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ELECTROCHEMICAL OXIDATION OF METHYLENEDIOXYAMPHETAMINES

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Summary—Four amphetamine derivatives bearing a methylenedioxy group at positions 3 and 4 of the benzene ring and differing in their substitution at C(6) were studied by differential pulse voltammetry in aqueous media. These experiments showed a single oxidation peak for the C(6)-H, -Br and -Cl compounds, while the C(6)-NO_2 analogue was not oxidized. The oxidation peak is interpreted as due to the removal of one electron from the aromatic electrophore with formation of a radical cation stabilized by the dioxole ring. The linear relationship between the peak current and the concentration of the derivatives is appropriate for development of a quantitative method for their determination. pK′ values were determined using both electrochemical and spectrophotometric methods.

The last few years have seen a renewal of interest in the psychotropic drugs 1-(3,4-methylenedioxyphenyl)-2-aminopropane or 3,4-methylenedioxyamphetamine (MDA) and its *N*-methyl and *N*-ethyl analogues (MDMA, "XTC", or "Adam", and MDEA or "Eve", respectively), owing to their disputed use in psychotherapy and to their neurotoxicity.[1,2] MDMA may be regarded as the prototype of a new class of drugs called "entactogens"[3,4] whose mechanism of action and structural requirements are almost totally unknown. It is only clear at this time that their subjective effects differ from those of the structurally similar phenylalkylamine hallucinogens, and that these effects are mainly due to their (S) isomers while the more potent hallucinogens possess the (R) configuration. Thus far, the exploration of this group of compounds has been limited to variations of the amine chain keeping the 1-(3,4-methylenedioxyphenyl) moiety intact and devoid of additional substitution, and has only very recently been extended to include the aminotetralin and aminoindan analogues.[3–5] Nevertheless, it is known that the bromination of MDA to afford 1-(6-bromo-3,4-methylenedioxyphenyl)-2-aminopropane leads to a non-hallucinogenic compound whose subjective effect was interpreted almost twenty years ago as "amphetamine-like" although this was only observed in the rather high dose range which is also required for MDA, MDMA and MDEA.[6,7]

As part of a synthetic program related to the known "entactogens", MDA and the above mentioned bromo derivative were prepared once more, as well as another two compounds substituted at C(6) of the benzene ring with a chlorine atom or a nitro group, respectively. In order to round out our vision of substituent effects on the electrochemistry of amphetamine analogues, obtained with a series of 1-(2,5-dimethoxyphenyl)-2-aminopropane derivatives substituted at C(4),[8] we have now studied the voltammetric behavior of MDA and its congeners.

The electrochemistry of amphetamine derivatives is an unexplored field of research. There are only three published papers related to: the nitro-reduction of 2,5-dimethoxy-4-nitroamphetamine (DON)[9] and 4,5,-dimethoxy-2-nitroamphetamine,[10] and the electrochemical behavior of several 4-substituted 2,5-dimethoxy-amphetamine derivatives.[8] From this latter work it is possible to conclude the existence of a correlation between the ring substitution and the oxidation potential. As the ring substitution in the amphetamine derivatives play an important role in their pharmacological activity, it may be possible to find empirical relationships between oxidation potentials and pharmacological activities.

EXPERIMENTAL

Reagents

MDA and its C(6)-bromo analogue were synthesized following published procedures.[6,7] Melting points are uncorrected. ^{1}H NMR spectra were recorded at 60 MHz in D_2O unless stated otherwise (chemical shifts in ppm from TMSPA-d_4).

1-(3,4-methylenedioxyphenyl)-2-aminopropane. (3,4-MDA)hydrochloride. M.p. 193.5–194.5° (i-PrOH-EtO); ^{1}H NMR (TFA) δ (from TMS) 1.53 (3H, d J = 6.4 Hz, C-CH3), 3.0 (2H, m, CH2), 3.8 (1H, m, CH), 6.02 (2H, s, OCH2O), 6.85 (3H, br s, ArH).

1-(3,4-methylenedioxyphenyl-6-nitrophenyl)-2-aminopropane. (3,4-MD-6-NA) nitrate. 1-(3,4-methylenedioxyphenyl)-2-aminopropane (2.00 g) was dissolved in 2*N* HNO_3 (5.5 ml) and treated dropwise, with efficient stirring and cooling, with 65% HNO_3 (6 ml). After several minutes the product separated out as a thick creamy precipitate which was diluted with water (18 ml), collected by filtration, resuspended in water, filtered again and dried. The yield was practically quantitative, m.p. 171° (decomp.) (EtOH); ^{1}H NMR δ (DMSO-d6) 1.19 (3H, d J = 6 Hz, C-CH3), 3.10 (2H, d J = 6 Hz, CH2), 3.5 (1H, m, CH), 6.27 (2H, s, OCH2O), 7.10 (1H, s, ArH), 7.67 (1H, s, ArH). Anal. C, 41.94; H, 4.59; N, 14.49%; calc. C10H13N207; C, 41.82; H, 4.56; N, 14.63%.

1-(6-bromo-3,4-methylenedioxyphenyl)-2-aminopropane(6-Br-3,4-MDA) hydrochloride. M.p. 221–222° (i-PrOH-acetone); ^{1}H NMR δ 1.32 (3H, d J = 6.5 Hz, C-CH3), 3.00 (2H, app. d Japp = 7 Hz, CH2), 3.7 (1H, m, CH), 6.00 (2H, s, OCH2O), 6.85 (1H, s, ArH), 7.12 (1H, s, ArH). Anal. C, 40.73; H, 4.49; N, 4.40%; calc. C10H13BrClNO2; C, 40, 76; H, 4.45; N, 4.75%.

1-(6-chloro-3,4-methylenedioxyphenyl)-2-aminopropane (6-Cl-3,4-MDA) hydrochloride. Prepared by LiAlH4 reduction of 1-(6-chloro-3,4-methylenedioxyphenyl)-2-nitropropene in Et_2O and precipitation of the salt; m.p. 222.5–223.5° (i-PrOH); ^{1}H NMR δ 1.33 (3H, d J = 6.4 Hz, C-CH3), 2.97 (2H, app. d Japp = 7 Hz, CH2), 3.7 (1H, m, CH), 6.01 (2H, s. OCH2O), 6.87 (1H, s, ArH), 7.00 (1H, s, ArH). Anal. C, 47.85; H, 5.28; N, 5.28%; calc. C10H13Cl2NO2: C, 48.02; H, 5.24; N, 5.60%.

Voltammetric experiments were carried out in buffered aqueous solutions containing 0.02*M* phosphoric acid with 0.02*M* acetic acid for pH 1–8.5 or 0.02*M* Na_2CO_3 for pH 8.5–12. The ionic strength was raised to 0.3*M* with $NaNO_3$.

For spectrophotometric experiments the Universal UV Spectroscopy buffer containing 0.1*M* in citric acid, potassium monophosphate, sodium tetraborate, TRIS and potassium chloride was used.

For both buffers, the pH was adjusted using HCl or NaOH. All reagents were p.a. grade.

The solid electrodes were routinely cleaned with chromic acid solution for 10 sec. This procedure permit to increase the reproducibility considerably.

Apparatus

A Tacussel CPRA thermostatic cell with three different working electrodes (platinum, glassy carbon and carbon paste) was employed. A platinum wire and a saturated calomel electrode were used as auxiliary and reference electrodes. A TACUSSEL model EDI rotating electrode assembly was used with platinum and glassy carbon electrodes. A METROHM carbon paste electrode with a geometric area of 38.5 mm^2 was also used.

Electrochemical data were obtained from an Inelecsa assembly equipped with the following elements:

(a) a generator–potentiostat type PDC-210.

(b) an interface containing 12-bit A/D and D/A converters, connected to a microprocessor with suitable software for fully automated control of the experiments and data acquisition. A Multitech, Apple II Plus-compatible microcomputer was used for data control, acquisition and treatment.

UV-Vis spectra were recorded using a SHIMADZU UV-160A spectrophotometer with 1 cm quartz cells.

A VARIAN Anaspect EM-360 (60 mHz) NMR spectrometer for NMR measurements was used.

RESULTS AND DISCUSSION

The present paper deals with the study of the voltammetric behavior of 3,4-methylenedioxyamphetamine (MDA) and its 6-chloro, 6-nitro and 6-bromo derivatives (Fig. 1).

In aqueous solution, using platinum, glassy carbon and carbon paste electrodes as working electrodes, MDA and the 6-chloro and 6-bromo derivatives produce an anodic peak which is

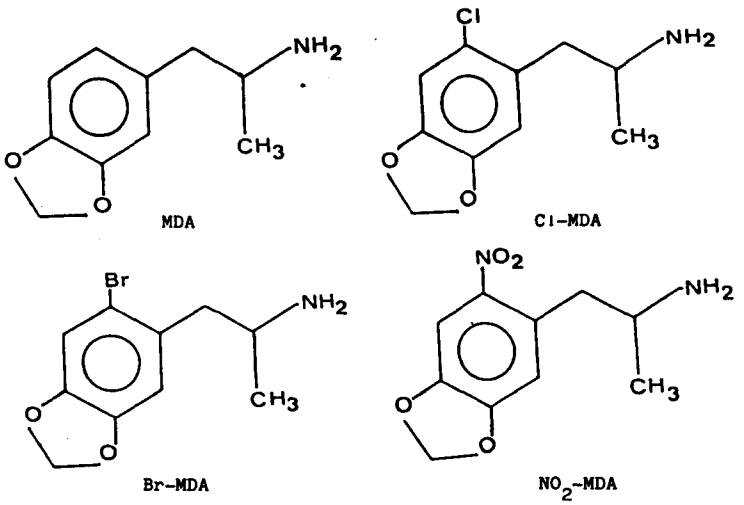

Fig. 1. Molecular structures of 3,4-methylenedioxyamphetamine (MDA) and its 6-chloro (Cl–MDA), 6-nitro (NO_2–MDA) and 6-bromo (Br–MDA) derivatives.

best resolved using differential pulse voltammetry. Unlike these compounds, the 6-nitro derivative of MDA did not reveal any anodic peak under these conditions. For comparative purposes, we also examined the electrochemical behavior of unsubstituted amphetamine and 2-methoxy amphetamine. No oxidation peaks were obtained for these compounds, suggesting that the anodic process observed with the other analogues does not involve the amine-substituted side chain but rather the 3,4-methylenedioxy-substituted benzene ring. In earlier work,[8] similar behaviour was observed for the 2,5-dimethoxyamphetamine derivatives.

The MDA and its chloro and bromo derivatives exhibited a single voltametric peak over the entire pH range studied, extending from pH 1 to 12. The peak potential (E_p)–pH plots (Fig. 2) show two linear segments for each compound, indicating that the electrode process is pH-dependent over the whole range. E_p decreases linearly with increasing pH; therefore, these compounds are oxidized more easily in more alkaline solutions as expected for common oxidative behavior. Moreover, the breaks in the E_p–pH plots can be ascribed to the voltammetric pK' values, showing that, when pH < pK' the dominant chemical species is the protonated amine and when pH > pK' the free base is more abundant. In order to confirm the above assumption we also studied the pH influence on the UV absorption spectra of the derivatives. In Fig. 3 we can observe the UV spectra of the amphetamine derivatives at two different pHs, displaying the strong pH-dependence of their UV band at approximately 210 nm. In the Fig. 4 plots of the absorptivity as a function of pH for the band at 210 nm are shown. From these curves we estimate the UV spectrophotometric pK_a values, which can be found in Table 1. The

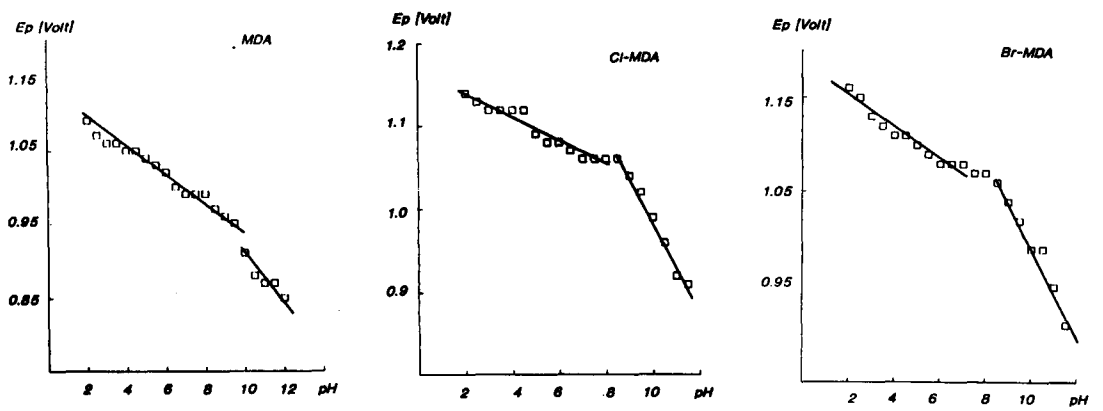

Fig. 2. Variation of the peak potential (E_p) with pH for the differential pulse voltammetric oxidation of the 3,4-methylenedioxyamphetamine derivatives. Voltammograms obtained on glassy carbon electrode.

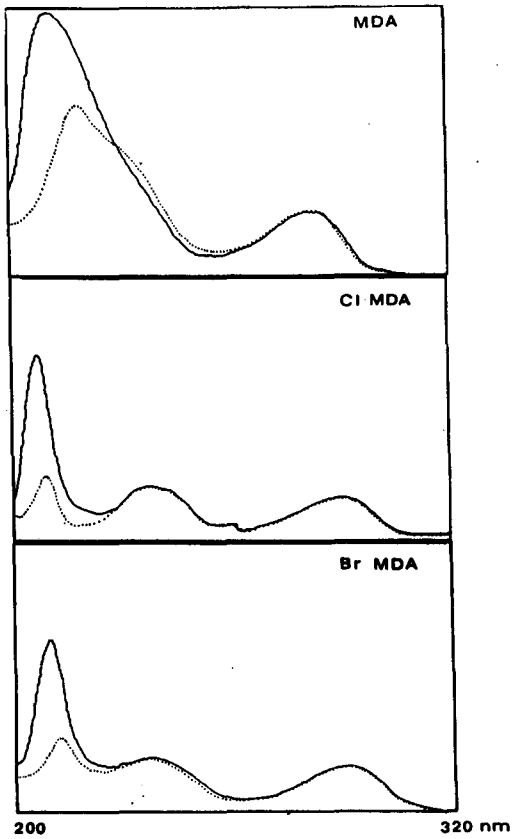

Fig. 3. UV spectra for the 3,4-methylenedioxyamphetamine derivatives. Solid line, pH = 2. Dashed line, pH = 10.

agreement between the voltammetric and the UV values supports the validity of the above assumption.

Examination of the peak current (i_p) values indicates that these fall (Fig. 5), and the peak broadens, as the pH rises. This may be due to a decrease in the heterogeneous rate constant affected by proton transfer.[11]

We also studied the electrochemical behavior of all derivatives under two different experimental conditions: a) variation of the temperature using a glassy carbon electrode as a working electrode, b) the effect of a platinum rotating disk electrode as a working electrode, maintaining a constant temperature (25°). From these experiments it was concluded that I_p does not exhibit variation with temperature or the rate of rotation, indicating that the oxidation rate is controlled by charge transfer without participation of the diffusion of the electroactive species to the electrode surface.

Table 1. pK' values obtained by voltammetric (glassy carbon and carbon paste electrodes) and spectrophotometric methods

Drug	Voltammetric C.P.E.	Voltammetric G.C.E.	Spectrophotometric
MDA	9.15	9.20	9.21
Cl–MDA	9.10	9.30	9.02
Br–MDA	8.75	8.80	9.12

One of the goals of this work was to evaluate the incidence of the C-6 substituent on the electrochemistry of these drugs. The experimental evidence here shown indicates that the electron acceptor or donor character of this substituent is directly related to the greater or lesser ability of oxidation. A simple explanation is that the electron-donating substituent increases the electron density of the aromatic ring π system, making it easier to remove an electron, thus producing a cation radical which is stabilized by the methylenedioxy group at C-3 and C-4. Consistent with this latter interpretation, the lowest E_p value was found for MDA itself, which lacks any substituent at C-6. Conversely, the chloro- and bromo-derivatives present higher E_p values. In these cases, the oxidation of the ring system is presumably more difficult due to the decreased stability of the resulting cation radical. In Fig. 2 we can see that

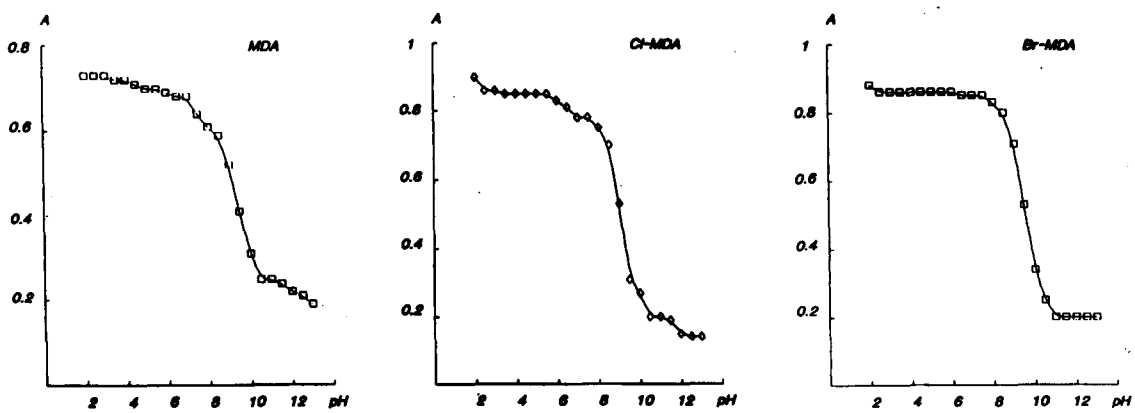

Fig. 4. Absorbance–pH plots for the band at 210 nm for the 3,4-methylenedioxyamphetamine derivatives.

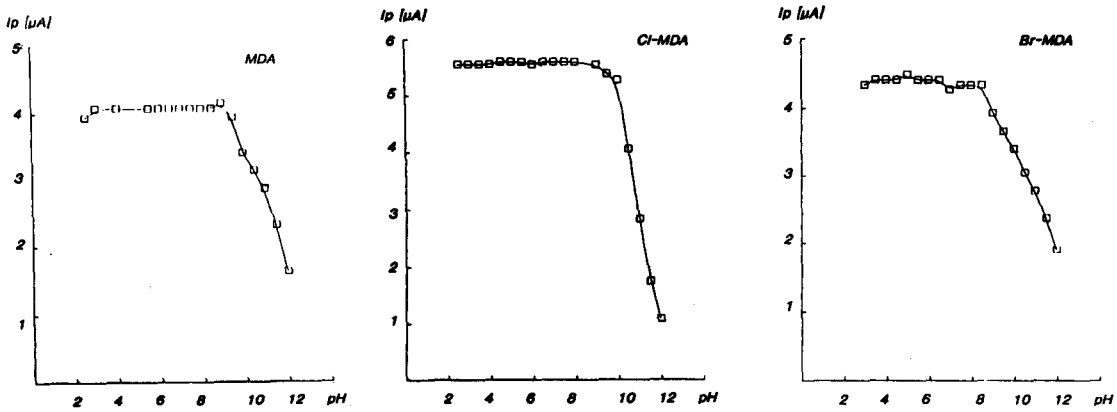

Fig. 5. Peak current–pH dependence for the amphetamine derivatives. Glassy carbon electrode, 25°.

in all the pH range the E_ps follow the order: MDA < Cl MDA < Br MDA. From the above results, it seems obvious that direct oxidation of the aromatic ring, influenced by the C-6 substituent, is occurring. Although, the suggestion that a carbocation is formed in an aqueous medium is arguable, it is quite justified in this case due to the stabilizing effect of the methylenedioxy group. This effect is well documented for a similar substituent, the methoxyl group.[12]

Linear sweep cyclic voltammetric experiments showed a single oxidation peak for all drugs, recording the voltammograms at sweep rates between 50 mV/sec and 5 V/sec at pH 7. From this study it can be seen that the potential peak shifts anodically by about 30 mV for each 10-fold increase in sweep rate. These results agree with an EC mechanism[13] with a one-electron transfer in the electrochemical step and a subsequent chemical reaction, *e.g.*, the electrochemical formation of a cation radical with a subsequent chemical reaction of this species. The chemical reaction of the cation radical is fast enough for the reduction of the cation radical not to occur in the time scale of the experiment.

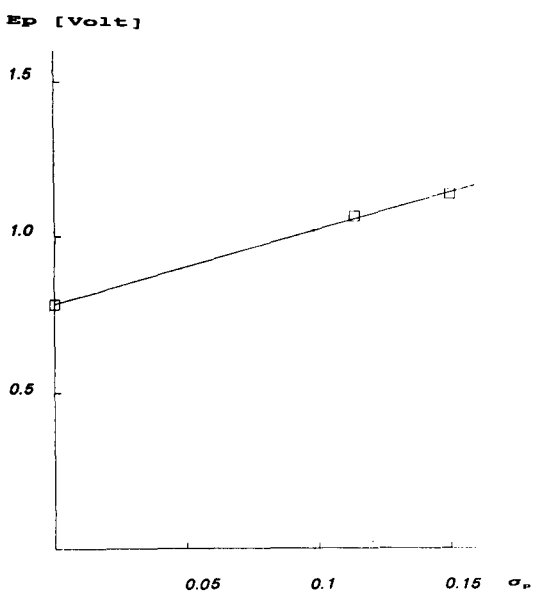

Fig. 6. Linear relationship between the extra-thermodynamic parameter, σ_p^+, and the peak potential of the amphetamine derivatives. pH 7 on glassy carbon electrode.

In order to confirm the relationship between E_p and the effect of the C-6 substituent on the oxidizable moiety, we consider σ_p^+ as an extra-thermodynamic parameter which combines the inductive and resonant effects of the substituents on the benzene ring. This factor can be applied to oxidation reactions involving resonance stabilization of a positive charge in an aromatic ring.[14] As can be seen in Fig. 7, there is a linear relationship between the σ_p^+ factor of the C-6 substituents and the E_ps experimentally obtained by differential pulse voltammetry. From the above behavior it is possible to extrapolate an E_p value for the 6-nitro derivative. The extrapolated value was 1460 mV, which is located in the discharge zone of the support electrolyte, explaining the absence of any oxidation peak for this derivative.

The voltammetric technique also can be used as an analytical tool to quantify these derivatives in aqueous solution. For this purpose we studied the dependence of the voltammetric peak on the concentration of the amphetamine derivatives. A linear relationship between the peak current and the concentration of the derivatives for 0.01 m*M* and 0.1 m*M* solutions at pH 7 were found (Table 2).

Peak potentials were independent of the concentration indicating that no adsorption or second order processes are involved. Reproducibility studies were carried out with the carbon

Table 2. Linear relations between peak current and concentrations of the drugs for 10 points between 0.01 m*M* and 0.1 m*M*. pH = 7.0, 25° and CPE as the working electrode. i_p (μA) = slope × C (*M*) + intercept

Drugs	Intercept	Slope × 10^{-5}	Correlation
MDA	1.306	1.668	0.996
Br–MDA	−0.168	1.625	0.994
Cl–MDA	−0.938	1.386	0.995

paste and glassy carbon electrodes, obtaining an average CV = 0.5 and 0.4% for the peak potentials and a CV = 3.8 and 3.4% for the peak currents, respectively. These results indicate good reproducibility and accuracy to develop a quantitative voltammetric assay for these derivatives. Furthermore, the behavior presented here would be useful for developing a method of HPLC with electrochemical detection. On the other hand, stability assays allow us to conclude that aqueous solutions of these compounds remain unchanged after 30 days at room temperature under normal room light.

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