*a* was treated with benzaldehyde in the presence of fused zinc chloride.

Condensation of I*c* with phenol in dry pyridine gave the phenoxy derivative I*f*. This condensation suggested the use of catechol instead of 2-aminophenol in the Turpin reaction with I*c*. Thus, catechol condensed with I*c* in dry pyridine to give 3,5-dinitro-4-

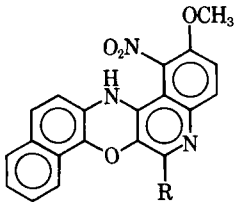


I*a*: $R_1 = CH_3, R_2 = R_4 = H, R_3 = OH$
I*b*: $R_1 = CH_3, R_2 = R_4 = NO_2, R_3 = OH$
I*c*: $R_1 = CH_3, R_2 = R_4 = NO_2, R_3 = Cl$
I*d*: $R_1 = CH{=}CH{-}C_6H_5, R_2 = R_4 = NO_2, R_3 = OH$
I*e*: $R_1 = CH{=}CH{-}C_6H_5, R_2 = R_4 = NO_2, R_3 = Cl$
I*f*: $R_1 = CH_3, R_2 = R_4 = NO_2, R_3 = OC_6H_5$



III*a*: R = H, R′ = CH_3
III*b*: R = Cl, R′ = CH_3
III*c*: R = H, R′ = $CH{=}CH{-}C_6H_5$
III*d*: R = Cl, R′ = $CH{=}CH{-}C_6H_5$

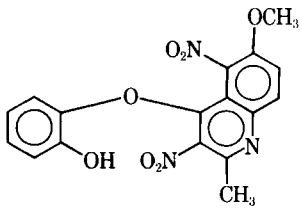
Scheme I



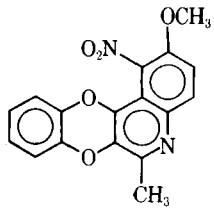
IV*a*: R = CH_3

IV*b*: R = CH=CH—C_6H_5

(2′-hydroxyphenoxy)-6-methoxy-2-methylquinoline (V), which gave 2-methoxy-6-methyl-1-nitro[1,4]benzodioxino[2,3-*c*]quinoline (VI) when gently warmed with alkali.



V



VI

EXPERIMENTAL[1]

3,5-Dinitro-4-hydroxy-6-methoxy-2-methylquinoline (I*b*)—Compound I*a* (12) (3 g, 0.0016 mole) was added portionwise with stirring to 30 g of cooled nitric acid (specific gravity 1.39), and the mixture was kept at room temperature for 24 hr and then poured onto 200 g of crushed ice. The precipitated material was collected, washed with water and then with ethanol, and recrystallized from dimethylformamide to give 2.8 g of I*b* (62%) as pale-yellow needles, mp 334–336° dec. The IR spectrum showed bands at 3300 (OH stretching), 1090 (—OCH_3), 1370 ($C_6H_5NO_2$), and 1550 (C=N) cm^{-1}. The NMR spectrum showed: *(a)* τ 7.3 (protons of the 2-CH_3 group, s), *(b)* τ 5.9 (protons of the 6-OCH_3 group, s), and *(c)* τ 2.3 and 1.7 (seven and eight aromatic protons, two doublets showing an *AB* system), indicating nitration at the 5-position.

Anal.—Calc. for $C_{11}H_9N_3O_6$: C, 47.32; H, 3.22; N, 15.05. Found: C, 47.54; H, 3.14; N, 14.85.

When a solution of I*b* in ethanol was treated with ammonia, ethylamine, diethylamine, or benzylamine, the corresponding salts were obtained.

With Ammonia—Anal.—Calc. for $C_{11}H_9N_3O_6 \cdot NH_3$: C, 44.59; H, 5.05; N, 18.92. Found: C, 44.55; H, 5.12; N, 18.88.

With Ethylamine—Anal.—Calc. for $C_{11}H_9N_3O_6 \cdot C_2H_5NH_2$: C, 48.15; H, 4.93; N, 17.28. Found: C, 48.15; H, 4.93; N, 16.99.

With Diethylamine—Anal.—Calc. for $C_{11}H_9N_3O_6 \cdot (C_2H_5)_2NH$: N, 15.90. Found: N, 15.90.

With Benzylamine—Anal.—Calc. for $C_{11}H_9N_3O_6 \cdot C_6H_5CH_2NH_2$: N, 14.51. Found: N, 14.51.

4-Chloro-3,5-dinitro-6-methoxy-2-methylquinoline (I*c*)—A mixture of 1 g (0.0036 mole) of I*b* and 5 ml of phosphorus oxychloride was refluxed for 30 hr. Excess phosphorus oxychloride was distilled off and the reaction mixture was cooled and then diluted with water. The precipitate formed after neutralization with ammonium hydroxide was filtered off, washed with water, and recrystallized from ethanol to give I*c*, 0.8 g (75%), as yellow needles, mp 210–212°.

Anal.—Calc. for $C_{11}H_8ClN_3O_5$: C, 44.37; H, 2.96; Cl, 11.93; N, 14.11. Found: C, 44.60; H, 3.30; Cl, 11.77; N, 14.23.

3,5-Dinitro-4-hydroxy-6-methoxy-2-styrylquinoline (I*d*)—A mixture of 1 g (0.0034 mole) of I*c*, 0.5 g (0.0047 mole) of benzaldehyde, and 0.01 g of fused zinc chloride was heated at 140–150° for 4 hr. The reaction mixture was cooled and treated with an alcoholic sodium hydroxide solution (20 ml, 2%). The solid product was collected, 0.5 g (41%), and recrystallized from dimethylformamide to give I*d* as colorless crystals, mp >350°.

Anal.—Calc. for $C_{18}H_{13}N_3O_6$: C, 58.86; H, 3.54; N, 11.44. Found: C, 58.32; H, 3.63; N, 11.07.

Attempted preparation of I*d* by treating I*b* with benzaldehyde in the presence of fused zinc chloride or acetic anhydride was unsuccessful.

4-Chloro-3,5-dinitro-6-methoxy-2-styrylquinoline (I*e*)—A mixture of 0.5 g (0.0014 mole) of I*d* and 5 g of phosphorus oxychloride was refluxed for 30 hr. Excess phosphorus oxychloride was distilled off and the reaction mixture was kept cold while being diluted with water and then basified with ammonium hydroxide. During continued cooling, a precipitate was formed. It was collected on a filter and washed several times with water and then with alcohol. Recrystallization from chloroform gave 0.3 g (57%) of I*e* as yellow crystals, mp 260–262°.

Anal.—Calc. for $C_{18}H_{12}ClN_3O_5$: C, 56.03; H, 3.11; Cl, 9.21; N, 10.89. Found: C, 56.26; H, 3.30; Cl, 9.21; N, 10.55.

3,5-Dinitro-4-phenoxy-6-methoxy-2-methylquinoline (I*f*)—A mixture of 0.6 g (0.002 mole) of I*c* and 0.28 g (0.003 mole) of phenol in 1 ml of dry pyridine was heated on a steam bath for 6 hr. After evolution of excess pyridine, the residue (0.5 g, 70%) was collected and recrystallized from ethanol to give I*f* as colorless needles, mp 140–141°.

Anal.—Calc. for $C_{17}H_{13}N_3O_6$: C, 57.46; H, 3.66; N, 11.83. Found: C, 57.46; H, 3.66; N, 11.47.

2-Methoxy-6-methyl-1-nitro-12*H*-quino[3,4-*b*][1,4]benzoxazine (III*a*)—A mixture of 2 g (0.0067 mole) of I*c*, 0.77 g (0.007 mole) of 2-aminophenol, and 1 g of sodium carbonate in 20 ml of ethanol was refluxed for 4 hr. Cooling and addition of water (100 ml) precipitated a material which was filtered off, washed with water and then with ethanol, and recrystallized from ethanol to give 1.2 g of III*a* (55%) as pale-yellow needles, mp 206–208°. The IR spectrum showed absorption bands at 3280 (NH-stretching) and 1590 (NH-bending) cm^{-1}. The NMR spectrum showed: *(a)* τ 7.2 (protons of the 2-CH_3 group, s), *(b)* τ 5.94 (protons of the 6-OCH_3 group, s), and *(c)* τ 2.4–3 (six aromatic protons, m).

Anal.—Calc. for $C_{17}H_{13}N_3O_4$: C, 63.16; H, 4.02; N, 13.00. Found: C, 62.76; H, 3.91; N, 12.92.

The hydrochloride salt was obtained by refluxing III*a* (0.32 g) with 15% HCl (10 ml) for 10 min. Cooling gave 0.25 g of the salt as bright-yellow needles, mp 254–256°.

Anal.—Calc. for $C_{17}H_{13}N_3O_4 \cdot HCl$: C, 56.74; H, 3.89; Cl, 9.87; N, 11.68. Found: C, 56.99; H, 3.89; Cl, 9.60; N, 11.68.

The salt regenerated the free base III*a* when gently warmed with water.

10-Chloro-2-methoxy-1-nitro-6-methyl-12*H*-quino[3,4-*b*][1,4]benzoxazine (III*b*)—This compound was prepared by the method used for III*a* from I*c* and 2-amino-4-chlorophenol to give III*b* (70%) as yellow needles from ethanol, mp 240–242°.

Anal.—Calc. for $C_{17}H_{12}ClN_3O_4$: C, 57.06; H, 3.36; Cl, 9.95; N, 11.75. Found: C, 57.20; H, 3.70; Cl, 10.00; N, 11.42.

Compounds III*a* and III*b* gave the styryl derivatives III*c* and III*d*, respectively, upon condensation with benzaldehyde in the presence of fused zinc chloride. The latter compounds were also obtained when I*e* was allowed to react with 2-aminophenol and 2-amino-4-chlorophenol, respectively.

When recrystallized from chloroform, III*c* had a melting point of 244–246°.

Anal.—Calc. for $C_{24}H_{17}N_3O_4$: C, 70.07; H, 4.13; N, 10.21. Found: C, 70.02; H, 4.13; N, 10.21.

When recrystallized from chloroform, III*d* had a melting point of 265–268°.

Anal.—Calc. for $C_{24}H_{17}ClN_3O_4$: C, 64.64; H, 3.59; N, 9.42. Found: C, 64.05; H, 3.91; N, 8.99.

In the same way, IV*a* was obtained by reacting I*c* with 2-amino-1-naphthol (60% yield). It was recrystallized from chloroform as violet crystals, mp 270–272°.

Anal.—Calc. for $C_{21}H_{15}N_3O_4$: C, 67.56; H, 4.02; N, 11.26. Found: C, 67.99; H, 3.98; N, 10.99.

Treatment of IV*a* with benzaldehyde in the presence of fused zinc chloride gave 40% yield of the styryl derivative IV*b*, mp 310–312°.

Anal.—Calc. for $C_{28}H_{19}N_3O_4$: C, 72.89; H, 4.12; N, 9.11. Found: C, 73.08; H, 4.02; N, 8.99.

3,5-Dinitro-4-(2′-hydroxyphenoxy)-6-methoxy-2-methylquinoline (V)—A mixture of 0.6 g (0.002 mole) of I*c*, 0.22 g (0.002 mole) of catechol, and 4 g (0.005 mole) of dry pyridine was heated on the steam bath for 10 hr. The residue left after evaporation of

[1] All melting points are uncorrected. The IR spectra were recorded with a Carl Zeiss Infracord spectrophotometer model UR 10 using KBr. NMR spectra were performed on a Varian A-60 spectrometer in $CDCl_3$ or CF_3COOH solution, using tetramethylsilane as an internal standard.

pyridine was extracted with ether and dried (sodium sulfate). After evaporation of the ether, the product was recrystallized from benzene to give 0.3 g (40%) of V as pale-yellow crystals, mp 128–130°.

Anal.—Calc. for $C_{17}H_{13}N_3O_7$: C, 54.98; H, 3.50; N, 11.32. Found: C, 54.80; H, 3.60; N, 11.40.

2-Methoxy-6-methyl-1-nitro[1,4]benzodioxino[2,3-*c*]quinoline (VI)—A stirred mixture of 0.37 g (0.001 mole) of V and 10 ml of an aqueous solution of sodium hydroxide (10%) was gently warmed on the steam bath for 2 hr. It was then cooled and neutralized with hydrochloric acid (pH 7). The precipitated product was recrystallized from ethanol to give 0.17 g (52%) of VI as yellowish-green needles, mp 208–210°. The IR spectrum showed the absence of an OH group. The NMR spectrum showed: *(a)* τ 7.41 (protons of the 6-CH_3 group, s), *(b)* τ 5.98 (protons of the 2-OCH_3 group, s), *(c)* τ 1.9 and 2.6 (three and four aromatic protons, two doublets showing an *AB* system), and *(d)* τ 3.02 (five aromatic protons, s).

Anal.—Calc. for $C_{17}H_{12}N_2O_5$: C, 62.96; H, 3.70; N, 8.64. Found: C, 63.06; H, 4.09; N, 8.46.

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Separation of Anthraquinone Glycosides and Aglycones Using Electropaperography

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Abstract □ A method was developed using an electropaperography apparatus for the separation of hydroxyanthraquinone derivatives, both as glycosides and as aglycones. By using this technique, a mixture of emodin, alizarin, and chrysophanic acid was separated and identified by paper chromatography. Similarly, emodin maltoside and chrysophanic acid maltoside were separated and identified, and the glycosides of senna leaflets were separated and their corresponding aglycones were identified.

Keyphrases □ Anthraquinone aglycones and corresponding glycosides—extraction and separation from plant material by electropaperography □ Senna leaflets—extraction, separation, and identification of glycosides and aglycones by electropaperography □ Electropaperography—separation and identification of anthraquinone glycosides and aglycones

For a number of years, considerable work has been done on the identification of naturally occurring hydroxyanthraquinone derivatives. Numerous methods can be found for the separation of anthraquinone derivatives in several vegetable laxative drugs. Most methods involve the use of either TLC (1) or column (2) or paper (3) chromatography. Characterizations of the anthraquinone derivatives by paper electrophoresis were first described by Core and Kirch (4) and Siesto and Bartoli (5). These workers used single strip paper electrophoresis to accomplish the separation of some hydroxyanthraquinones. The R_f value of each individual compound was determined after each run.

The principle of applying, to a flowing liquid inside a porous medium, an electric field at right angles to the central stream of the mixture to be separated was initially proposed by several investigators (6–9). This method, usually referred to as electropaperography or continuous electrophoresis, is particularly useful for the preparative separations of multicomponent mixtures. The electrolyte solution flows in a direction normal to the lines of force of the electrical field, and the mixture to be separated is added continuously at a small spot in the flowing medium. Components of the mixture are deflected in different directions according to their electrophoretic mobilities and, after passage through the entire plane of the flowing medium, can be collected continuously at various positions. Electropaperography differs from other methods in one important and fundamental respect, *i.e.*, the more slowly migrating components are not required to move over a path that has been previously traversed by the more rapidly migrating components; as a result, absolute separations can be effected. The main purpose of this research was to use this method in the separation of certain plant anthraquinone derivatives, a method that has not been reported previously for these compounds.