intensity) 369 (1.1, M^+), 207 (24), 192 (100), 135 (98), 107 (24). Anal. (HRMS) Calcd for $C_{22}H_{27}NO_4$: 369.1938. Found: 369.1929.

4-*O*-(2-Aminoethyl)hexestrol (6). A mixture of the amide **5** (0.2 g) in 5 mL of 1 M B_2H_6 in THF was refluxed for 16 h, at which time TLC indicated the absence of starting material. Methanol was added and evaporated to remove boric acid as methyl borate. This procedure was repeated five times, and the residue was triturated with EtOAc and dried over P_2O_5 to give product **6** (0.12 g, 60%): mp 205–207 °C; TLC, R_f (EtOAc) 0.00, R_f (MeOH) 0.14; IR (Nujol) ν_{max} 2900–3500 (OH, NH_2) cm^{-1}, absence of amide band; MS, m/z (relative intensity) 178 (12), 135 (100), 107 (41). The diacetate of **6** was prepared: mp 134–135 °C; MS, m/z (relative intensity) 397 (0.04, M^+), 354 (0.5), 177 (37), 135 (100). Anal. ($C_{24}H_{31}NO_4$) C, H, N.

4-*O*-[2-(Carbethoxyamino)ethyl]hexestrol (7). To a mixture of the amine **6** (16 mg) in THF (2 mL) and pyridine (1 mL) was added ethyl chloroformate (4.5 mg, 1 equiv) in THF (0.45 mL). After the mixture was stirred for 5 min at 25 °C, the solvent was evaporated. The residue was taken up in EtOAc and processed as described for **2**. An analytical sample was purified on HPLC (E_t = 11.2 min): mp 88–89 °C; TLC, R_f (system I) 0.20, R_f (system II) 0.66; ^1H NMR ($CDCl_3$) δ 0.52 (t, J = 7 Hz, 6, $CHCH_2CH_3$), 1.16–1.42 (m, 7, OCH_2CH_3 and CCH_2CH_3), 2.44–2.47 (m, 2, CH) 3.55–3.62 (q, J = 5 Hz, 2, OCH_2CH_2N), 4.01–4.05 (t, J = 5 Hz, 2, OCH_2CH_2N), 4.09–4.17 (q, J = 7 Hz, 2, $COOCH_2$), 6.76–6.85 (dd, J = 8 Hz, 4, Ar H), 6.99–7.07 (dd, J = 8 Hz, 4, Ar H); MS, m/z (relative intensity) 385 (2, M^+), 340 (2), 250 (90), 204 (30), 135 (100). Anal. ($C_{23}H_{31}NO_4$) C, H, N.

C-11 Labeled 2 and 7. The $^{11}COCl_2$ was collected in EtOH for a period of 10 min to yield 38–65 mCi. To this was added a solution of the amine **1** or **6** (25 μg in EtOH) containing 1% Et_3N. The reaction mixture was left at room temperature for 10 min and injected on the HPLC column, and the radioactive peak of [^{11}C]**2** or [^{11}C]**7** was collected. The elution solvent was evaporated under a stream of helium, and the product redissolved in ethanol. Radiochemical purity was verified by TLC in both solvent systems I and II. Specific activities obtained varied from 4 to 10 Ci/mmol for [^{11}C]**2** and from 1 to 2.5 Ci/mmol for [^{11}C]**7** after HPLC purification. The nondecay-corrected radiochemical yields (after HPLC purification, about 20 min after the end of bombardment) were usually 5–10% for [^{11}C]**2** and were occasionally as low as 0.5–1.0%. In the case of the [^{11}C]**7**, the yields varied between 1 and 2%.

Estrogen Receptor Binding Assay. The affinity of the estradiol and hexestrol derivatives for estrogen receptors was determined by competition studies using the Dextran-coated charcoal method.[21] Cytosol was prepared from calf uteri. Tissue samples were homogenized at 4 °C in 5 mM phosphate buffer, pH 7.4, containing 10 mM thioglycerol and 10% glycerol. After centrifugation (1 h at 105000g), the supernatant was diluted with phosphate buffer to 1–1.8 mg of protein/mL. The [^{3}H]estradiol (NEN, 130–170 Ci/mmol) was used as the radioligand at 10^{-9} M in the final incubation mixture. Test compounds were used at seven different concentrations, from 10^{-11}–10^{-5} M. Incubations were performed at 4 °C for 4 h, in triplicate. Radioactivity in charcoal-treated incubation supernatants (0.5 mL) was mixed with 10 mL of toluene-based scintillation fluid (4.0 g of PPO, 0.05 g of POPOP, 1 L of toluene) and was measured in a Searle Analytic 92 apparatus with a counting efficiency of 50%. Additional concentrations of test compounds were studied about the 50% competition point in order to obtain a graphic estimate of the concentration required for 50% displacement of the radioligand from the receptor. The RBA value of a competitor was established by using the ratio of unlabeled estradiol concentration required for 50% receptor displacement of the corresponding [^{3}H]estradiol and the competitor concentration required for the same effect, multiplied by 100.[22]

In Vivo Studies. Immature female Fisher rats (3–4 weeks old, ca. 50 g) were used to study tissue uptake of [^{11}C]**2** and [^{11}C]**7**. Solutions of 0.2–0.3 mL containing 1–10 μCi (corresponding to a mass of about 1 pmol) of these radioactive compounds were injected via the caudal vein. If designed to show the blockage of specific receptor uptake, the solution was fortified with 18 μg of nonradiolabeled estradiol. The animals were killed by cardiac puncture 10 min after injection, and tissue samples were taken and counted for radioactivity. The animals were paired in such a manner that always two of them were injected simultaneously with the same ^{11}C-labeled estrogen preparation, one without and one with carrier estradiol. The activity distribution in untreated animals and animals treated with estradiol was compared by the paired Student's t tests. A probability value of $p < 0.05$ was considered as significant.

Acknowledgment. We are grateful to the Medical Research Council of Canada for financial support and for providing one of us (G.W.) with a visiting scientist award.

Registry No. **1**, 20989-33-7; **2**, 88803-29-6; [^{11}C]**2**, 88803-30-9; **3**, 84-16-2; **4**, 88803-31-0; **5**, 88803-32-1; **6**, 88803-33-2; **7**, 88803-34-3; [^{11}C]**7**, 88803-35-4; **8**, 50-28-2; **9**, 53-16-7; **10**, 5982-51-4; [^{11}C]ethyl chloroformate, 80495-06-3.

Synthesis and Physicochemical and Neurotoxicity Studies of 1-(4-Substituted-2,5-dihydroxyphenyl)-2-aminoethane Analogues of 6-Hydroxydopamine

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In an attempt to evaluate the possible relationship between the neurotoxicity of 6-hydroxydopamine and the redox properties and electrophilic reactivity of the 6-hydroxydopamine–*p*-hydroquinone/*p*-quinone system, we have synthesized a series of 6-hydroxydopamine analogues in which the C_4-hydroxy group is replaced with various electron-donating and electron-withdrawing substituents. With the aid of cyclic voltammetry, the formal oxidation potentials ($E^{\circ\prime}$) for the *p*-hydroquinone/*p*-quinone redox couples and the rates of cyclization of the *p*-quinones to the corresponding *p*-iminoquinones were determined. As expected, electron-rich *p*-hydroquinones were easily oxidized to the *p*-quinones, which underwent cyclization slowly, whereas the oxidation of electron-poor *p*-hydroquinones required higher voltages and yielded *p*-quinones, which cyclized readily at pH 7.4. The neurotoxic potential of these compounds showed that in vivo destruction of noradrenergic terminals, as measured by inhibition of norepinephrine uptake by rat heart slices, occurred only with those analogues bearing electron-donating substituents. Potent neurotoxic properties were associated only with the 4-amino and 4-hydroxy derivatives, both of which form *p*-quinones, which do not cyclize readily at pH 7.4. These results support the thesis that the *p*-quinone derived from 6-hydroxydopamine may be an important species in the mediation of the neurodestruction caused by 6-hydrodopamine.

The dopamine oxidation product 1-(2,4,5-trihydroxyphenyl)-2-aminoethane [6-hydroxydopamine (6-OHDA, **1**)] is an effective sympatholytic agent[1,2] that selectively destroys noradrenergic and dopaminergic nerve terminals.[3,4]

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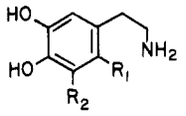
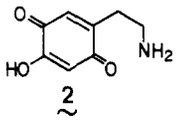
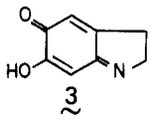
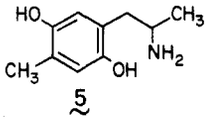




1: R_1=OH, R_2=H

4: R_1=H, R_2=OH

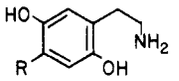




Although the precise molecular mechanisms underlying the neurodegenerative properties of 6-OHDA are not well established, it is generally accepted that the process is mediated by oxidation of the *p*-hydroquinone functionality to the corresponding *p*-quinone **2** and/or products derived from compound **2**, such as the *p*-iminoquinone **3**. Such electrophilic oxidation products alkylate nucleophilic functionalities present on macromolecules, a process that may be the initial step leading to destruction of the nerve terminals. The ease of oxidation of 6-OHDA, especially at physiological pH,[5] and the electrophilic properties of *p*-quinones/*p*-iminoquinones[6] support this proposal. Furthermore, administration of radiolabeled 6-OHDA to rats leads to extensive and irreversible binding of the label to spleen and heart tissues.[2] Several studies have confirmed that the covalent interaction of one or more of these oxidation products with intraneuronal proteins can be correlated with the neurodegeneration produced by 6-OHDA.[4,7,8] Similar covalent binding is observed in incubates of radiolabeled 6-OHDA with bovine serum albumin, as well as other model proteins.[9-11] Finally, the catecholamine *O*-methyltransferase inhibiting properties of 6-OHDA and analogues are thought to be dependent on similar pathways and may involve cyclic species at higher oxidation states than the *p*-quinone/*p*-iminoquinone systems.[12,13]

In an effort to characterize further the chemical events responsible for the neurotoxicity of 6-OHDA, we have undertaken the synthesis and analysis of the chemical and neurotoxic properties of a selected series of 1-(4-substituted-2,5-dihydroxyphenyl)-2-aminoethane analogues of 6-OHDA. These compounds retain the *p*-hydroquinone

Chart I. Structures of 6-OHDA and Analogues

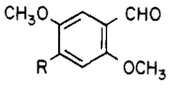


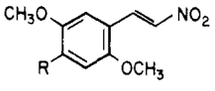
1, R = OH	10, R = NH_2
6, R = H	11, R = Br
7, R = CH_3	12, R = CN
8, R = OCH_3	13, R = COOH
9, R = NO_2	14, R = Cl

functionality present in 6-OHDA. The absence of cytotoxic properties in the isomeric 5-OHDA analogue **4**[14] and the retention of such properties in 1-(2,5-dihydroxy-4-methylphenyl)-2-aminopropane (**5**)[15] suggest that the *p*-hydroquinone group may be important to the neurotoxic properties of 6-OHDA.

The series of compounds prepared (Chart I) includes structures bearing a wide spectrum of electron-withdrawing and electron-donating groups at the C_4 position. It was anticipated that the electronic characteristics of the C_4 substituents would affect the ease with which these 6-OHDA analogues undergo oxidation, as well as the electrophilicity and, hence, potential alkylating properties of the corresponding *p*-quinones (and *p*-iminoquinones). With the aid of electrochemical and norepinephrine reuptake inhibition studies, we have compared the ease of *p*-hydroquinone oxidation and rates of *p*-quinone cyclization with the neurotoxic properties of the compounds listed in Chart I. The results suggest that the *p*-quinone **2** may be the principal species responsible for the neurodegenerative properties of 6-OHDA.

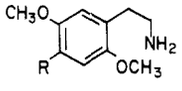
Synthesis. The majority of the compounds listed in Chart I were prepared from commercially available 2,5-dimethoxybenzaldehyde (**6a**), which was readily converted

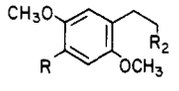




	R
6a	H
7a	CH_3
8a	OCH_3

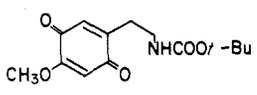
	R
6b	H
7b	CH_3
8b	OCH_3



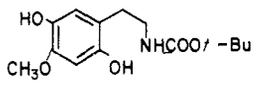


	R
6c	H
7c	CH_3
8c	OCH_3
11c	Br
12c	CN
13c	COOH
14c	Cl

	R	R_2
8d	CH_3O	NH-*t*-butoxy
9d	NO_2	NH-Ac
10d	NH_2	NH-Ac
11d	Br	N-phthalimide
12d	CN	N-phthalimide
14d	Cl	N-phthalimide



8e



8f

to 1-(2,5-dimethoxyphenyl)-2-aminoethane (**6c**) via the intermediate 1-(2,5-dimethoxyphenyl)-2-nitroethene (**6b**).

(1) Tranzer, J. P.; Thoenen, H. *Experientia* **1968**, *24*, 155.
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Bromination of **6c** proceeded regiospecifically to generate the corresponding 4-bromo derivative **11c**. Following protection of the amino functionality as its phthalimido derivative **11d**, reaction with cuprous cyanide and cuprous chloride according to the procedure of Friedman and Shechter[16,17] gave the corresponding phthalimido cyano and chloro products **12d** and **14d**, respectively. Cleavage of the phthalimido group present in **14d** to yield **14c** was readily accomplished by reaction with hydrazine. The corresponding reaction with the cyano intermediate **12d** required careful monitoring to avoid attack of the cyano group by hydrazine. Base hydrolysis of the cyano group present in **12c** provide the corresponding C_4-carbonyl intermediate **13c**. The dimethyl ether groups present in **6c**, **11c**, **12c**, **13c**, and **14c**, were readily cleaved by treatment with BBr_3 to provide the desired *p*-hydroquinones **6**, **11**, **12**, **13**, and **14**, respectively.

Nitration of 1-(2,5-dimethoxyphenyl)-2-aminoethane (**6c**) with nitric acid in acetic anhydride and acetic acid yielded the 4-nitro intermediate as its acetamido derivative **9d**. Reduction of **9d** with granular tin in concentrated HCl gave the corresponding anilino compound **10d**. The *N*-acetyl and *O*-methyl groups of these two intermediates were cleaved simultaneously by heating in 48% HBr to give the *p*-hydroquinones **9** and **10**, respectively.

Synthesis of the 4-methyl analogue **7** was achieved by a slightly modified route. Vilsmeier formylation of 2,5-dimethoxytoluene (**15**) proceeded regiospecifically to yield 2,5-dimethoxy-4-methylbenzaldehyde (**7a**). Elaboration of the side chain was accomplished by reaction of **7a** with nitromethane in the presence of ammonium acetate, followed by reduction of the resulting 1-(2,5-dimethoxyphenyl)-2-nitroethene (**7b**) with $LiAlH_4$ to the corresponding aminoethane derivative **7c**. Reaction of **7c** with BBr_3 provided the desired C_4-methyl analogue **7**.

The synthesis of the C_4-methoxy compound **8** has been reported.[18] Since the overall yield in this synthesis was extremely low, an alternate and more efficient route was sought. A synthetic scheme had to be devised that would leave intact the methyl ether group at C_4 while providing the *p*-hydroquinone moiety. The selective oxidation of 1,4-dimethoxybenzene derivatives with ceric ammonium nitrate has been reported to yield the corresponding *p*-quinones.[19] This oxidative cleavage reaction was performed on the *tert*-butoxycarbonyl derivative **8d** to give the *p*-quinone intermediate **8e**. Reduction of **8e** with sodium dithionite to **8f**, followed by acid cleavage of the *tert*-butoxycarbonyl protecting group, yielded the desired C_4-methoxy analogue **8**.

For the most part, all of the reactions proceeded in acceptable yields, except some of those generating the *p*-hydroquinone products. Because of the ease of oxidative polymerization of those *p*-hydroquinones bearing electron-donating substituents at C_4, the isolation and purification steps had to be carried out at low pH. This was achieved by cation-exchange chromatography using dilute HCl to load the resin and 4 N HCl to elute the product. The final products could be crystallized from ethanol as their hydrochloride salts, which proved to be reasonably stable when stored in the cold. All new compounds were fully characterized spectroscopically and by elemental analysis.

Electrochemical Studies. With the aid of cyclic voltammetric techniques,[20] we have examined the redox chemical behavior of the *p*-hydroquinones listed in Chart I. The working electrode employed in our studies was a thin-layer platinum electrode that, because of its design, confines the entire reactant within a 10^{-3}-cm layer at the electrode surface.[21] As a result, each reactant molecule has access to the electrode surface. This eliminates diffusion-controlled mass transfer equilibria generally associated with conventional electrodes.[22] The attractive features of this technique include high sensitivity and the ease of achieving steady-state conditions at the electrode surface.

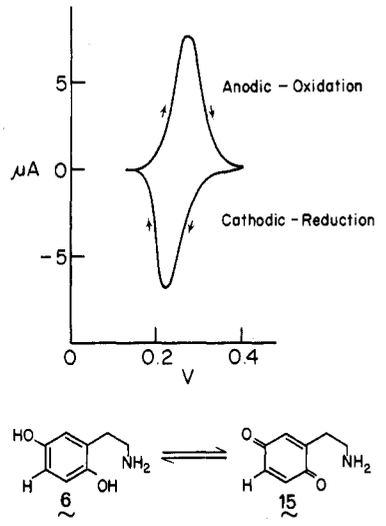

Figure 1. Upper: cyclic voltammogram of compound **6** (0.25 mM) in Robinson–Britton buffer at pH 3 with a scan rate of 2 mV/s. Lower: structural changes occurring during cyclic voltammetry.

Typical cyclic voltammograms of the *p*-hydroquinones are illustrated by the scans of the C_4-protio analogue **6** obtained at pH 3 and 7.4, shown in Figures 1 and 2, respectively. At low pH, a single redox couple associated with the *p*-hydroquinone/*p*-quinone (**6** ⇄ **15**) system is observed. The forward, anodic sweep (toward the more positive potential) generates a peak at +0.27 V, which corresponds to the oxidation of the parent *p*-hydroquinone to the corresponding *p*-quinone. On the reverse, cathodic sweep (toward the more negative potential), a peak at +0.23 V observed, which corresponds to the reduction of the *p*-quinone to the *p*-hydroquinone. The formal oxidation potential of this compound at pH 3 is +0.25 V (midpoint between the two peak potentials). From the unity ratio of the cathodic and anodic peak currents, one can conclude that this is a reversible redox process.[23] At pH 7.4, the *p*-quinone undergoes a chemical follow-up reaction (Figure 2). At a scan rate of 10 mV/s (Figure 2a), the first forward sweep displays the initial oxidation of the *p*-hydroquinone **6** to the corresponding *p*-quinone **15** (peak A) with a peak potential of +0.07 V. On the reverse sweep, reduction of the remaining *p*-quinone is observed at 0.00 V (peak B). There is, however, an additional reduction peak at −0.13 V (peak C). On the second forward sweep, a peak at −0.07 V (peak D) is detected. This new oxidation peak was not present on the first sweep. The redox system represented by peaks C and D can be identified with the

(16) Friedman, L.; Shechter, H. *J. Org. Chem.* **1961**, *26*, 2522.
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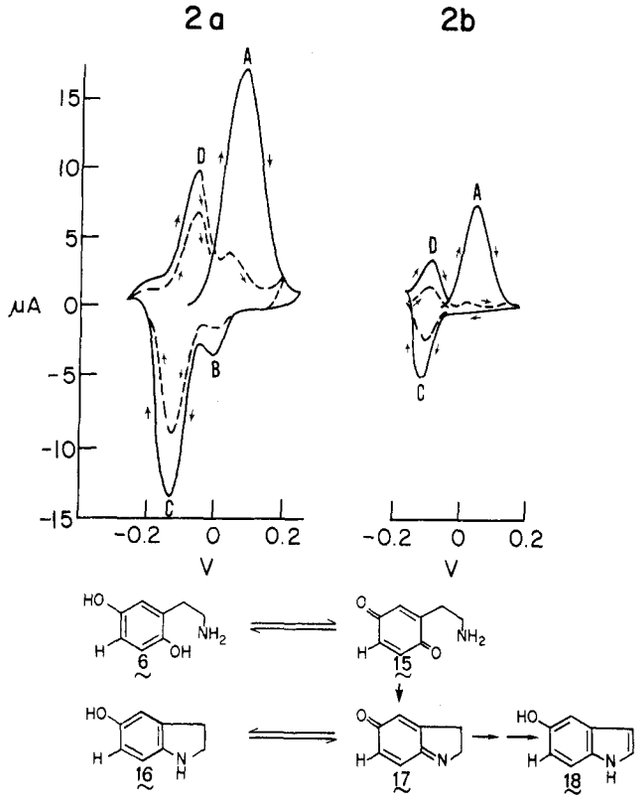

Figure 2. Upper left (2a): cyclic voltammogram of compound **6** (0.25 mM) in Robinson–Britton buffer at pH 7.4 at a scan rate of 10 mV/s: (—) first sweep; (- - -) second sweep. Upper right (2b): cyclic voltammogram of compound **6** (0.25 mM) in Robinson–Britton buffer at pH 7.4 at a scan rate of 2 mV/s. Lower: structural changes occurring during cyclic voltammetry.

indoline (**16**)–p-iminoquinone (**17**) redox couple. The cyclization reaction probably is irreversible, since the concentration of starting *p*-hydroquinone at the electrode surface decreases with time. This is reflected in the large decrease in intensity of peak A on the second forward sweep. Subsequent cycles under the same conditions give basically the same pattern of curves, with the exception that all of the peaks gradually decrease in intensity because of the loss of electroactive species. This loss probably is due to rearrangement of the *p*-iminoquinone to the corresponding indole **18**. Redox processes involving indole **18** are not observed at these particular sweep potential intervals. Figure 2b shows the cyclic voltammogram of compound **6** at the slower scan rate of 2 mV/s. Peak B (reduction of the *p*-quinone) is hardly detectable in this tracing because at this slower scan rate the longer time period between the anodic (oxidative) and cathodic (reductive) scans allows for complete cyclization of the intermediate *p*-quinone formed during the forward (oxidative) scan. The formal oxidation potentials ($E°'$) of these analogues obtained from the cyclic voltammetric plots are listed in the second column of Table I.

Thin-layer chronopotentiometry[24] was employed to measure the relative rates of cyclization of the *p*-quinone analogues to the corresponding *p*-iminoquinones. In this procedure, the voltage is set at an oxidative potential (E_{ox}; see Figure 2a and Table I) that will oxidize the *p*-hydroquinone to the *p*-quinone. The auxiliary electrode then is disconnected for a delay period of t seconds (varies from 1.5 to 40 s, depending on approximate cyclization rates), which results in the disruption of the redox chemical reaction. However, cyclization can still proceed. After this delay period, the auxiliary electrode is reconnected, and the potential is reversed by setting the voltage to the resting potential (E_{rest}; see Figure 2a and Table I) to reduce the previously electrogenerated *p*-quinone. The measured current corresponds to the amount of *p*-quinone originally formed that has not undergone cyclization. As the delay period increases, more of the *p*-quinone undergoes cyclization. These data then can be fitted to a first-order log plot (that is, plotting log *p*-quinone concentration vs. time) to obtain a straight line for this first-order cyclization reaction. Figure 3 depicts the log plot for the cyclization of the *p*-quinone derived from the C_4-bromo analogue **11**. The observed cyclization rates for the various *p*-quinone analogues are summarized in Table I. The cyclization reactions of the C_4-amino- and C_4-hydroxy-*p*-quinone derivatives proceed too slowly to measure, whereas the cyclization reactions of the C_4-cyano- and C_4-nitro-*p*-quinone derivatives proceed too rapidly to measure within the time frame used in these experiments.

Table I. Formal Oxidation Potentials ($E°'$), and Oxidation (E_{ox}) and Resting (E_{rest}) Potentials Used in the Measurements of the *p*-Quinone Cyclization Rates (K_{cyc})[a]

C_4	formal $E°'$, V	$E°'$,[b] V	E_{rest},[b] V	$K_{cyc} \times 10^{-2}$ s^{-1}
NH_2	−0.150	*c*	*c*	*c*
OH	−0.140	*c*	*c*	*c*
OCH_3	−0.082	+0.05	−0.14	0.45
CH_3	−0.042	+0.14	−0.10	1.89
Cl	−0.035	+0.15	−0.05	2.33
Br	−0.030	+0.14	−0.03	2.96
H	−0.010	+0.18	−0.06	6.30
COOH	+0.100	+0.28	+0.08	8.59
CN	+0.122	*d*	*d*	*d*
NO_2	+0.260	*d*	*d*	*d*

[a] Measurements were made in Britton–Robinson buffer at pH 7.4. [b] Potentials used in rate measurements. [c] Cyclization rates too slow to be measured. [d] Cyclization rates too fast to be measured.

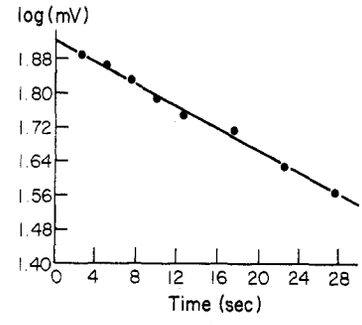

Figure 3. Log concentration of *p*-quinone vs. time plot for C_4-bromo analogue **11** in Britton–Robinson buffer at pH 7.4.

Neurotoxicity Studies. Animal studies were undertaken in order to evaluate the relative neurotoxicity of the various 6-OHDA analogues. The percent inhibition of tritiated norepinephrine (NE) uptake by heart atria isolated from rats previously administered the test compounds was used as an index of neurotoxicity. This technique is based on the observation that once noradrenergic neurons are destroyed, they lose their ability to take up NE. This assay, which is well established for 6-OHDA,[25] provides a direct measurement of the extent of neurodegeneration caused by the test compound.

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Table II. Percent Norepinephrine Uptake Inhibition (Facilitation) in Rat Atria Caused by 6-OHDA and Analogues[a]

	% NE uptake inhibn at the following doses								
C_4	730 μmol/kg	486 μmol/kg	365 μmol/kg	243 μmol/kg	122 μmol/kg	97 μmol/kg	73 μmol/kg	49 μmol/kg	24 μmol/kg
NH_2	*b*	*b*	*b*	*b*	*b*	91 ± 2	80 ± 3	60 ± 1	20 ± 8
OH		87 ± 5		85 ± 5	79 ± 1	79 ± 4	74 ± 3	68 ± 4	45 ± 20
OCH_3	*b*	*b*	26 ± 20						
CH_3	*b*	*b*	24 ± 12	27 ± 3	19 ± 4		16 ± 5		
Br	(19 ± 1)	(15 ± 5)		10 ± 2	0				
Cl	(10 ± 2)	0							
H		0							
COOH		0							
CN		0							
NO_2		0							

[a] Each value represents the mean plus or minus the SD of 12 measurements (four measurements per animal). [b] Rats expired within 30 min after injection.

Twenty-four hours following intravenous administration of the drug, the rats were sacrificed, and the atria were removed, sectioned, and incubated at pH 7.4 and 37 °C with tritium-labeled NE. After sonication and centrifugation, the supernatant fraction was counted for released radioactivity. The data obtained from these studies are summarized in Table II.

Results and Discussion

In an effort to gain insight into the contributions the oxidation products of 6-OHDA may make to the neurodegenerative process, we have synthesized a series of C_4-substituted *p*-hydroquinone analogues of 6-OHDA (Chart I) that exhibit a wide range of oxidation potentials and that yield *p*-quinones and *p*-iminoquinones displaying varying degrees of electrophilic reactivity. Table I summarizes the results from the electrochemical studies, which established the formal potentials ($E^{\circ\prime}$) for the oxidation of the *p*-hydroquinones to the corresponding *p*-quinones. The relative ease of oxidation of the analogues is in accordance with the electronic effects of the C_4 substituents. Electron-donating groups facilitate oxidation ($E^{\circ\prime}$ more negative), whereas electron-withdrawing groups retard this process ($E^{\circ\prime}$ more positive).

The observed rates of cyclization of the *p*-quinones at pH 7.4 are presented in the last column of Table I. As expected, the rates increase with increasing electron-withdrawing character of the C_4 substituents. The C_4-amino- and C_4-hydroxy-*p*-quinone derivatives cyclize too slowly to measure, whereas the C_4-cyano- and C_4-nitro-*p*-quinone derivatives cyclize too rapidly to measure within the time frame used in the experiment.

Table II summarizes the neurotoxicity data. Although all of these analogues are structural variants only at C_4, differences in metabolism, biodisposition and affinity for the neuronal membrane pump may lead to significant differences in intraneuronal concentrations. Therefore, the structure–activity correlations described below must be considered tentative.

The C_4-amino analogue appears to be the most toxic compound of the series. Rats expired shortly after administration of this compound at doses as low as 122 μmol/kg. This compound also is a very potent neurotoxin. At the 97 μmol/kg dose, 91% inhibition of NE uptake was observed. The dose of 6-OHDA required for 87% inhibition is almost five times this concentration (486 μmol/kg). Based on the activity of the C_4-amino compound, neurotoxicity does not appear to be limited to catecholamines as such. The C_4-methoxy and C_4-methyl analogues also display neurotoxic activity, although this activity is considerably less than that observed with 6-OHDA and the C_4-amino compound. Complete dose–response curves could not be obtained for these compounds because of their acute toxicity.

The C_4-chloro- and C_4-bromo analogues show very similar biological activity and chemical properties. They have almost identical $E^{\circ\prime}$ values and similar rates of cyclization of the corresponding *p*-quinones. The facilitation of NE uptake exhibited by these two 6-OHDA analogues suggests that they displace NE from its storage sites but, presumably because of their high redox potentials, are not converted extensively to the neurotoxic quinone species. On the other hand, the absence of facilitation of NE uptake by the protio, carboxy, cyano, and nitro analogues (all of which display higher formal oxidation potentials) implies that other factors (metabolism, biodisposition, and inaccessibility to NE storage sites) contribute to the in vivo interaction of these compounds with NE. The rapid cyclization rates of the *p*-quinones derived from these amines also should be noted. Even if they were to reach neuronal storage sites and undergo oxidation, the rapid cyclization of the resulting quinones would preclude the presence of high concentrations of the putative neurotoxic *p*-quinones.

In summary, our data suggest that ease of oxidation may be an important factor in the neurodegeneration caused by these compounds, since only those analogues that are readily oxidized (6-OHDA and the C_4-amino compound) are potent neurotoxins. Additionally, the resulting *p*-quinone species may occupy a central role in these events, since the *p*-quinones derived from the neurotoxic 6-OHDA and C_4-amino analogue also undergo cyclization very slowly, whereas the *p*-quinones derived from the other analogues cyclize rapidly at pH 7.4. In a similar vein, Borchardt et al.[13] concluded that inhibition of catechol *O*-methyltransferase by 6-aminodopamine was due predominantely to active-site alkylation of the enzyme by 6-aminodopaquinone. Additionally, Graham[26] has attributed the greater toxicity of dopamine (vs. norepinephrine and epinephrine) to the slower rate with which the quinone derived from this compound undergoes cyclization compared to the rates of cyclization of the corresponding quinones derived from norepinephrine and epinephrine.[27] Finally, the isolation of a glutathione adduct of 6-OHDA from brain tissue treated in vivo with 6-OHDA[6] provides direct evidence for the in situ oxidation of 6-OHDA and the reactivity of the corresponding quinone with bionucleophiles.

Experimental Section

Melting points were determined on a Thomas Hoover melting point apparatus and are uncorrected. NMR spectra were recorded

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on a Varian FT-80 instrument. Chemical shifts are reported in part per million (ppm) relative to Me_4Si ($CDCl_3$ and Me_2SO-d_6) as internal standard. Infrared spectra were recorded on a Perkin-Elmer 337 grating IR spectrophotometer. Elemental analyses were performed by the Microanalytical Laboratory, University of California, Berkeley.

1-(2,5-Dimethoxyphenyl)-2-nitroethene (6b). A mixture of 2,5-dimethoxybenzaldehyde and nitromethane was condensed as described by Sugasawa et al.[28] to give **6b** as yellow crystalline needles: mp 118–119 °C (lit.[28] mp 119–120 °C); ^1H NMR ($CDCl_3$) δ 8.13 (d, J = 13.2 Hz, 1 H, vinylic proton), 7.76 (d, J = 13.2 Hz, 1 H vinylic proton), 6.95 (s, 2 H, Ar H) 6.90 (s, 1 H, Ar H), 3.85 (s, 3 H, OCH_3), 3.75 (s, 3 H, OCH_3), 3.75 (s, 3 H, OCH_3).

1-(2,5-Dimethoxyphenyl)-2-aminoethane (6c). Into a 1-L three-necked flask equipped with a magnetic stirrer and pressure-equalizing dropping funnel were added 400 mL of dry THF and $LiAlH_4$ (20 g, 0.53 mol). The system was purged with dry N_2 and cooled with an ice bath. After the mixture was stirred for 30 min, a solution of **6b** (25 g, 0.12 mol) in 150 mL of dry THF was added dropwise. This mixture was heated under reflux overnight. The excess hydride was destroyed by the successive addition of 20 mL of a THF/water mixture (1:1), 20 mL of 4 N NaOH, and 60 mL of water. The mixture was filtered, and the filter cake was digested with 50 mL of THF. The combined solvent was dried (K_2CO_3) and evaporated to an oil. Bulb to bulb distillation [oven temperature 110–120 °C (0.05 mmHg)] afforded 16.4 g (76%) of **6c** as a clear, colorless oil: ^1H NMR ($CDCl_3$) δ 6.66 (s, 3 H, Ar H), 3.70 (s, 6 H, OCH_3), 2.75 (m, 4 H, CH_2), 1.27 (br s, exchanges with D_2O, 2 H, NH_2). The HCl salt was prepared by dissolving 1 g of free base in 6 mL of 2-propanol and adding concentrated HCl until the pH was distinctly acidic. Upon addition of Et_2O, a white precipitate appeared. Recrystallization from EtOH provided a white crystalline product: mp 138–139 °C (lit.[29] mp 139 °C).

1-(2,5-Dihydroxyphenyl)-2-aminomethane Hydrochloride (6·HCl). To a solution of amine **6c** (17.44 g, 96 mmol) in 100 mL of CH_2Cl_2 cooled to −78 °C was added BBr_3 (10.88 mL, 220 mol) dropwise. The mixture was allowed to stir for 2 days at room temperature. After coooling, the reaction was quenched by the dropwise addition of 150 mL of MeOH. The brown, solid residue obtained after evaporation of the solvent was dissolved in a small volume of water and chromatographed on a cationic exchange column (300 g of Dowex AG 50W-X4, 50–100 mesh, washed with 300 mL of 2 N HCl and 300 mL of water). The column was washed with water until the eluent was free of halide (silver nitrate test). The product was eluted with 4 N HCl, and the eluent was lyophilyzed to give an off-white solid. Recrystallization from EtOH afforded 10 g (50%) of **6** as a white crystalline solid: mp 165–166 °C; ^1H NMR (D_2O) δ 6.85 (s, 3 H, Ar H), 3.13 (m, 4 H, CH_2). Anal. ($C_8H_{11}NO_2$·HCl) C, H, N, Cl.

1-(2,5-Dimethoxy-4-methylphenyl)-2-nitroethene (7b). A mixture of 2,5-dimethoxy-4-methylbenzaldehyde (**7a**; 14.5 g, 80 mmol),[30] NH_4OAc (2.58 g, 0.34 mol), and nitromethane (30 mL, 0.49 mol) was allowed to react in the same way as described for the synthesis of **6b**. Recrystallization from 95% EtOH yielded 13.2 g (73%) of **7b** as bright yellow needles: mp 118–119 °C (lit.[31] mp 118–119 °C); ^1H NMR ($CDCl_3$) δ 8.10 (d, J = 12.5 Hz, 1 H, vinylic proton), 7.75 (d, J = 12.5 Hz, 1 H, vinylic proton), 6.53 (s, 1 H, Ar H), 6.47 (s, 1 H, Ar H), 3.87 (s, 3 H, OCH_3), 3.80 (s, 3 H, OCH_3), 2.25 (s, 3 H, CH_3).

1-(2,5-Dihydroxy-4-methylphenyl)-2-aminoethane Hydrochloride (7·HCl). The synthesis of **7**·HCl was achieved by treatment of 1-(2,5-dimethoxy-4-methylphenyl)-2-aminoethane (**7c**;[28] 16.7 g, 86 mmol) with BBr_3 (25 mL, 0.28 mol) as described in the synthesis of **6**. Ion-exchange chromatography, followed by recrystallization from EtOH, provided 9.59 g (55%) of **7** as an off-white crystalline solid: mp 188–189 °C; ^1H NMR (D_2O) δ 6.82 (s, 1 H, Ar H), 6.79 (s, 1 H, Ar H), 3.35 (m, 2 H, CH_2), 2.75 (m, 2 H, CH_2), 2.22 (s, 3 H, CH_3). Anal. ($C_9H_{13}NO_2$·HCl) C, H, N.

1-(2,4,5-Trimethoxyphenyl)-2-nitroethene (8b). The reaction of 2,4,5-trimethoxybenzaldehyde (16.1 g, 82 mmol), NH_4OAc (3 g, 39 mmol), and nitromethane (32 mL, 0.52 mol) proceeded as described for the preparation of **6b**. Recrystallization from EtOH afforded 12.4 g (63%) of **8b** as orange-red needles: mp 130–131 °C (lit.[18] mp 130–132 °C); ^1H NMR ($CDCl_3$) δ 8.16 (d, J = 13.5 Hz, 1 H, vinylic proton), 7.75 (d, J = 13.5 Hz, 1 H, vinylic proton), 6.90 (s, 1 H, Ar H), 6.52 (s, 1 H, Ar H), 3.87 (s, 3 H, OCH_3), 3.85 (s, 3 H, OCH_3), 3.81 (s, 3 H, OCH_3).

1-(2,4,5-Trimethoxyphenyl)-2-aminoethane (8c). Nitroethene **8b** (20 g, 83.7 mmol) and $LiAlH_4$ (15 g, 0.20 mol) were allowed to react in a similar way as described for the synthesis of **6c**. Bulb to bulb distillation [oven temperature 140–150 °C (0.1 mmHg)] afforded 9.1 g (52%) of **8c** as a clear, colorless oil: ^1H NMR ($CDCl_3$) δ 6.65 (s, 1 H, Ar H), 6.45 (s, 1 H, Ar H), 3.87 (s, 3 H, OCH_3), 3.82 (s, 3 H, OCH_3), 3.78 (s, 3 H, OCH_3), 2.73 (m, 4 H, CH_2), 2.2 (br s, exchanges with D_2O, NH_2). The HCl salt was prepared as described for **6c**, mp 187–188 °C (lit.[18] mp 187–188 °C).

1-(2,4,5-Trimethoxyphenyl)-2-(*tert*-butoxyamino)ethane (8d). To a solution of amine **8c** (8.5 g, 40 mmol) in 100 mL of THF was added triethylamine (9.5 mL) and 25 mL of water, followed by 2-[[(*tert*-butoxycarbonyl)oxy]imino]-2-phenylacetonitrile (12.3 g, 50 mmol). The mixture was stirred for 3 h at room temperature and warmed gently on a steam bath for 15 min. The mixture was then acidified with aqueous citric acid (5%, 50 mL), and the aqueous layer was extracted with ether (2 × 100 mL). The combined organic layers were dried (K_2CO_3) and concentrated to an oily residue, which solidified upon cooling. Recrystallization from EtOH yielded 11 g (88%) of **8d** as a white solid: mp 78–79 °C; ^1H NMR ($CDCl_3$) δ 6.68 (s, 1 H, Ar H), 6.51 (s, 1 H, Ar H), 3.87 (s, 3 H, OCH_3), 3.82 (s, 3 H, OCH_3), 3.30 (t, J = 6.6 Hz, 2 H, CH_2), 2.73 (t, J = 6.6 Hz, 2 H, CH_2), 1.42 (s, 9 H, CH_3). Anal. ($C_{16}H_{25}NO_5$) C, H, N.

2-Methoxy-5-[2-[(*tert*-butoxycarbonyl)amino]ethyl]-*p*-quinone (8e). Ceric ammonium nitrate (48 g, 87 mmol) in 75 mL of water was added dropwise to a stirred solution of **8d** (11 g, 35 mmol) in 250 mL of acetonitrile. After the addition was complete, the mixture was stirred for 1 h at room temperature and then extracted with CH_2Cl_2 (2 × 150 mL). The extract was dried (K_2CO_3) and evaporated to an oil. Treatment with EtOH (100%) resulted in crystallization of a yellow product. Recrystallization of the solid gave 3.65 g (37%) of **8e**: mp 126–127 °C; ^1H NMR ($CDCl_3$) δ 6.55 (s, 1 H, allylic proton), 5.95 (s, 1 H, allylic proton), 3.83 (s, 3 H, OCH_3), 3.33 (t, J = 6.5 Hz, 2 H, CH_2), 2.69 (t, J = 6.5 Hz, 2 H, CH_2), 1.45 (s, 9 H, CH_3). Anal. ($C_{14}H_{19}NO_5$) C, H, N.

1-(2,5-Dihydroxy-4-methoxphenyl)-2-[(*tert*-butoxycarbonyl)amino]ethane (8f). A solution of sodium dithionate (2.5 g, 12 mmol) in 30 mL of water was added dropwise to a stirred solution of **8e** (2.8 g, 10 mmol) in 50 mL of CH_2Cl_2. This mixture was stirred for 1 h at room temperature, extracted with CH_2Cl_2 (2 × 100 mL), dried ($MgSO_4$), and evaporated to a chalky solid residue. The solid was recrystallized from EtOH to yield 1.6 g (57%) of **8f** as a white solid: mp 157–158 °C; ^1H NMR ($CDCl_3$) δ 6.60 (s, 1 H, Ar H), 6.47 (s, 1 H, Ar H), 5.10 (br s, exchanges with D_2O, 1 H, NH), 3.82 (s, 3 H, OCH_3), 3.17 (m, 2 H, CH_2), 2.71 (m, 2 H, CH_2), 1.45 (s, 9 H, CH_3). Anal. ($C_{14}H_{21}NO_5$) C, H, N.

1-(2,5-Dihydroxy-4-methoxyphenyl)-2-aminoethane Hydrochloride (8·HCl). A mixture of freshly distilled trifluoroacetic acid (2 mL) and **8f** (1.6 g, 5.6 mmol) was stirred for 1 h at room temperature. Evaporation yielded a purplish solid residue, which was taken up in a small volume of water and chromatographed on a cation-exchange column (10 g of dry resin, Dowex AG 50W-X4, 50–100 mesh, washed with 75 mL of 2 N aqueous HCl and 100 mL of water). This column was eluted with water until the eluent was free of halide (silver nitrate test). The product was eluted with 4 N HCl. The combined eluents were lyophilyzed to give an off-white solid, which was recrystallized from EtOH to afford 1 g (80%) of **8**·HCl: mp 188–189 °C (lit.[32] mp 176–179

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°C); ^{1}H NMR (D_2O) δ 6.82 (s, 1 H, Ar H), 6.69 (s, 1 H, Ar H), 3.90 (s, 3 H, OCH_3), 3.15 (m, 4 H, CH_2). Anal. ($C_9H_{13}NO_3$·HCl) C, H, N.

1-(2,5-Dimethoxy-4-nitrophenyl)-2-(acetylamino)ethane (9d). To an ice-cold solution of amine **6c** (14.1 g, 67 mmol) in a mixture of acetic acid (22 mL, 0.37 mol) and acetic anhydride (36 mL, 0.35 mol) was added nitric acid (18 mL, 0.29 mol) dropwise. The mixture was stirred at 0 °C for 1 h. The yellow solid that separated was suction filtered and recrystallized from EtOH to yield 18.45 g (80%) of **9d** as yellow crystals: mp 142.5–143 °C; ^{1}H NMR ($CDCl_3$) δ 7.41 (s, 1 H, Ar H), 6.92 (s, 1 H, Ar H), 3.92 (s, 3 H, OCH_3), 3.83 (s, 3 H, OCH_3), 2.90 (m, 4 H, CH_2), 1.38 (s, 3 H, $COCH_3$). Anal. ($C_{12}H_{16}N_2O_5$) C, H, N.

1-(2,5-Dimethoxy-4-nitrophenyl)-2-aminoethane Hydrochloride (9·HCl). Aqueous HBr (3.5 mL, 48%, distilled over $SnCl_2$) was added to **9d** (4.0 g, 13.5 mmol). The mixture was heated under reflux for 6 h and then evaporated to dryness to give a reddish-brown solid residue. The solid was taken up in 5 mL of water and chromatographed on a cationic exchange resin (25 g of Dowex 50W-X4, 50–100 mesh); the product eluted with 4 N HCl. The eluent was lyophilyzed to yield a red solid. Recrystallization from EtOH yielded 2.0 g (57%) of **9**·HCl: mp 200 °C with decomposition; ^{1}H NMR (Me_2SO-d_6) δ 7.80 (br s, exchanges with D_2O, 3 H, NH_3), 2.83 (m, 4 H, CH_2). Anal. ($C_8H_{10}N_2O_4$·HCl) C, H, N, Cl.

1-(2,5-Dimethoxy-4-aminophenyl)-2-(acetylamino)ethane (10d). To a solution of **9d** (15 g, 56 mmol) in 23 mL of concentrated hydrochloric acid was added Sn (16.6 g, 140 mmol) portionwise. The orange–red solution was heated for 1 h on a steam bath and filtered hot through glass–wool to remove unreacted tin. Evaporation of the solvent gave a greyish solid residue. Recrystallization from EtOH yielded 6.37 g (48%) of **10d**: mp 139–140 °C; ^{1}H NMR ($CDCl_3$) δ 6.57 (s, 1 H, Ar H), 6.32 (s, 1 H, Ar H), 5.67 (br s, exchanges with D_2O, NH), 3.79 (s, 3 H, OCH_3), 3.75 (s, 3 H, OCH_3), 3.34 (q, 2 H, CH_2), 2.71 (t, 2 H, CH_2), 1.90 (s, 3 H, $COCH_3$). Anal. ($C_{12}H_{18}N_2O_3$) C, H, N.

1-(2,5-Dihydroxy-4-aminophenyl)-2-aminoethane Hydrochloride (10·HCl). Compound **10d** (4.84 g, 21.4 mmol) and 48% HBr (5 mL, distilled over $SnCl_2$) were allowed to react in a similar fashion as described for the synthesis of **9**. Ion-exchange chromatography, followed by recrystallization from EtOH, yielded 3 g (68%) of **10**·HCl: mp 230 °C with decomposition; ^{1}H NMR (D_2O) δ 7.39 (s, 1 H, Ar H), 6.95 (s, 1 H, Ar H), 3.07 (m, 4 H, CH_2). Anal. ($C_8H_{12}N_2O_2$·HCl) C, H, N, Cl.

1-(2,5-Dihydroxy-4-bromophenyl)-2-aminoethane Hydrochloride (11·HCl). Amine **11c**[33] (11.1 g, 43 mmol) and BBr_3 (13 mL, 137 mmol) were allowed to react in a similar way as described for the synthesis of **6**. Ion-exchange chromatography, followed by recrystallization from EtOH, yielded 6.3 g (53%) of **11**·HCl as an off-white solid: mp 200 °C with decomposition; ^{1}H NMR (D_2O) δ 7.17 (s, 1 H, Ar H), 6.91 (s, 1 H, Ar H), 3.05 (m, 4 H, CH_2). Anal. ($C_8H_{10}BrNO_2$·HCl) C, H, N, Cl.

1-(2,5-Dimethoxy-4-bromophenyl)-2-(phthalimidoamino)ethane (11d). To a solution of **11c** (7.24 g, 27.8 mmol) and phthalic anhydride (4.5 g, 30 mmol) in 100 mL of DMF (distilled) was added molecular sieves. The reaction mixture was heated under reflux overnight and, after cooling, was suction filtered to remove the molecular sieves. Treatment with CH_2Cl_2 resulted in the crystallization of yellow needles. Recrystallization from EtOH provided 7.57 g (69%) of **11d**: mp 141.5–142 °C; ^{1}H NMR ($CDCl_3$) δ 7.65 (m, 4 H, Ar H), 6.97 (s, 1 H, Ar H), 6.68 (s, 1 H, Ar H), 3.94 (t, J = 7.3 Hz, 2 H, CH_2) 3.72 (s, 3 H, OCH_3), 3.71 (s, 3 H, OCH_3), 2.96 (t, J = 7.3 Hz, 2 H, CH_2). Anal. ($C_{18}H_{16}BrNO_4$) C, H, N, Br.

1-(2,5-Dimethoxy-4-cyanophenyl)-2-(phthalimidoamino)-ethane (12d). Compound **11d** (7.57 g, 19.4 mmol) and cuprous cyanide (2.0 g, 22.3 mmol) in 150 mL of DMF were heated under reflux for 5 h. The mixture was poured into a solution containing hydrated ferric chloride (6 g), 1.48 mL of concentrated HCl, and 9 mL of water. The solution was maintained at a temperature of 60–70 °C for 20 min to decompose the complex and then extracted with CH_2Cl_2. The organic phase was washed with dilute aqueous HCl (100 mL), dried ($MgSO_4$), and evaporated to give a white solid. Recrystallization from EtOH provided 5.98 g (91%) of **12d** as white needles: mp 194–195 °C; IR (KBr) 2200 (C≡N) cm^{-1}; ^{1}H NMR ($CDCl_3$) δ 7.70 (m, 4 H, Ar H), 6.91 (s, 1 H, Ar H), 6.73 (s, 1 H, Ar H), 3.97 (t, J = 6.8 Hz, 2 H, CH_2), 3.88 (s, 3 H, OCH_3), 3.72 (s, 3 H, OCH_3), 3.4 (t, J = 6.8 Hz, 2 H, CH_2). Anal. ($C_{19}H_{16}N_2O_4$) C, H, N.

1-(2,5-Dimethoxy-4-cyanophenyl)-2-aminoethane (12c). Phthalimide **12d** (9.2 g, 27.38 mmol) and hydrazine (2.2 mL, 68.55 mmol, 98%) in 50 mL of anhydrous EtOH were heated under reflux for 15 min. After the mixture was cooled, the phthalazinedione was filtered off, and the filtrate was evaporated to give a solid residue. The solid was dissolved in H_2O and extracted with $CHCl_3$. The organic extract was washed with aqueous Na_2CO_3 (10%, 3 × 100 mL), dried (Na_2SO_4), and evaporated to give 3.6 g (64%) of **12c**: ^{1}H NMR ($CDCl_3$), 6.96 (s, 1 H, Ar H), 6.30 (s, 1 H, Ar H), 3.88 (s, 3 H, OCH_3), 2.80 (m, 4 H, CH_2), 1.20 (br s, exchanges with D_2O, 2 H, NH_2). The HCl salt (precipitated from ether) was recrystallized from EtOH, mp 220–222 °C. Anal. ($C_{11}H_{14}N_2O_2$·HCl) C, H, N.

1-(2,5-Dihydroxy-4-cyanophenyl)-2-aminoethane Hydrochloride (12·HCl). Amine **12c** (3.6 g, 17.5 mmol) and BBr_3 (1.7 ml, 17.9 mmol) were allowed to react in the same way as described for the preparation of **6**. Ion-exchange chromatography, followed by recrystallization from EtOH, yielded 1.5 g (40%) of **12**·HCl: mp 230–231 °C with decomposition; ^{1}H NMR (Me_2SO-d_6) δ 7.83 (br s, exchanges with D_2O, 2 H, OH), 6.94 (s, 1 H, Ar H), 6.81 (s, 1 H, Ar H), 2.92 (m, 4 H, CH_2). Anal. ($C_9H_{10}N_2O_2$·HCl) C, H, N, Cl.

1-(2,5-Dihydroxy-4-carboxyphenyl)-2-aminoethane Hydrochloride (13·HCl). To a solution of amine **12c** (12 g, 58 mmol) in 50 mL of EtOH was added 100 mL of 25% aqueous NaOH. The resulting solution was held under reflux for 5 h. After cooling, the mixture was treated with Dowex 50 × 8 cationic resin until the pH reached 1. The resin was collected and washed with water, and then the product was eluted with concentrated NH_4OH. The filtrate was concentrated in vacuo to a solid residue, which was triturated with acetone and crystallized from EtOH to yield 2.3 g (17%) of **13c**: mp 135–137 °C; ^{1}H NMR (D_2O) δ 7.11 (s, 1 H, Ar H), 6.94 (s, 1 H, Ar H), 3.83 (s, 3 H, OCH_3), 3.81 (s, 3 H, OCH_3), 3.26 (m, 4 H, CH_2). Without further purification, this amine (4.6 g, 22 mmol) and BBr_3 (1.7 mL, 17.9 mmol) were allowed to react in a similar way as described for the synthesis of **6a**. Ion-exchange chromatography, followed by crystallization from EtOH, yielded 290 mg (60%) of **13**·HCl as a white crystalline solid: mp 260–261 °C; ^{1}H NMR (Me_2SO-d_6) δ 8.00 (br s, exchanges with D_2O, 3 H, OH), 7.23 (s, 1 H, Ar H), 6.70 (s, 1 H, Ar H), 2.89 (br s, 4 H, CH_2). Anal. ($C_9H_{11}NO_4$·HCl) C, H, N, Cl.

1-(2,5-Dimethoxy-4-chlorophenyl)-2-(phthalimidoamino)ethane (14d). Compound **11d** (14.94 g, 38.3 mmol) and cuprous chloride (4.5 g, 45 mmol) were allowed to react in a way similar to that described for the synthesis of **12d**. Recrystallization from hot EtOH provided 12.18 g (92%) of **14d** as yellow needles: mp 138–140 °C; ^{1}H NMR ($CDCl_3$) δ 7.70 (m, 4 H, Ar H), 6.82 (s, 1 H, Ar H), 6.70 (s, 1 H, Ar H), 3.94 (t, 2 H, CH_2), 3.71 (s, 6 H, OCH_3), 2.95 (t, 2 H, CH_2). Anal. ($C_{18}H_{16}NO_4Cl$) C, H, N, Cl.

1-(2,5-Dimethoxy-4-chlorophenyl)-2-aminoethane (14c). Phthalimide **14d** (12.18 g, 35 mmol) and hydrazine (2.86 mL, 87.5 mmol, 98%) in 60 mL of anhydrous EtOH were allowed to react as described for the synthesis of **12c**. Bulb to bulb distillation [oven temperature 145–155 °C (0.05 mmHg)] afforded 5.16 g (67.8%) of **14c** as a clear, colorless oil: ^{1}H NMR ($CDCl_3$) δ 6.87 (s, 1 H, Ar H), 6.76 (s, 1 H, Ar H), 3.84 (s, 3 H, OCH_3), 3.76 (s, 3 H, OCH_3), 2.77 (m, 4 H, CH_2), 1.37 (br s exchanges with D_2O, 2 H, NH_2). The HCl salt (precipitated from ether) was crystallized from EtOH, mp 220–221 °C. Anal. ($C_{10}H_{14}NO_2$·HCl) C, H, N.

1-(2,5-Dihydroxy-4-chlorophenyl)-2-aminoethane Hydrochloride (14·HCl). Amine **14c** (5.16 g, 24 mmol) and BBr_3 (6.82 mL, 72 mmol) were allowed to react as described for the synthesis of **6**. Ion-exchange chromatography, followed by recrystallization from EtOH, yielded 3.2 g (60%) of **14**·HCl as a white crystalline solid: mp 210–211 °C; ^{1}H NMR (D_2O) δ 6.94 (s, 1 H, Ar H), 6.85 (s, 1 H, Ar H), 3.27 (m, 2 H, CH_2), 2.91 (m, 2 H, CH_2). Anal. ($C_8H_{10}NO_2Cl$·HCl) C, H, N, Cl.

Electrochemistry. Cyclic voltammetry was utilized for these studies. This is a rapid voltage-scanning technique that employs

(33) Shulgin, A. T.; Carter, M. F. *Physchopharmacol. Commun.* **1975** *1*, 93.

a stationary working electrode in quite solution and a repetitive, triangular (isosceles) wave potential sweep between it and a reference electrode. The anodic and cathodic currents are recorded as a function of the applied triangular potential sweep on an X–Y recorder.

The working electrode used was a thin-layer electrode that consisted of a section of precision-bore, heavy-wall Pyrex tubing cut to the appropriate size (0.1235 ± 0.0002 in. i.d. × 9 mm o.d. × 11 mm length) by means of a wet-wheel saw, with care being taken to produce true, carefully finished ends. The thin-layer cavity consisted of a close-fitting platinum rod (0.6-in. length × 0.1215-in. diameter). This platinum electrode could be removed from the glass container for cleaning and reinserted without undue contamination.

Reproducible electrode surface pretreatment was accomplished as follows. The platinum electrode was removed from the glassware and heated to redness for 10 min in an oxidizing methane–oxygen flame, allowed to cool for a few seconds in air, and then inserted into the capillary. A cyclic current-potential curve was recorded in 1 M $HClO_4$ from +1.30 to −0.105 V vs. AgCl, which served the double purpose of removing the last traces of absorbed organic materials and confirming the cleanliness of the surface.

All solutions were prepared from reagent grade chemicals. Triply distilled water was employed, the second distillation being from a solution of 10^{-3} M $KMnO_4$ in 10^{-3} M NaOH. The test compounds were dissolved in Britton–Robinson buffers at a concentration of 0.25 mM. All solutions were deoxygenated with a vigorous stream of nitrogen, and all studies were performed under an atmosphere of nitrogen.

Formal oxidation potentials were obtained at scan rates of either 2 or 10 mV/s. The reported potentials were measured relative to a Ag/AgCl electrode in 1 M NaCl solution.

To measure the cyclization rates, we oxidized the *p*-hydroquinones at a constant potential (E_{ox}) and observed the current-time decays following different time delays to allow for cyclization to occur. At a later time, the electrode was switched to a potential (E_{rest}) sufficient to reduce the *p*-quinone previously electrogenerated (see Table I). The measured current at this potential corresponds to the amount of *p*-quinone that has not been consumed by cyclization. From a knowledge of the delay time (which can vary from 1.5 to 40 s) and the amount of *p*-quinone consumed, one can calculate the cyclization rate of the *p*-quinone.

Animal Studies. Male Sprague–Dawley rats purchased from Simonson Co. and weighing 120 ± 10 g were utilized. The rats were allowed food and water ad libidium. The appropriate dose of each compound was dissolved in 0.9% saline containing 0.1% ascrobic acid to prevent autoxidation prior to delivery and was administered intravenously in a volume of 0.2 mL. Control animals were administered 0.2 mL of vehicle. The data reported in Table II are the mean plus or minus SD values obtained from 12 determinations with three drug-treated animals and 12 determinations with three control animals for each dose studied. The percent uptake inhibiton (facilitation) was calculated as the difference between the values obtained for the control vs. drug treated animals.

Injections were made with a 1-cm^3 tuberculin syringe (27-gauge needle) in the left femoral vein. The rats were put under light anesthesia with halothane, and a small incision (1 in.) was made in the left leg to expose the vein. After injection, the incision was stitched up with autoclips. The rats were sacrificed 24 h after injection by decapitation. The atria were removed and sectioned in the cold into four 10–15-mg pieces.

The incubation medium used was a Kreb–Ringer bicarbonate buffer, which consisted of NaCl (6.92 g/L), KCl (0.35 g/L), $CaCl_2 \cdot 2H_2O$ (0.19 g/L), KH_2PO_4 (0.16 g/L), $MgSO_4$ (0.14 g/L), $NaHCO_3$ (2.10 g/L), dextrose (1.8 g/L), and ascorbic acid (0.2 g/L). This buffer has a pH of 7.35. The requisite amount of [7-^{3}H]norepinephrine (New England Nuclear, 2.7 Ci/mmol) was added to the medium to provide a 10^{-7} M solution (0.27 μCi/mL) of the catecholamine. The sections were placed in individual vials, 1-mL aliquots of [^{3}H]norepinephrine-containing medium was added, and the samples were incubated under an atmosphere of 95% O_2–5% CO_2 at 37 °C for 30 min in a metabolic shaker. The tissue sections were removed and placed in fresh vials, and a 1-mL aliquots of [^{3}H]norepinephrine-free medium was added. The incubation was resumed as before for another 10 min. Each section was then sonicated with 0.5 mL of 0.4 N aqueous $HClO_4$. The resulting homogenates were centrifuged to provide pellets, and 0.2-mL aliquots of the supernatants were removed and placed in individual scintillation vials containing 15-mL portions of Aquasol (New England Nuclear). The solutions were then counted for released radioactivity.

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Registry No. 1, 1199-18-4; **6**, 21581-41-9; **6**·HCl, 88440-94-2; **6b**, 40276-11-7; **6c**, 3600-86-0; **7**, 81255-52-9; **7**·HCl, 88440-95-3; **7a**, 4925-88-6; **7b**, 25505-64-0; **7c**, 24333-19-5; 8, 38411-82-4; **8**·HCl, 13062-74-3; **8b**, 24160-51-8; **8c**, 15394-83-9; **8d**, 88440-96-4; **8e**, 88440-97-5; **8f**, 88440-98-6; **9**, 41241-39-8; **9**·HCl, 88440-99-7; **9d**, 88441-00-3; **10**, 41241-40-1; **10**·HCl, 88441-01-4; **10d**, 88441-02-5; 11, 81255-55-2; 11·HCl, 88441-03-6; **11c**, 66142-81-2; **11d**, 88441-04-7; **12**, 88441-05-8; **12**·HCl, 88441-06-9; **12c**, 88441-07-0; **12c**-HCl, 88441-08-1; **12d**, 88453-16-1; **13**, 88441-09-2; **13**·HCl, 88441-10-5; **13c**, 88441-11-6; **14**, 88441-12-7; **14**·HCl, 88441-13-8; **14c**, 88441-14-9; **14c**·HCl, 88441-15-0; **14d**, 88441-16-1; 2,4-dimethoxybenzaldehyde, 93-02-7; nitromethane, 75-52-5; 2,4,5-trimethoxybenzaldehyde, 4460-86-0.