(6 H, t). MS: *m/e* (relative intensity) 280 (M^+, 0.5), 279 (4), 265 (65), 252 (25), 250 (13), 235 (16), 176 (100), 160 (21), 148 (25), 133 (30), 120 (39), 105 (76), 91 (99).

From the oxidation of **1n** the following compounds were isolated (SiO_2, hexane/ethyl acetate, 8:2).

Diethyl α-[3-(4′-Methoxyphenyl)-3-acetoxypropyl]-malonate (4n). ^1H NMR ($CDCl_3$, δ): 6.92 (d, 2 H, H_2, H_6), 7.32 (d, 2 H, H_3, H_5), 5.73 (dd, 1 H, CHOAc), 4.22 (q, 4 H, CH_2CH_3), 3.80 (s, 3 H, OCH_3), 3.38 (dd, 1 H, $CH(COOEt)_2$), 2.25–2.5 (m, 4 H, CH_2CH_2), 2.04 (s, 3 H, $OCOCH_3$), 1.28 (t, 6 H, CH_2CH_3). MS: *m/e* (relative intensity) 352 (M^+, 11), 309 (8), 265 (22), 217 (12), 192 (70), 150 (100), 137 (32), 43 (8).

1,1-Dicarbethoxy-3-acetoxy-6-methoxyindan (3n). ^1H NMR ($CDCl_3$, δ): 6.9 (dd, 1 H, H_6), 7.12 (d, 1 H, H_2), 7.25 (dd, 2 H, H_5), 6.17 (dd, 1 H, CHOAc), 3.21 and 2.71 (dd, 2 H, CH_2), 2.06 (s, 3 H, $OCOCH_3$), 1.26 (t, 6 H, CH_2CH_3). MS: *m/e* (relative intensity) 350 (M^+, 1), 308 (20), 307 (27), 291 (23), 234 (98), 217 (100), 205 (18), 189 (20), 161 (40), 145 (41), 133 (12), 43 (13).

Tetraethyl 1,6-Bis(4′-methoxyphenyl)-1,1,2,2-hexanetetracarboxylate (5n). ^1H NMR ($CDCl_3$, δ): 6.81 (d, 4 H, H_2, H_6), 7.12 (d, 4 H, H_3, H_5), 4.28 (m, 8 H, CH_2CH_3), 3.76 (s, 6 H, OCH_3), 2.64 (m, 4 H, $ArCH_2$), 2.29 (m, 4 H, $ArCH_2CH_2$), 1.31 (t, 12 H, CH_2CH_3).

Diethyl α-(4-Phenyl-4-acetoxybutyl)malonate (6) (isolated in 80% yield from **1q**). ^1H NMR ($CDCl_3$, δ): 7.46–7.25 (m, 5 H), 5.75 (t, 1 H), 4.18 (q, 4 H), 3.30 (t, 1 H), 2.18–1.5 (m, 8 H), 1.25 (t, 6 H). MS: *m/e* (relative intensity) 290 (M^+, 3), 245 (5), 216 (11), 199 (12), 173 (11), 130 (100), 129 (17), 107 (15), 91 (11), 42 (36).

From the oxidation mixture of compound **1r** the dimer **7r** was isolated in 40% yield (SiO_2, hexane/ethyl acetate 1% gradient starting from 9:1).

Tetraethyl 1,2-Bis(3-phenoxypropyl)-1,1,2,2-ethanetetracarboxylate (7r). ^1H NMR ($CDCl_3$, δ): 7.8–7.43 (m, 4 H), 7.4–7.1 (m, 6 H), 4.22 (q, 8 H), 3.98 (t, 4 H), 2.63–1.80 (m, 8 H), 1.30 (t, 12 H). ^{13}C NMR ($CDCl_3$): 169.62 (C_q, COOEt), 158.95 (CH, C_1), 129.31 (C_q, C_2, C_6), 120.54 (CH, C_4), 114.53 (CH, C_3, C_5), 62.75 (C_q, $C(COOEt)_2$), 67.84 (CH_2O), 61.56 ($COOCH_2CH_3$), 28.03 (OCH_2CH_3), 25.69 ($OCH_2CH_2CH_2$), 13.86 ($COOCH_2CH_3$).

Registry No. **1a**, 26395-09-5; **1b**, 34795-65-8; **1c**, 118598-36-0; **1d**, 118598-37-1; **1e**, 78383-16-1; **1f**, 1787-17-3; **1g**, 118598-38-2; **1h**, 118598-39-3; **1i**, 118598-40-6; **1j**, 118598-41-7; **1k**, 118598-42-8; **1l**, 6628-68-8; **1m**, 111171-90-5; **1n**, 118598-43-9; **1p**, 118598-44-0; **1q**, 78573-23-6; **1r**, 6345-89-7; **1s**, 118598-45-1; **2a**, 115860-33-8; **2b**, 118598-46-2; **2c**, 118598-47-3; **2d**, 118598-48-4; **2e**, 118598-49-5; **2f**, 118598-50-8; **2g**, 118598-51-9; **2h**, 118598-52-0; **2i**, 118598-53-1; **2j**, 118598-54-2; **2k**, 118598-55-3; **2l**, 115860-31-6; **2m**, 118598-56-4; **2m′**, 118598-57-5; **2n**, 118598-58-6; **2p**, 118598-59-7; **2p′**, 118598-60-0; **2q**, 118598-61-1; **2r**, 118598-62-2; **2s**, 118598-63-3; **3l**, 118629-54-2; **3n**, 118598-65-5; **4l**, 118598-71-3; **4n**, 118598-66-6; **5l**, 118598-64-4; **5n**, 118598-67-7; **6**, 118598-68-8; **7r**, 118598-69-9; diethyl α-(2-phenylethyl)-α-hydroxymalonate, 118598-70-2; manganese(III) acetate, 993-02-2.

Supplementary Material Available: Characterization data for **2a–s** (4 pages). Ordering information is given on any current masthead page.

Electrochemical Oxidation of 5-Hydroxytryptamine in Acidic Aqueous Solution

Monika Z. Wrona and Glenn Dryhurst*

Department of Chemistry and Biochemistry, University of Oklahoma, Norman, Oklahoma 73019

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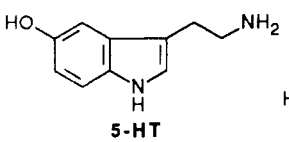
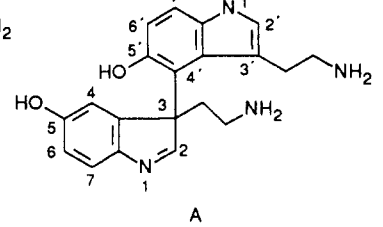
The electrochemical oxidation of 5-hydroxytryptamine (5-HT) has been studied in aqueous solution at pH 2.0 using a pyrolytic graphite electrode. Eight major products formed in the early stages of oxidation of millimolar solutions of 5-HT have been isolated and their structures elucidated by spectroscopic techniques.

The electrochemical oxidation of 5-hydroxytryptamine (5-HT) in acidic (1.05 M $HClO_4$) acetonitrile at a platinum electrode yields a single dimeric product in 80% yield. ^1H and ^{13}C NMR evidence indicates that this dimer is the asymmetric 3,4′-linked indolenine–indole A. Under the

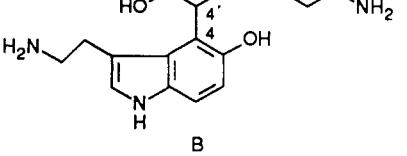


5-HT



A

latter conditions cyclic voltammetric evidence suggests that the initial electrode reaction involves a one-electron abstraction to produce a radical cation with the unpaired electron located at the C(3) position of 5-HT. Coupling of this radical with 5-HT then apparently yields a 3,4′-dimer radical cation. A disproportionation-like second electron transfer then occurs in solution between the 5-HT radical cation and the 3,4′-dimer radical cation to generate 5-HT and, following deprotonation, A. In an earlier report we showed that electrochemical oxidation of very low concentrations (≥30 μM) of 5-HT in acidic (0.01 M HCl) aqueous solution at a pyrolytic graphite electrode also proceeds through the initial formation of a radical intermediate.[2] At low applied potentials (i.e., corresponding to the rising segment of the first voltammetric oxidation peak of 5-HT), the major product is the symmetrical 4,4′-dimer (B) along with a much smaller amount of an



B

asymmetric dimer which, based primarily on ^1H NMR data, was proposed to be 5,5′-dihydroxy-1,4′-bitryptamine. At higher applied potentials the initial radical intermediate is further oxidized (1 e, 1 H^+) to a very reactive quinone imine. Nucleophilic attack by water on the latter intermediate forms 4,5-dihydroxytryptamine, which is rapidly

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oxidized (2 e, 2 H^+) to tryptamine-4,5-dione. This dione then undergoes further chemical and electrochemical reactions.[2] Direct comparison of published results of electrochemical oxidations in acidic acetonitrile and aqueous acidic solutions is difficult owing to the large differences in the concentrations of 5-HT employed. Furthermore, the electrochemical oxidation of 5-HT in aqueous solution is controlled to a significant extent by adsorption of reactant and product(s) precluding conclusive mechanistic deductions based on electroanalytical studies. Formation of asymmetric dimer A as the major product of electrochemical oxidation of millimolar concentrations of 5-HT in acetonitrile and the symmetric dimer B as the major product in very dilute aqueous solutions suggests a fundamental difference in mechanism in these two media. We have examined this difference further by investigating the electrochemical oxidation of 5-HT in the millimolar concentration range in aqueous solution at pH 2. In this preliminary report we present information about several new, structurally interesting oxidation products of 5-HT which provide additional insights into the oxidation mechanism. The structure of one dimer, reported earlier, has been corrected.

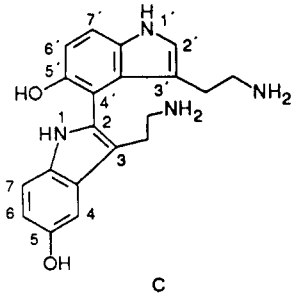
Results

Linear sweep voltammograms (5 mV s^{-1}) of 5-HT (1 mM) in aqueous 0.01 M HCl at the pyrolytic graphite electrode show three well-defined oxidation peaks. Peak potentials (E_p) for peaks I_a, II_a, and III_a are 0.45, 0.59, and 1.01 V, respectively. Because oxidation peak I_a overlapped somewhat with peak II_a, controlled potential electrolyses were carried out at $E_{p/2}$ for peak I_a in order to minimize interference from the peak II_a reaction.

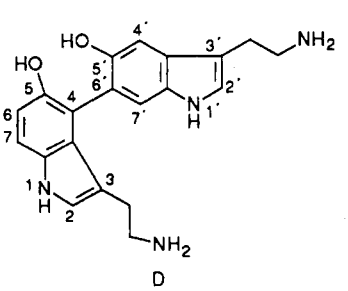
HPLC analysis of the solution formed after a few minutes electrolysis of 1 mM 5-HT showed a rather complex mixture of products (Figure 1). Longer electrolysis times resulted in even more complex mixtures as the result of secondary chemical and electrochemical reactions. In this report discussion will be limited to the eight major products formed in the early stages of the electrochemical reaction corresponding to HPLC peaks 1, 8, 10, 11, 13–15 and 17. HPLC peak 18 was due to at least three minor products, which have not yet been separated and identified.

The compounds eluted under HPLC peaks 1, 11, and 14 are formed when micromolar concentrations of 5-HT are oxidized and have been identified as B, tryptamine-4,5-dione, and 4-chloro-5-hydroxytryptamine, respectively. Structural information concerning the latter products has been presented elsewhere.[2]

The compounds eluted under HPLC peaks 8 and 10 are the asymmetric 2,4′- and 4,6′-dimers C and D, respectively. HPLC peak 17 appears to represent the major product based on the chromatogram shown in Figure 1. However,



C



D

at the detector wavelength employed (260 nm), this product, E, exhibits a very intense absorption maximum whereas all other major products exhibit absorption min-

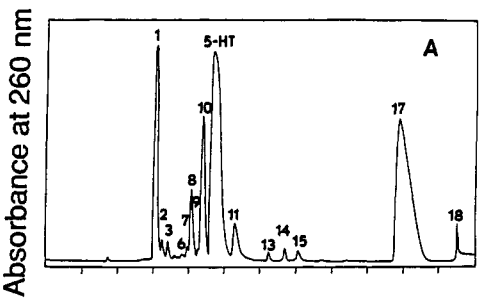
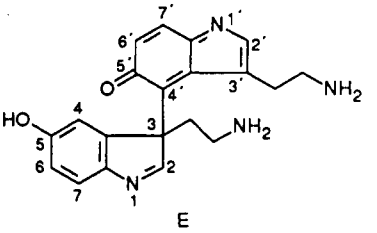


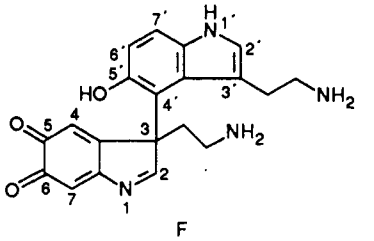
Figure 1. High-performance liquid chromatogram of the product mixture obtained after controlled potential electrochemical oxidation of 1.1 mM 5-hydroxytryptamine in 0.01 M HCl at 0.525 V for 25 min. Chromatographic conditions are given in the Experimental Section.

ima. Pale pink E was not very stable in aqueous solution and hence it was not possible to isolate this compound in a pure form. However, E shows a voltammetric reduction peak (E_p = 0.23 V) when dissolved in the HPLC mobile phase. Controlled potential reduction of E in the latter solution at −0.05 V caused its pink color to fade. HPLC



E

analysis of the colorless solution showed a single major product having the same retention time as HPLC peak 15. The reduced form of E, after desalting and freeze-drying, showed FAB-MS and ^1H NMR spectra that were identical with the asymmetric 3,4′-linked indolenine–indole dimer A. Coulometry showed that the electrochemical reduction of E (at −0.05 V) to A and the electrochemical oxidation of A to E (at 0.3 V) in 0.01 M HCl were 2 e processes. Thus, E is probably the quinone imine oxidation product of A. Additional evidence for the 3,4′ linkage in A and its oxidation product E is provided by the compound responsible of HPLC peak 13. This compound, F, is the first



F

(of several) decomposition products formed from E in acid solution and clearly contains the 3,4′ linkage between indolenine and indole residues. The exact reaction pathway from E to F remains to be determined.

Discussion

Controlled potential electrooxidation of millimolar concentrations of 5-HT in aqueous solution at pH 2 yields at least four simple dimers, A, B, C, and D. Based upon HPLC analyses the order of decreasing yield of these dimers is B > A[4] > D > C. The dimer formed in highest

(4) Compound 15 is oxidized to dimer 17 at potentials corresponding to peak I_a of 5-HT. The yield of 15 was estimated as the sum of 17 and 15 calculated on the basis of their HPLC peak heights and molar absorptivities.

yield is the 4,4′-linked compound B and all of the other fully characterized dimers have at least one 5-HT residue linked at the C(4)-position. This suggests that the predominant form of the radical generated in the initial heterogeneous one-electron abstraction from 5-HT is that where the unpaired electron is located at the C(4)-position. The fact that dimers have been isolated linked at the 4,4′-, 2,4′-, 4,6′-, and 3,4′-positions suggests that in aqueous solution the initially formed radical intermediate exists in several resonance forms. In acidic acetonitrile, however, the primary radical species that is formed is apparently that in which the unpaired electron is located at the C-(3)-position.[1] A major impetus to our studies has been to utilize information derived from electrochemical studies as a guide to understanding certain biological oxidations of 5-HT.[2] It remains to be seen if the biological oxidation chemistry is better mimicked by electrochemical oxidation in aqueous or nonaqueous media.

Experimental Section

5-Hydroxytryptamine hydrochloride (5-HT·HCl) was obtained from Sigma (St. Louis, MO). Sephadex LH-20 was obtained from Pharmacia (Piscataway, NJ). Equipment for electrochemical studies has been described elsewhere.[5] A pyrolytic graphite electrode (Pfizer Minerals, Pigments and Metals Division, Easton, PA) having an approximate surface area of 3.6 mm^2 was used for voltammetry. Controlled potential electrolyses employed two plates of pyrolytic graphite as the working electrode (total surface area ca. 70 cm^2) dipping into 40 mL of a solution containing 5-HT. All potentials are referred to the saturated calomel reference electrode (SCE) at ambiant temperature (22 ± 3 °C).

High-performance liquid chromatography (HPLC) employed a Bio-Rad gradient system and a reversed-phase column (Brownlee Laboratories, Santa Clara, CA, RP-18, 25 × 0.7 cm). Two mobile phase solvents were employed. Solvent A was prepared by adding 70 mL of MeCN and 7 mL of concentrated NH_3 to 923 mL of water. The pH of this solution was then adjusted to 3.14 by addition of concentrated HCOOH. Solvent B was prepared by adding 400 mL of MeCN and 7 mL of NH_3 to 593 mL of water. This solution was then adjusted to pH 3.5 by addition of HCOOH. For HPLC separations the following conditions were employed: 0–30 min, 100% solvent A at a flow rate of 0.7 mL min^{-1}; 30–55 min, 100% solvent A at a flow rate of 2.5 mL min^{-1}; 55–60 min, 100% solvent B at a flow rate of 3 mL min^{-1}. The eluent was monitored at 260 nm with a Gilson Holochrome UV detector.

Low- and high-resolution fast atom bombardment mass spectrometry (FAB-MS) was carried out with a VG Instruments Model ZAB-E mass spectrometer. ^1H NMR spectra (300 MHz) were obtained with a Varian Model 300 XL spectrometer. NMR spectra were obtained using Me_2SO-d_6 and D_2O as solvents, and assignments were made on the basis of spin decoupling experiments. UV–visible spectra were recorded on a Hitachi 100-80 spectrophotometer.

Isolation and Identifications of Products. In a typical preparative controlled potential electrolysis 9 mg of 5-HT·HCl was dissolved in 40 mL of 0.01 M HCl (ca. 1 mM 5-HT) contained in the working electrode compartment of a conventional three-electrode cell.[5] Electrolyses at 0.525 V were continued until HPLC analysis showed that nearly all (≥90%) 5-HT had been oxidized. At that point another 9 mg of 5-HT·HCl was added, and the electrolysis was continued. This was repeated one more time so that a total of 27 mg of 5-HT·HCl was oxidized. Two-milliliter aliquots of the resulting deep purple solution were then injected into the HPLC system. The products responsible for HPLC peaks 1, 8, 10, 11, 14, 15, and 17 (Figure 1) were collected individually as they eluted. The combined eluent for each product was then passed through a column of Sephadex LH-20 (105 × 2-cm) with H_2O/MeOH (9:1, v/v) adjusted to pH 2.0 with HCl as the mobile phase. A flow rate of 20 mL h^{-1} was employed. The latter step served to separate each product from ammonium formate. The desalted product solutions were then freeze-dried.

3-(2-Aminoethyl)-3-[3′-(2-aminoethyl)-5′-hydroxyindol-4′-yl]-5-hydroxyindolenine (A). In 0.01 M HCl the UV spectrum of **15** showed bands at λ_{max} (log ϵ_{max}) 305 (3.85), 277 (3.81), 220 sh (4.33) nm. FAB-MS (thioglycerol matrix) gave a pseudomolecular ion (MH^+) at m/e 351.1827 ($C_{20}H_{23}N_4O_4$, calcd m/e 351.1821). Thus, **15** is a simple dimer of 5-HT (MW = 350 g ($C_{20}H_{22}N_4O_2$)). ^1H NMR spectrum (Me_2SO-d_6): δ 2.42 (m, 1 H, CH_2), 2.70 (m, 3 H, CH_2), 3.52 (m, 4 H, CH_2), 6.08 (s, 1 H, C(2)-H), 6.45 (dd, $J_{6,7}$ = 8.1 Hz, $J_{4,6}$ = 2.1 Hz, 1 H, C(6)-H), 6.51 (d, $J_{6,7}$ = 8.1 Hz, 1 H, C(7)-H), 6.60 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(6′)-H), 6.82 (d, $J_{4,6}$ = 2.1 Hz, 1 H, C(4)-H), 7.16 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(7′)-H), 7.34 (d, $J_{1',2'}$ = 2.1 Hz, 1 H, C(2′)-H), 8.22 (s, 3 H, NH_3^+), 8.45 (s, 3 H, NH_3^+), 11.15 (d, $J_{1',2'}$ = 2.4 Hz, 1 H, N(1′)-H). IR spectrum of **15** (cm^{-1}): 3400 (br s, NH, OH stretches), 1610 (s, N=C stretch), 1495 and 1460 (s, aromatic C=C stretches). The ^{13}C NMR spectrum of **15** has been reported elsewhere.[1] The intense IR absorption band at 1610 cm^{-1} provides additional support for the N(1)=C(2) group and therefore for the indolenine residue in **15**.[8]

5,5′-Dihydroxy-2,4′-bitryptamine (C). In 0.01 M HCl the UV spectrum of C showed bands at λ_{max} ($\log_{10} \epsilon_{max}$[9]) 295 (4.04), 285 sh (4.01), 214 (4.49) nm. FAB-MS (3-nitrobenzyl alcohol matrix) showed an intense pseudomolecular ion (MH^+) at m/e 351. Thus, C is a dimer of 5-HT. ^1H NMR (Me_2SO-d_6): δ 2.15 (m, 4 H, CH_2CH_2), 2.43 (m, 4 H, CH_2CH_2), 6.64 (dd, $J_{6,7}$ = 8.7 Hz, $J_{6,4}$ = 2.1 Hz, 1 H, C(6)-H), 6.86 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(6′)-H), 6.92 (d, $J_{4,6}$ = 2.1 Hz, 1 H, C(4)-H), 7.13 (d, $J_{6,7}$ = 8.7 Hz, 1 H, C(7)-H), 7.16 (d, $J_{1',2'}$ = 2.1 Hz, 1 H, C(2′)-H), 7.30 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(7′)-H), 7.64 (br s, 3 H, NH_3^+), 7.91 (br s, 3 H, NH_3^+), 8.72 (br s, ~2 H, 2 OH), 10.79 (s, 1 H, N(1)-H), 10.96 (d, $J_{1',2'}$ = 2.1 Hz, 1 H, N(1′)-H). In D_2O: δ 6.89 (dd, $J_{6,7}$ = 8.7 Hz, $J_{4,6}$ = 2.1 Hz, 1 H, C(6)-H), 6.97 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(6′)-H), 7.13 (d, $J_{4,6}$ = 2.4 Hz, 1 H, C(4)-H), 7.25 (s, 1 H, C(2)-H), 7.44 (d, $J_{6,7}$ = 8.7 Hz, 1 H, C(7)-H), 7.52 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(7′)-H). These ^1H NMR spectra indicate that in dimer C one C(4) proton is missing, causing the C(6) proton in the same ring to give a doublet rather than a doublet of doublets as is the case for the other 5-HT residue. In the latter residue the signal due to the C(2) proton is absent. This is confirmed by one indolic NH proton giving a singlet resonance. Accordingly, C must be 5,5′-dihydroxy-2,4′-bitryptamine.

5,5′-Dihydroxy-4,6′-bitryptamine (D). In 0.01 M HCl the UV spectrum of D showed bands at λ_{max} ($\log_{10} \epsilon_{max}$[9]) 296 (3.94), 277 (3.93), 210 (4.44) nm. FAB-MS (3-nitrobenzyl alcohol matrix) gave an intense pseudomolecular ion (MH^+) at m/e 351 (100%). Thus D is a simple dimer of **5-HT**. ^1H NMR (Me_2SO-d_6): δ 2.42 (m, 2 H, CH_2), 3.01 (m, 2 H, CH_2), 3.08 (m, 2 H, CH_2), 3.44 (m, 2 H, CH_2), 6.77 (d, $J_{6,7}$ = 8.7 Hz, 1 H, C(6)-H), 6.98 (s, 1 H, C(4′)-H), 7.02 (s, 1 H, C(7′)-H), 7.06 (d, $J_{1,2}$ = 2.4 Hz, C(2)-H), 7.14 (s, 1 H, C(2′)-H), 7.17 (d, $J_{6,7}$ = 8.7 Hz, 1 H, C(7)-H), 7.52 (br s, 3 H, NH_3^+), 8.03 (br s, 1 H, OH), 8.12 (br s, 3 H, NH_3^+), 8.30 (br s, 1 H, OH), 10.64 (d, $J_{1',2'}$ = 2.1 Hz, 1 H, N(1′)-H), 10.72 (d, $J_{1,2}$ = 2.4 Hz, 1 H, N(1)-H). Addition of D_2O caused the signals at 7.52, 8.03, 8.12, 8.30, 10.64, and 10.72 ppm to disappear, and the doublet at 7.06 ppm to collapse into a singlet. These NMR spectra indicate that a C(4) proton in one 5-HT residue and a C(6) proton in the second residue are missing. Thus, D is 5,5′-dihydroxy-4,6′-bitryptamine.

3-(2-Aminoethyl)-3-[3′-(2-aminoethyl)-5′-oxoindol-4′-yl]-5-hydroxyindolenine (E). In 0.01 M HCl the UV–visible spectrum of E showed bands at λ_{max} ($\log_{10} \epsilon_{max}$[9]) 450 (2.70), 295 sh (3.84), 262 (4.28) nm. Cyclic voltammograms of E in 0.01 M HCl showed a quasi-reversible reduction peak at 0.27 V. Controlled potential electrochemical reduction of E immediately after it eluted from the HPLC column caused the pink solution to become colorless. HPLC analysis of the resulting solution showed a single major product having the same retention time as A. The reduced form of E, after desalting and freeze-drying, had FAB-MS and ^1H NMR spectra that were identical of A. Compound A could be quantitatively transformed to E by controlled potential

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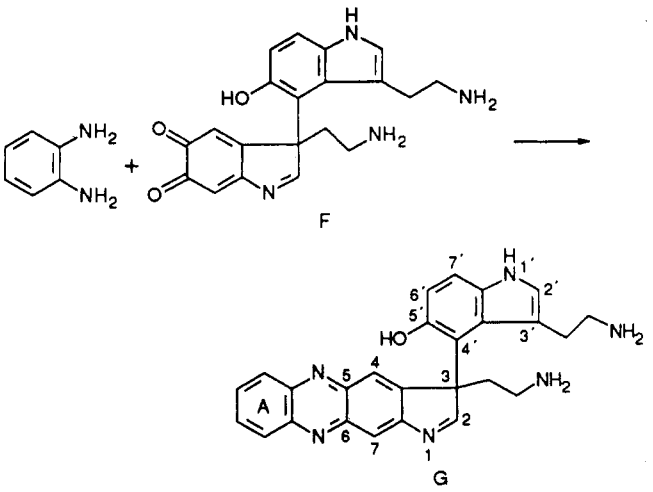
(9) Molar absorptivity values are based on the assumption that this compound was isolated as the dihydrochloride salt.

Scheme I



electrochemical oxidation at 0.30 V in 0.01 M HCl.

3-(2-Aminoethyl)-3-[3′-(2-aminoethyl)-5′-hydroxyindol-4′-yl]indolenine-5,6-dione (F). In 0.01 M HCl this bright orange compound shows absorption bands at λ_{max} = 458, 282, 215 nm. FAB-MS (thioglycerol matrix) showed intense ions at m/e 367.1770 ($C_{20}H_{23}N_4O_3$, calcd m/e 367.1770) and at m/e = 365.1614 ($C_{20}H_{21}N_4O_3$, calcd m/e = 365.1614). These results indicate that under the FAB-MS conditions employed F is partially reduced. Similar effects with other easily reducible compounds have been noted by other workers.[6,7] The FAB-MS results show that F is a dimer consisting of one residue of 5-HT and a second residue that contains an additional oxygen atom.

Compound F was not stable in either Me_2SO-d_6 or D_2O as evidenced by the gradual disappearance of ^1H NMR peaks and the appearance of many new peaks with the result that a very complex spectrum was ultimately obtained. The mass and UV–visible spectra of F along with partial interpretation of the NMR spectrum suggested that this compound possessed an *o*-quinone residue. This was confirmed by reacting F with *o*-phenylenediamine using a procedure described elsewhere.[2] After purification of the resulting product by chromatography on Sephadex LH-20[2] a stable, golden product (λ_{max} in 0.01 M HCl: 530 sh, 498, 390, 316 sh, 280, 231 nm) was obtained. FAB-MS (glycerol matrix) on this compound showed an intense pseudomolecular ion (MH^+) at m/e 437.2090 ($C_{26}H_{25}N_6O_1$, calcd m/e 437.2090). ^1H NMR (Me_2SO-d_6): δ 2.68 (m, 1 H, CH_2), 2.90 (m, 3 H, CH_2), 3.61 (m, 4 H, CH_2CH_2), 6.48 (s, 1 H, C(2)-H), 6.71 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(6′)-H), 7.01 (s, 1 H, C(4)-H), 7.26 (d, $J_{6',7'}$ = 8.7 Hz, 1 H, C(7′)-H), 7.48 (s, 1 H, C(2′)-H), 7.76 (m, 1 H, ring A), 7.86 (m, 1 H, ring A), 7.95 (s, 1 H, C(7)-H), 8.10 (m, 2 H, ring A), 8.32 (s, 3 H, NH_3^+), 8.45 (s, 3 H, NH_3^+), 9.28 (s, 1 H, OH), 11.37 (d, $J_{1',2'}$ = 2.4 Hz, N(1′)-H). Addition of D_2O caused the resonances at 8.32, 8.45, 9.28, and 11.37 to disappear. These spectral data indicate that this compound has the structure G (Scheme I). Such a structure can only be formed by condensation of *o*-phenylenediamine with dimer F, which contains a 5,6-dione residue, as illustrated in Scheme I. The ^1H NMR spectrum of G had two important features in common with that of A. Thus, in Me_2SO-d_6, the C(2) proton of G (δ 6.48) and A (δ 6.08) both show a high-field shift compared to that of 5-HT (δ 7.13). The high-field region of the ^1H NMR spectrum of A and G show an unresolved multiplet centered at δ 3.52 and δ 3.61, respectively, corresponding to the ethylamino side chain of a 5-HT residue. In addition, both compounds show complex systems for the other side chain. For A these appear as δ 2.42 (m, 1 H) and 2.70 (m, 3 H) and for B δ 2.68 (m, 1 H) and 2.90 (m, 3 H). These systems are consistent with the presence of chiral centers at the C(3)-positions in both compounds.

The structures A–G described above are based upon spectral methods of analysis. Establishment of these structures beyond any doubt would require independent synthesis.

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Theoretical Studies on 2- and 3-Azacyclopentadienylidene. An Analysis of Aromatic versus Nonaromatic π-Systems and Nonbonded-Pair Interactions

Daniel J. Pasto

Department of Chemistry, University of Notre Dame, Notre Dame, Indiana 46556

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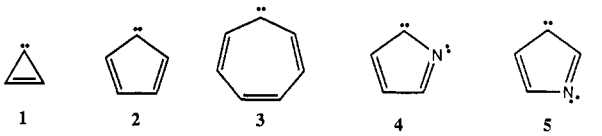
High-level ab initio calculations have been carried out on the various electronic configurations of the 2- and 3-azacyclopentadienylidenes **4** and **5**. Calculations have been carried out at the 6-31G* level with full geometry optimization with single point calculations being carried out at the MP2/6-31G* level. In each system the lowest energy configuration is calculated to be the five π-electron, σ,π triplet state having a $1b_1^2,2b_1^2,1a_2^1$ π configuration. The six π-electron, closed-shell states are very close in energy and, in fact, may be lower in energy than the pure triplet states. The LUMO's of the six π-electron, closed-shell species have a dominant contribution from the vacant σ orbital on the carbene carbon atom and are very low in energy (~+1.1 eV). The results of these calculations suggest that the lowest energy derivatives of **4** and **5** should exhibit chemistry typical of either, or both, triplet diradical and highly reactive electrophilic closed-shell singlet species. This is consistent with various experimental observations. The four π-electron, closed-shell singlet states are considerably higher in energy than the six π-electron, closed-shell singlet states. The highest-energy states are calculated to be the four and six π-electron, open-shell triplets states which are the triplet excited states of the four and six π-electron closed-shell singlet states. In the lowest energy states, the configurations of **4** are calculated to be lower in energy, while in the higher energy states, the configurations of **5** are calculated to be lower in energy. These energy differences are interpreted in terms of stabilizing delocalization effects and destablilzing electron repulsion effects.

Introduction

Cyclic π carbenes such as **1-3** have received considerable attention both theoretically and experimentally. The lowest member of the series, cyclopropenylidene (**1**), has been subjected to a number of theoretical infestigations. High-level ab initio calculations on the closed-shell singlet and open-shell triplet states of **1** using the Huzinaga–Dunning double-ζ (DZ) and double-ζ plus polarization (DZ + P) basis sets indicated that the $\pi^2\sigma^2$ singlet $^1A_1$ state lies



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