POLAROGRAPHIC AND VOLTAMMETRIC DETERMINATION OF 1-(4-METHOXYPHENYL)-3-METHYLTRIAZENE

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The polarographic behaviour of 1-(4-methoxyphenyl)-3-methyltriazene in a mixed aqueous–methanolic medium was investigated by tast polarography and differential pulse polarography at a dropping mercury electrode, constant-potential coulometry at a large area mercury electrode and cyclic voltammetry at a hanging mercury drop electrode. A mechanism has been proposed for its polarographic reduction and optimal conditions have been found for its determination by tast polarography in the concentration range 2–100 μmol l^{-1} and by differential pulse polarography at a dropping mercury electrode, differential pulse voltammetry at a hanging mercury drop electrode and linear scan voltammetry at a hanging mercury drop electrode in the concentration range 0.2–100 μmol l^{-1}. The sensitivity of the determination can be further improved through adsorptive accumulation of the test substance on the surface of the hanging mercury drop electrode; fifteen-second accumulation in stirred solution allows determination in the range 0.01–0.1 μmol l^{-1}.

Key words: 1-(4-Methoxyphenyl)-3-methyltriazine; Differential pulse polarography; Adsorptive stripping voltammetry.

The derivatives of 1-phenyl-3-methyltriazene are genotoxic substances that are assumed to act through an alkylation mechanism[1–3]. In addition, this type of substances exhibits carcinostatic action[4–6]; in this connection, detailed studies have been carried out of its acute toxicity[7], metabolism[8] and protolytic splitting mechanism[9,10]. Thus, there is a need for sensitive analytical methods for the determination of trace amounts of these substances. Because of the ready reducibility of triazenes[11], modern polarographic and voltammetric methods are suitable for this purpose. These methods have been found useful for the determination of trace amounts of pharmaceutically important 2′-carbamoyl[12], 3′-carbamoyl[13], 4′-carbamoyl[14], 4′-bromo[15], 4′-phenylazo[16,17] and 2′-nitro[18] derivatives of 1-phenyl-3,3-dimethyltriazene. A number of other works dealing with the mechanism or analytical utilization of the polarographic reduction of various types of triazenes can be found in the monograph[11] or in ref.[12]. In the framework of a

systematic study of the use of modern polarographic and voltammetric methods for the analysis of chemical carcinogens, this work describes the use of tast polarography and differential pulse polarography (DPP) at a classical dropping mercury electrode (DME), differential pulse voltammetry (DPV) at a hanging mercury drop electrode (HMDE), linear scan voltammetry (LSV) at HMDE and adsorptive stripping voltammetry (AdSV) for the determination of the so-far unstudied compound 1-(4-methoxyphenyl)-3-methyltriazene (**1**) [CAS Name: 1-Triazene, 1-(4-methoxyphenyl)-3-methyl; CAS Registry Number: 53477-43-3]. In addition, cyclic voltammetry at HMDE was used to investigate the reversibility of the reduction and potentiostatic coulometry was employed to determine the number of electrons exchanged. In view of the sparing solubility of the test substance in water, a mixed aqueous–methanolic solvent was used, as proved convenient for previously investigated 1-phenyl-3,3-dimethyltriazene derivatives[12–18].

EXPERIMENTAL

Reagents

The stock solution of 1-(4-methoxyphenyl)-3-methyltriazene in methanol (c = 1 mmol l^{-1}) was prepared by dissolving 0.0413 g of the substance (Research Institute for Organic Syntheses, Pardubice-Rybitví) in p.a. solvent. The purity of the substance was controlled by paper and thin-layer chromatography and by measuring the spectra of a methanolic solution in UV region[19]. More diluted solutions were prepared by exact dilution of the stock solution. All the solutions were stored in the dark. It followed from a spectrophotometric study of the stability of these solutions that the solution with a concentration of 1 mmol l^{-1} must be prepared fresh once a month, 0.1 mmol l^{-1} every week and 0.01 mmol l^{-1} daily. The other chemicals employed were of p.a. purity (Lachema, Brno, Czech Republic). Water was doubly distilled in a quartz apparatus. The methanol used was stored only in glass vessels as contact with polyethylene led to extraction of substances that unfavourably affected the determination of the lowest concentrations.

Apparatus

A PA 3 polarographic analyzer was employed together with an XY 4106 recorder (both from Laboratorní pristroje, Prague, Czech Republic). Measurements were carried out using a three-electrode arrangement with a platinum wire auxiliary electrode and saturated silver chloride reference electrode, to which all the potential values are referred. The parameters of the classical DME used in tast and DP polarography were as follows: At a mercury reservoir height of h = 36 cm, the flow rate was m = 5.51 mg s^{-1} and the drop time was τ = 2.12 s (at an applied voltage of 0 V in 0.1 M KCl). Where not stated otherwise, work with the DME was carried out at a polarization rate 5 mV s^{-1}, controlled drop time of 1 s, mercury reservoir height of 36 cm and modulation amplitude in differential pulse polarography of –100 mV.

DPV, AdSV and cyclic voltammetric measurements were carried out using a static mercury drop electrode SMDE 1 (Laboratorní pristroje, Prague, Czech Republic) connected as a hanging mercury drop electrode (HMDE). The capillary employed had a diameter of 0.146 mm and the maximum drop size attainable was employed, obtained by opening the valve for 160 ms. Where not stated otherwise, work with the HMDE was carried out at a polarization rate 20 mV s^{-1} and modulation amplitude in

differential pulse voltammetry of –50 mV. Oxygen was removed from the measured solutions by bubbling for ten minutes with nitrogen, which was purified by passing through a solution of chromium(II) ions in dilute hydrochloric acid over zinc amalgam. A prebubler containing a water–methanol mixture in the same ratio as in the polarographed solution was placed prior to polarographic vessel.

Coulometric measurements were performed on an OH 404 coulometric analyzer (Radelkis, Budapest, Hungary) in a 200 ml vessel. A mercury pool at the vessel bottom served as the cathode, a platinum sheet electrode served as the anode. The cathode and anode compartments were separated with a frit. The saturated calomel electrode used was an OH 993 (Radelkis, Budapest, Hungary). The solution was stirred with a magnetic stirrer during the electrolysis, and the measurements were conducted under nitrogen.

Spectrophotometric measurements were conducted on a Specord M 400 instrument (Zeiss, Jena, Germany) using 1 cm quartz cells.

The solution pH was measured with an OP 211/1 digital pH meter using a combined pH electrode Crytur 01-29 (Monokrystaly, Turnov, Czech Republic). Calibration was carried out using acetate, borate and phosphate buffers in 50% methanol[20].

All the measurements were carried out at laboratory temperature.

Procedures

The calibration curves were measured in triplicate and evaluated by the least squares linear regression method. The limit of determination was calculated as the tenfold standard deviation from 7 analyte determinations at the concentration corresponding to the lowest point on the appropriate calibration straight line[21].

The procedure for the determination of the number of electrons exchanged using constant potential coulometry at a large area mercury electrode was as follows: 50 ml of Britton–Robinson buffer, pH 7.1, and 40 ml of methanol were measured into the coulometric vessel and the solution was bubbled with nitrogen with constant stirring. Simultaneously, pre-electrolysis was commenced at a selected constant potential. After about 20 min, the residual current value decreased below 0.2 mA and no longer changed. Then the appropriate circuit parameters were adjusted for automatic compensation of the residual current and 10.00 ml of the solution of 1-(4-methoxyphenyl)-3-methyltriazene in methanol (c = 1 mmol l^{-1}), previously freed of oxygen, were added with constant bubbling and stirring. Electrolysis was terminated when the current value decreased to that of the residual current (ca after 60 min) and the charge consumed was found by digital integration of the current passed. The course of the reduction was monitored spectrophotometrically and polarographically by taking 10 ml of solution from the coulometric vessel at the given time intervals and measuring tast polarogram and UV and visible spectrum. The sampling was carried out prior to the beginning of the coulometric reduction and after reduction of 25, 50, 75 and 100% of the substance (calculated relative to the determined number of electrons exchanged at the constant potential).

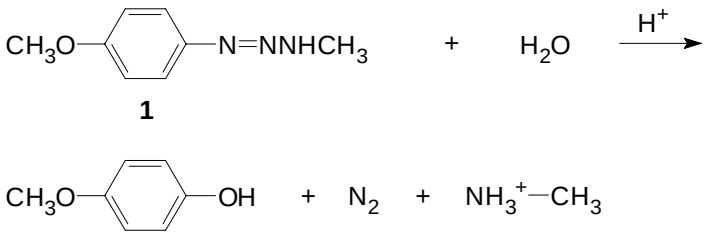
RESULTS AND DISCUSSION

Study of the Polarographic Behaviour of 1-(4-Methoxyphenyl)-3-methyltriazene

Tast polarography. It can be seen from Fig. 1 that 1-(4-methoxyphenyl)-3-methyltriazene gives one or two waves in dependence on pH. The limiting current (I_{lim}^{1}) of the first wave at pH < 4.5 is practically negligible; it rapidly increases at pH > 5.4 and reaches its maximum value at pH 7.6. It rapidly decreases at higher pH values and at

pH > 10 this wave practically disappears. The half-wave potential ($E^1_{1/2}$) of this wave shifts to more negative values with increasing pH and this dependence has asymptotic character (see Table I). Simultaneously, at pH > 7.6 a new wave appears with a half-wave potential ($E^2_{1/2}$) of about –1.45 V. The limiting current (I^2_{lim}) of this wave is practically constant within pH range of 10–12. However, this wave is difficult to utilize analytically, as it coincides with the decomposition of the base electrolyte. For the same reason it was not possible to carry out its logarithmic analysis. However, the shape of this wave indicates an irreversible process. This wave apparently corresponds to direct polarographic reduction of unprotonated molecules of 1-(4-methoxyphenyl)-3-methyltriazene.

The decrease in the limiting current of the first wave at pH < 5.4 can be explained in terms of protolysis of the test substance[10] according to Eq. (*A*).



(*A*)

This protolytic decomposition results in the decrease of the limiting current of the first wave with time at pH < 7. Assuming that this decomposition is a first-order reaction, the following equation can be used:

$$\ln I_t = \ln I_0 - kt$$

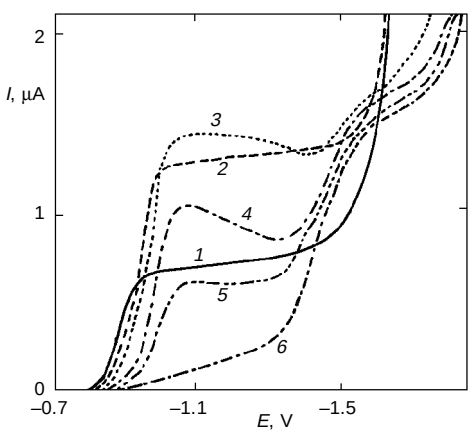


FIG. 1

Tast polarograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 0.1 mmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1). pH: *1* 5.4; *2* 6.3; *3* 7.6; *4* 8.6; *5* 9.2; *6* 10.8

where I_0 is the limiting current at the beginning of the reaction of protolysis, I_t the limiting current at time t and k the first-order rate constant. The linear dependence of ln I_t on time, which has been established, indicates that the reaction is pseudo-monomolecular in its behaviour. The rate constant, calculated as the negative value of the slope of the above mentioned straight line, is equal to 0.044 min^{-1} at pH 5.4.

It follows from the logarithmic analysis of the tast polarograms that the electrode process is irreversible in the pH range 5–10. This was also confirmed by the cyclic voltammetry at a hanging mercury drop electrode (see further). At pH 7.6, the limiting current of the first wave measured by classical DC polarography is directly proportional to the square root of the mercury reservoir height confirming that this is a diffusion-controlled process. However, the dependence of the current values on the square root of the mercury reservoir height for the region of current decrease around –1.3 V does not pass through the origin, indicating mixed diffusion-kinetic control. The fact that the slope of the $E_{1/2}$ vs pH dependence does not correspond to the relationship $dE_{1/2}/dpH = -mRT/\alpha n_a F$ indicates[22] that a substance adsorbed on the electrode surface undergoes the preliminary protonation reaction. The observed decrease in the limiting

TABLE I

The effect of pH on the tast polarograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 0.1 mmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1)

pH[a]	pH[b]	$E^1_{1/2}$, V	I^1_{lim}, μA	α_1[c], mV	$E^2_{1/2}$, V	I^2_{lim}, μA
2.05	2.5	–[d]	–[d]	–[d]	–[f]	–[f]
3.25	4.0	–[d]	–[d]	–[d]	–[f]	–[f]
3.99	4.5	–[d]	–[d]	–[d]	–[f]	–[f]
5.07	5.4	–0.88	0.61	80.1	–[f]	–[f]
5.74	6.3	–0.92	1.24	85.0	–[f]	–[f]
6.87	7.6	–0.94	1.29	80.3	–[g]	–[g]
8.09	8.6	–0.97	0.94	70.8	–1.45[h]	0.78[h]
9.08	9.2	–0.98	0.52	70.4	–1.45[h]	0.99[h]
10.02	10.0	–0.99	0.14	70.3	–1.45[h]	1.11[h]
10.80	10.8	–[e]	–[e]	–[e]	–1.45[h]	1.12[h]
11.66	11.4	–[e]	–[e]	–[e]	–1.45[h]	1.12[h]

[a] Value corresponding to the Britton-Robinson buffer used. [b] Value corresponding to the mixed aqueous-methanolic solution. [c] Slope of the logarithmic analysis. [d] The more positive wave is too low to be evaluated. [e] The more positive wave completely disappears. [f] The more negative wave is not observable. [g] The more negative wave is too low to be evaluated. [h] The values given are only approximate because the more negative wave is difficult to evaluate as it coincides with the decomposition of the base electrolyte.

current in the region around –1.3 V at pH around 9 is apparently connected with a decrease in the rate of surface protonation as a consequence of desorption of the substance at potentials that are far more negative than potential of the electrocapillary zero, where maximum adsorption of uncharged molecules can be expected.

Constant potential coulometry. Potentiostatic coulometry at a large-area mercury electrode at pH 7.6 and a constant potential of –1.2 V yielded a value of $n = 4.2$ for the number of exchanged electrons (see Fig. 2). Tast polarographic monitoring of the coulometric reduction of 1-(4-methoxyphenyl)-3-methyltriazene ($c = 0.1$ mmol l^{-1}) at constant potential –1.2 V in mixed Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6 (see Fig. 3) revealed the disappearance of the polarographically active triazene group and the formation of a product which gave anodic wave at –0.2 V.

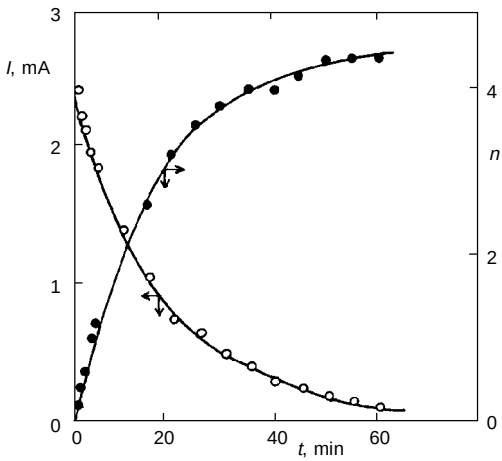


FIG. 2
Dependence of current I and calculated number of electrons exchanged per 1 molecule n on time during the coulometric reduction of 1-(4-methoxyphenyl)-3-methyltriazene ($c = 0.1$ mmol l^{-1}) at constant potential –1.2 V in mixed Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6

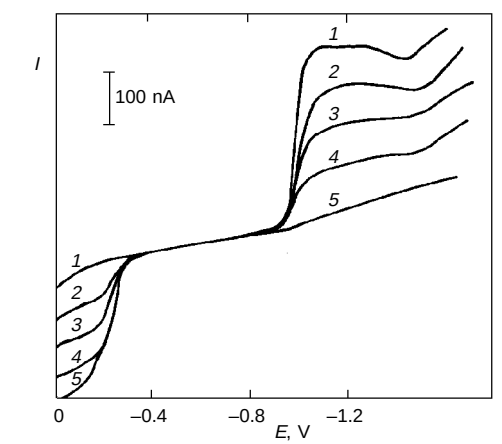
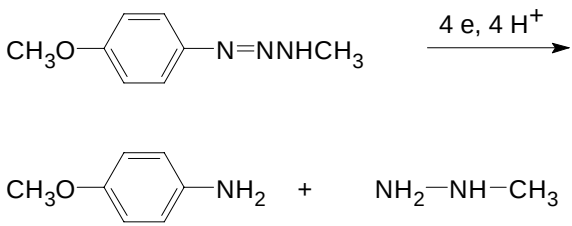


FIG. 3
Tast polarographic monitoring of the coulometric reduction of 1-(4-methoxyphenyl)-3-methyltriazene ($c = 0.1$ mmol l^{-1}) at constant potential –1.2 V in mixed Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6. Polarograms correspond to the passage of charge required to reduce 0% (*1*), 25% (*2*), 50% (*3*), 75% (*4*) and 100% (*5*) of the test substance (calculated with respect to the n value established at the potential applied)

Cyclic voltammetry at a hanging mercury drop electrode. Cyclic voltammogram of 1-(4-methoxyphenyl)-3-methyltriazene in mixed Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6 (see Fig. 4) demonstrates the irreversible character of the observed process. The height of the cathodic peak at –1 V is not directly proportional to either the polarization rate or the square root of this quantity, so that it can be assumed that this is a complex overall process controlled both by the diffusion of the test substance to the electrode surface and also by its adsorption on this surface. The anodic peak at –0.2 V is apparently connected with the presence of an aromatic amine formed by the reduction of 1-(4-methoxyphenyl)-3-methyltriazene.

Proposed reduction mechanism. It follows from the above observations that 1-(4-methoxyphenyl)-3-methyltriazene undergoes four-electron irreversible reduction at pH around 7. It can be assumed by analogy with the polarographic behaviour of previously studied 1-phenyl-3,3-dimethyltriazene derivatives[12–18] that the polarographic reduction of the test substance will occur according to Eq. (*B*).



(*B*)

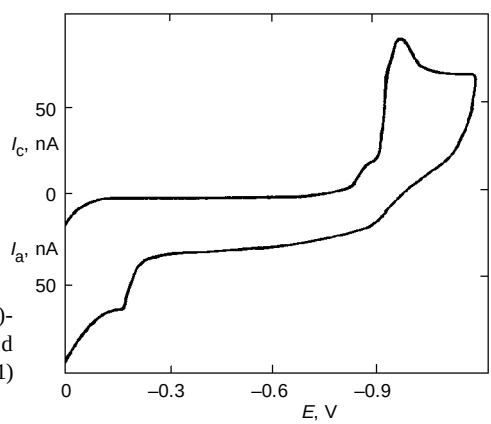


FIG. 4
Cyclic voltammogram of 1-(4-methoxyphenyl)-3-methyltriazene (c = 10 μmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6 and polarization rate of 50 mV s^{-1}

Analytical Utilization of the Polarographic Reduction of 1-(4-Methoxyphenyl)-3-methyltriazene

Tast polarography. From an analytical point of view, the highest and most easily measured curves were obtained in medium with pH 7.6. Simultaneously, solutions polarographed in this medium are sufficiently stable (the height of the wave of 0.1 mmol l^{-1} 1-(4-methoxyphenyl)-3-methyltriazene does not change after 60 min within experimental error), in contrast to acidic solutions, where acid hydrolysis leads to decrease in the wave height after 20 or 30 min by 43 and 67%, respectively, at pH 5.4. The calibration dependencies are linear at pH 7.6 in the range 2–100 μmol l^{-1} and their parameters are given in Table II together with the calculated limit of determination.

Differential pulse polarography at a dropping mercury electrode. It can be seen from Table III that 1-(4-methoxyphenyl)-3-methyltriazene yields one or two peaks in differential pulse polarography, in dependence on the pH value. The effect of pH on the peak position (E_p) and height (I_p) reflects the effect of pH on the behaviour of the test substance in tast polarography. For analytical purposes, the first peak is preferable since the second peak is rather close to the decomposition of the base electrolyte. It can be seen from Fig. 5 that highest, best developed and most readily evaluated first peak was

TABLE II
Parameters of the calibration straight lines for the determination of 1-(4-methoxyphenyl)-3-methyltriazene by various polarographic and voltammetric methods in Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6

Method	c, μmol l^{-1}	Slope mA mol^{-1} l	Intercept nA	Correlation coefficient	L_Q[a], μmol l^{-1}
Tast	20–100	12.2	–28.1	0.9966	–
	2–10	14.8	–0.8	0.9998	2
DPP/DME	20–100	19.0	–43.8	0.9983	–
	2–10	22.0	–2.6	0.9984	–
	0.2–1[b]	22.3	–0.4	0.9973	0.2
LSV/HMDE	2–10	5.51	0.5	0.9978	–
	0.2–1[b]	7.23	0.2	0.9980	0.3
DPV/HMDE	2–10	5.3	–0.9	0.9992	–
	0.2–1[b]	3.9	–0.1	0.9990	0.1
AdSV/HMDE	0.01–0.1[c]	140.5	–0.2	0.9996	0.01

[a] Limit of determination. [b] Tenfold diluted Britton–Robinson buffer–methanol (1 : 1) medium, pH 7.2. [c] Tenfold diluted Britton–Robinson buffer–methanol (99 : 1), pH 7.1, 15 s accumulation in stirred solution at –0.4 V.

obtained in Britton–Robinson buffer–methanol medium (1 : 1) at pH 7.6. Simultaneously, the height of this peak does not change after 60 min within experimental error. Nevertheless, the polarographic curves at lower analyte concentration (bellow 1 μmol l^{-1}) should be recorded soon after preparation of the polarographed solution, preferably

TABLE III
The effect of pH on the differential pulse polarograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 0.1 mmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1)

pH[a]	pH[b]	E_{p1}, V	I_{p1}, μA	E_{p2}, V	I_{p2}, μA
2.05	2.5	–[c]	–[c]	–[d]	–[d]
3.25	4.0	–0.67	0.05	–[d]	–[d]
3.99	4.5	–0.70	0.05	–[d]	–[d]
5.07	5.4	–0.84	1.18	–[d]	–[d]
5.74	6.3	–0.87	2.03	–[d]	–[d]
6.87	7.6	–0.94	2.08	–1.45	0.46
8.09	8.6	–0.95	1.37	–1.45	1.02
9.08	9.2	–0.97	0.47	–1.45	1.04
10.02	10.0	–0.98	0.15	–1.45	1.34
10.80	10.8	–[e]	–[e]	–1.45	1.12
11.66	11.4	–[e]	–[e]	–1.45	0.87

[a] Value corresponding to the Britton–Robinson buffer used. [b] Value corresponding to the mixed aqueous–methanolic solution. [c] The more positive peak is too low to be evaluated. [d] The more negative peak is not observable. [e] The more positive peak completely disappears.

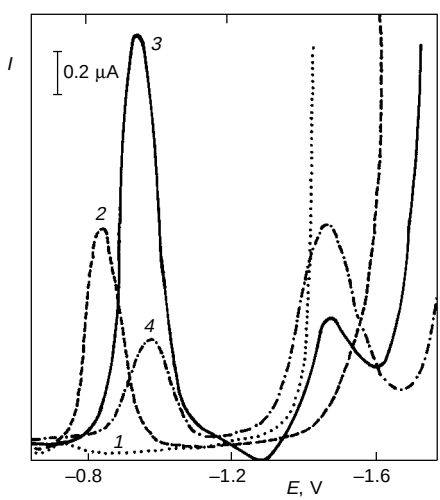


FIG. 5
Differential pulse polarograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 0.1 mmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1). pH: *1* 4.5; *2* 5.4; *3* 7.6; *4* 10.0

after a constant time from the preparation has elapsed. The calibration curves are linear under these conditions in the range 0.2–100 μmol l^{-1} and their parameters are given in Table II. (The peak height was measured from the straight line connecting the minima on both sides of the peak.) It is preferable to carry out DPP determination in the concentration range 0.2–1 μmol l^{-1} in a mixture of tenfold diluted Britton–Robinson buffer–methanol (1 : 1) with pH 7.2. (As the concentration of the aqueous Britton–Robinson buffer decreases, the peak heights corresponding to impurities in the chemicals employed for its preparation also decrease, leading to a smoother base electrolyte curve.)

Linear scan voltammetry at a hanging mercury drop electrode. The effect of pH on the behaviour of 1-(4-methoxyphenyl)-3-methyltriazene is depicted in Fig. 6. It was found that the test substance gives two to three peaks in dependence on pH. The first, most positive and rather low peak is probably connected with adsorptive phenomena rather than with faradaic processes. The third, most negative peak is difficult to evaluate since it coincides with the decomposition of the base electrolyte. From an analytical point of view, the highest, best developed and most easily evaluated second peak was obtained in Britton–Robinson buffer–methanol (1 : 1) medium at pH 7.6. The height of this peak can easily be evaluated as the distance between the peak maximum and the straight line constructed by the prolongation of the linear part of the base electrolyte curve before the onset of the peak. The calibration curves constructed in this way are linear within the concentration range 0.2–10 μmol l^{-1} and their parameters are given in Table II. For the above given reasons, tenfold diluted Britton–Robinson buffer–methanol (1 : 1) medium at pH 7.2 was used in the lowest concentration region.

Differential pulse voltammetry at a hanging mercury drop electrode. The effect of pH on the differential pulse voltammograms of the analyte is documented by Fig. 7.

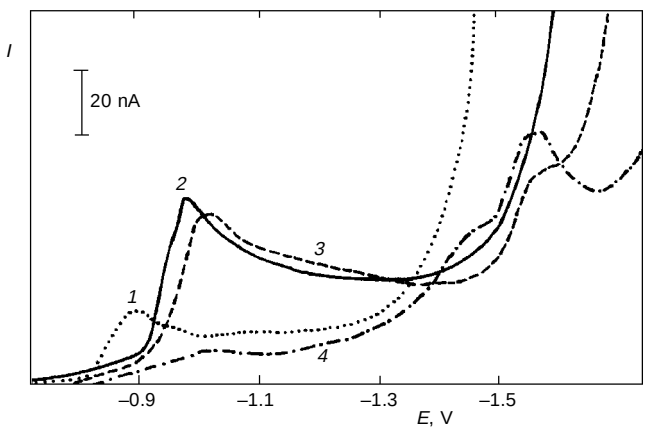


FIG. 6

Linear scan voltammograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 10 μmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1). pH: *1* 5.4; *2* 7.6; *3* 9.2; *4* 10.8

Analogously do LSV at HMDE, 1-(4-methoxyphenyl)-3-methyltriazene yields two to three peaks in dependence on pH. The height, position and shape of these peaks reflect the behaviour of the test substance at previously investigated polarographic and voltammetric techniques. Again, the first peak (most positive one, which is probably connected with adsorptive processes) and the third peak (most negative one, which coincides with the decomposition of the base electrolyte) are of no analytical use. The highest, best developed and most easily evaluated second peak was obtained at pH 7.6 (see Fig. 7). Therefore, Britton–Robinson buffer–methanol (1 : 1) medium at pH 7.6 was chosen as optimum. The height of the second peak was again evaluated from the straight line connecting the minima on both sides of the peak. The calibration curves constructed in this way are linear within the concentration range 0.2–10 μmol l^{-1} and their parameters are given in Table II. As with previously used techniques, tenfold diluted Britton–Robinson buffer–methanol (1 : 1) medium at pH 7.2 was used in the lowest concentration region.

Adsorptive stripping voltammetry at a hanging mercury drop electrode. A further increase in the sensitivity of the determination can be attained through adsorptive accumulation of the test substance on the surface of a hanging mercury drop electrode. The following dependence of the peak height I_p on the time t elapsed between formation of the hanging mercury drop and recording of the voltammogram was found in tenfold diluted Britton–Robinson buffer–methanol (99 : 1) stirred medium at pH 7.1 and an analyte concentration of 0.1 μmol l^{-1}.

t, s	0	10	15	20	30
I_p, nA	4.9	8.3	11.8	9.5	9.0

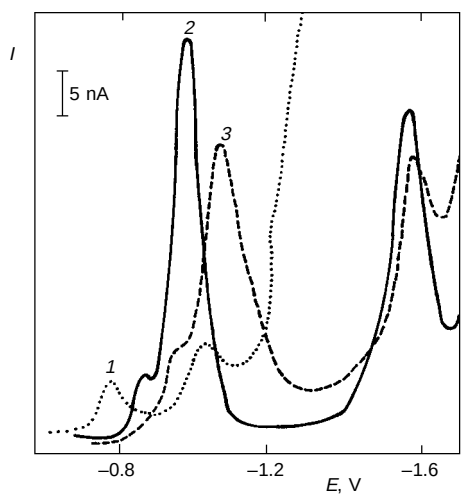


FIG. 7
Differential pulse voltammograms of 1-(4-methoxyphenyl)-3-methyltriazene (c = 10 μmol l^{-1}) in mixed Britton–Robinson buffer–methanol medium (1 : 1). pH: *1* 4.5; *2* 7.6; *3* 9.2

A decrease in the peak current observed at accumulation times longer than 15 s can be explained by maximum possible coverage of the working electrode by the adsorbed substance and by the passivation of the electrode whose surface is not renewed during the measurement. The adsorptive accumulation can be increased by decreasing the methanol content in the voltammetric solution, which can be explained both by the higher solubility of the analyte in methanol and also by the competitive adsorption of the molecules of this solvent on the HMDE surface.

The following dependence of the peak height I_p on the potential of accumulation E_{acc} was found in tenfold diluted Britton–Robinson buffer–methanol (99 : 1) stirred medium at pH 7.1 and an analyte concentration of 0.1 μmol l^{-1}.

E_{acc}, V	0	–0.1	–0.2	–0.3	–0.4	–0.5	–0.6
I_p, nA	10.5	10.5	12.0	13.3	14.0	13.7	11.6

Therefore, the following optimum conditions were chosen for AdSV determination of 1-(4-methoxyphenyl)-3-methyltriazene: E_{acc} = –0.4 V, t_{acc} = 15 s, stirred solution of tenfold diluted Britton–Robinson buffer–methanol (99 : 1) medium at pH 7.1. At the end of the accumulation period in a stirred solution, the stirrer was switched off and the differential pulse voltammogram was recorded after 15 s had elapsed to allow the solution to become quiescent. In such conditions the peak height, measured from the straight line connecting the minima on both sides of the peak, was linearly dependent on analyte concentration across the region of 0.01–0.1 μmol l^{-1}. The calibration straight line parameters are given in Table II. The determination cannot be additionally sensitised by extending the time of accumulation, decreasing the content of methanol or diluting the Britton–Robinson buffer used.

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