(magnesium sulfate), and the solvent was removed by evaporation under reduced pressure, giving the desired ketone.

This procedure was used to isolate 2-piperidinoacetophenone and gave a yield of 91%, bp 102–104° (0.2 mm Hg) [lit. (10) bp 180–181° (26 mm Hg)]; IR: ν_{max} (neat) 3050, 2925, 2840, 2790, 2747, 1684, 1672, 1590, 1570, 1440, 1374, 1290, 1274, 1250, 1230, 1210, 1190, 1168, 1150, 1120, 1100, 1069, 1030, 1012, 985, 955, 850, 740, 705, and 680 cm^{-1}; UV: λ_{max} 243 nm (ϵ 9670).

Anal.—Calc. for $C_{13}H_{17}NO$: C, 76.81; H, 8.43; N, 6.89. Found: C, 76.63; H, 8.40; N, 6.78.

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Synthesis of 5-Hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]-amino]hydantoin, a Metabolite of Dantrolene

RALPH L. WHITE [x] and THOMAS J. SCHWAN

Abstract □ The synthesis and structural elucidation of 5-hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin, a compound proposed as a metabolite of dantrolene sodium, are reported. In addition, a chromatographic comparison of the biological and synthesized materials is made.

Keyphrases □ Dantrolene metabolite—synthesis and structural elucidation of 5-hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin □ 5-Hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]-amino]hydantoin—dantrolene metabolite, synthesis and structural elucidation

Dantrolene sodium, 1-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin sodium salt hydrate[1], is a peripherally acting, novel, skeletal muscle relaxant (1–4). Metabolism of dantrolene sodium was reported to proceed through both reductive and nonreductive pathways (5). A polarographic method was developed for simultaneous determination of dantrolene (I) with its reduced and nonreduced metabolites (5).

Subsequently, a procedure was described for determining dantrolene alone and for estimating the amounts of two metabolites (6). One of these metabolites was referred to as Metabolite A (5, 6). Metabolite A now has been isolated, and evidence accumulated in these laboratories suggests that its structure is 5-hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin (II) (Scheme I). This paper is concerned with the synthesis and structural elucidation of II and its subsequent chromatographic comparison with the isolated Metabolite A.

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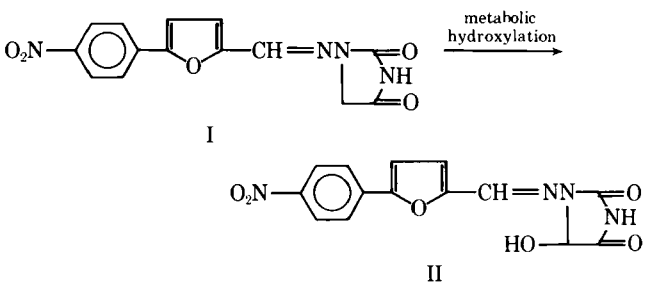
DISCUSSION

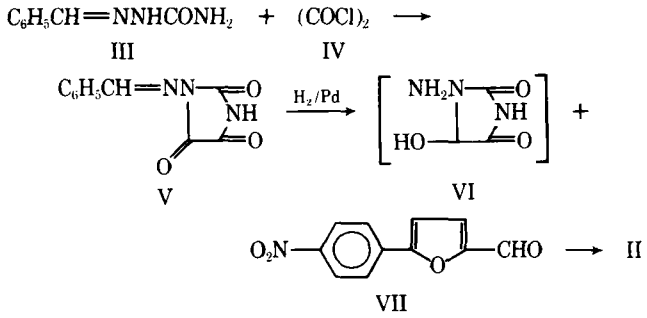
Cyclization of benzaldehyde semicarbazone (III) with oxalyl chloride (IV) allowed isolation of 1-(benzylideneamino)parabanic acid (V). By a combined catalytic hydrogenation and hydrogenolysis of V to 1-amino-5-hydroxyhydantoin (VI) and subsequent condensation of VI *in situ* with 5-(*p*-nitrophenyl)furfural (VII), II was prepared (Scheme II).

The NMR signal of the N_3—H proton (11.3 ppm) of II compared favorably with the N_3—H proton of dantrolene (11.3 ppm). In contrast, the N_1—H proton of the positional isomer 3-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin was observed at 8.3 ppm, close to the phenyl protons (7). Thus, the NMR spectrum of II supports catalytic reduction of the carbonyl of V in the 5-position rather than the 4-position; *i.e.*, II is a 1-amino-5-hydroxyhydantoin rather than a 3-amino-5-hydroxyhydantoin.

Compound II was unstable in the presence of base. When II was



Scheme I



Scheme II

dissolved in dilute aqueous sodium hydroxide solution (Scheme III), an orange solid precipitated from the solution; it was shown to be the semicarbazone (VIII) by IR comparison with an authentic sample. The hydrolysis of 5-hydroxyhydantoin to urea was reported previously (8).

To assure the identity of Metabolite A with that of synthesized II, the two materials were cochromatographed on silica gel thin-layer plates in four solvent systems. There was no separation of the two components in any system.

In summary, the synthesis of II was accomplished, and the assignment of the structure of Metabolite A as II in these laboratories was shown to be correct by cochromatography of the biological and synthesized samples. The ring opening of synthetic Metabolite A in base to 5-(*p*-nitrophenyl)furfural semicarbazone also was found.

EXPERIMENTAL[2]

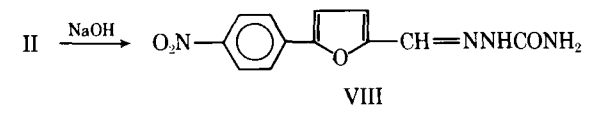
1-(Benzylideneamino)parabanic Acid (V)—In a 3-liter flask equipped with a condenser, drying tube, stirrer, and dropping funnel was placed 114 g (0.70 mole) of benzaldehyde semicarbazone in 1.0 liter of anhydrous ether. Oxalyl chloride (64 ml, 0.75 mole) was added dropwise during 0.5 hr rapidly enough to maintain a gentle reflux, and the solution then was stirred without heating for another 2.5 hr. The solid was collected by filtration and recrystallized from 3.5 liters of 2-propanol to yield 119 g (74%) of V, mp 205–207°.

Anal.—Calc. for $C_{10}H_7N_3O_3$: C, 55.30; H, 3.25; N, 19.35. Found: C, 55.19; H, 3.24; N, 19.30.

5-Hydroxy-1-[[5-(*p*-nitrophenyl)furfurylidene]amino]hydantoin (Metabolite A) (II)—In 500 ml of methanol were placed 87 g (0.40 mole) of V and 20 g of 5% palladium-on-carbon (50% moisture). Hydrogenation on a Parr apparatus gave uptake of two equivalents of hydrogen in 2 hr. An additional 20 g of catalyst was then added. Uptake of the third equivalent required 7 hr, with a total of 92% of the theoretical hydrogen uptake being observed.

The reduced solution was filtered, and the catalyst was washed with 500 ml of methanol. The combined methanolic filtrates were added to 87 g (0.40 mole) of VII (9), 2.0 liters of methanol, and 40 ml of concentrated hydrochloric acid. The mixture was refluxed for 2 hr and then filtered while hot. The collected solid was dried to give 20 g (15%) of II, mp 229–230°; NMR: δ 5.75 (d, 1H, CH, J = 9 Hz, exchange to s), 7.15 (d, 1H, furan CH, J = 3.5 Hz), 7.45 (d, 1H, OH, J = 9 Hz, exchange), 7.48 (d, 1H, furan CH, J = 3.5 Hz), 8.03 (d, 2H, phenyl CH, J = 9 Hz), 8.25 (s, 1H, azomethine CH), 8.36 (d, 2H, phenyl CH, J = 9 Hz), and 11.3 (broad s, 1H, NH, exchange); IR (potassium bromide): 3.1, 5.6, and 5.8 μm; mass spectrum M^+: *m/e* 330.

Anal.—Calc. for $C_{14}H_{10}N_4O_6$: C, 50.91; H, 3.05; N, 16.97. Found: C, 51.05; H, 3.10; N, 16.93.



Scheme III

1-[[5-(*p*-Nitrophenyl)furfurylidene]amino]hydantoin (Dantrolene) (I)—The synthesis of this compound was reported previously (1); NMR: δ 4.40 (s, 2H, CH), 7.04 (d, 1H, furan CH, J = 4 Hz), 7.42 (d, 1H, furan CH, J = 4 Hz), 7.80 (s, 1H, azomethine CH), 7.98 and 8.32 (d of d, 4H, phenyl CH, J = 9 Hz), and 11.3 (broad s, 1H, NH, exchange).

5-(*p*-Nitrophenyl)furfural Semicarbazone (VIII) from II—To II (200 mg, 0.61 mmole) was added 100 ml of 0.5% sodium hydroxide, and the orange solution was shaken occasionally over 4 hr. Solid separated during that time. The mixture was filtered, and the collected solid was allowed to dry for 1 hr and washed with ether (20 ml). The IR spectrum of this product matched that of authentic 5-(*p*-nitrophenyl)furfural semicarbazone. The yield was 67 mg (32%), mp 223–226°. No depression was observed for a mixed melting point.

5-(*p*-Nitrophenyl)furfural Semicarbazone (VIII) from VII—To VII (9) (50 g, 0.25 mole) dissolved in warm dimethylformamide was added an aqueous solution of semicarbazide hydrochloride (30 g, 0.27 mole). The resulting solution was allowed to stand for 10 min and was then poured into a large volume of water. The resulting precipitate was recrystallized from acetic acid to give VIII (36%), mp 230–232°. A second recrystallization from acetic acid gave the analytical material, mp 236–237° [lit. (10) mp 193–194°[3]].

Anal.—Calc. for $C_{12}H_{10}N_4O_4$: C, 52.55; H, 3.68; N, 20.43. Found: C, 52.49; H, 3.57; N, 20.31.

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[x] To whom inquiries should be directed.

[2] Melting points were determined on a Mel-Temp apparatus and are corrected. A Varian Associates A-60A instrument was used for NMR spectra. All spectra were run in dimethyl sulfoxide-d_6 with tetramethylsilane as the internal standard. A Perkin-Elmer model 137B recording spectrophotometer was used for IR spectra. The mass spectrum was run on a Hitachi Perkin-Elmer RMV-7 mass spectrometer at Northern Illinois University, DeKalb, Ill.

[3] No reason for the difference in melting point is apparent.