
CHEMICAL DESTRUCTION OF 4-NITROBIPHENYL IN LABORATORY WASTE AND ITS MONITORING BY DIFFERENTIAL PULSE POLAROGRAPHY AND VOLTAMMETRY AND BY HIGH-PERFORMANCE LIQUID CHROMATOGRAPHY*

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A method for destruction of the carcinogenic 4-nitrobiphenyl in laboratory waste was elaborated based on its reduction with powdered zinc and oxidation of the produced 4-aminobiphenyl with potassium permanganate. The efficiency of the method can be checked by high-performance liquid chromatography, differential pulse polarography, and differential pulse voltammetry.

The study of physical, chemical, and biological properties of chemical carcinogens inevitably causes production of laboratory wastes contaminated by these compounds. Therefore, methods suitable for their destruction have been in the centre of interest during last several years¹⁻⁴. One of the suspected human carcinogens is 4-nitrobiphenyl, which is enzymatically reducible with the formation of 4-aminobiphenyl⁵. The proposed destruction of the latter⁶ by means of permanganate is efficient from the chemical point of view, however it leads to products which are mutagenic⁷, *i.e.* noxious from the biological point of view. Therefore, we preferred to reduce 4-nitrobiphenyl to 4-aminobiphenyl, which can easily be liquidated by oxidation with permanganate⁹ to give definitely nonmutagenic products¹⁰. Powdered zinc was chosen as the most suitable reducing agent.

EXPERIMENTAL

Apparatus and Chemicals

Polarographic and voltammetric records were made on a polarographic analyser of the type PA 2 connected with a type XY 4103 pen recorder (Laboratorní přístroje, Prague) in a three-electrode mode. All measurements were carried out at the room temperature with a saturated calomel reference electrode and a platinum wire auxiliary electrode. The polarization rate in

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differential pulse polarographic measurements was 10 mV s^{-1} , the drop time was kept equal to 1 s unless otherwise indicated, the height of the mercury column was 36 cm, and the modulation amplitude was 100 mV. The dropping mercury electrode had a rate of flow $m = 2.83 \text{ mg s}^{-1}$ and drop time $t = 2.82 \text{ s}$ ($h = 36 \text{ cm}$) in 0.1 M-KCl at 0 V against s.c.e. Differential pulse voltammetric measurements were carried out with a stationary glassy carbon disc electrode R.D.E. 1 (Laboratorní přístroje, Prague) at a polarization rate of 5 mV s^{-1} , pulse amplitude 12.5 mV, and interval between pulses 1 s. The analysed solutions were deaerated by 5 min bubbling with nitrogen which passed through an alkaline anthraquinone-2-sulphonate solution and a solution of Cr(II) ions in dilute HCl in contact with Zn amalgam. Chromatographic measurements were carried out on a high-performance liquid chromatograph (Pye Unicam, USA) with a type LC UV detector and LC-XPD pump in combination with a TZ 4 100 pen recorder (Laboratorní přístroje, Prague). A 150 mm long all-glass column of 3.4 mm internal diameter with a 30 mm long pre-column of the same internal diameter were used. Both were filled with Separon SI, C 18 (reversed phase) of $10 \mu\text{m}$ particle diameter. A mobile phase methanol-acetonitrile-aqueous solution of $1.5 \text{ mmol l}^{-1} \text{ KH}_2\text{PO}_4$ and $1.5 \text{ mmol l}^{-1} \text{ K}_2\text{HPO}_4$ (3 : 1 : 2) at a rate of flow of 1 ml min^{-1} was used. The detection wavelength was 300 nm for 4-nitrobiphenyl and 254 nm for 4-aminobiphenyl. The stock solutions of $5 \cdot 10^{-3} \text{ mol l}^{-1}$ 4-aminobiphenyl and $5 \cdot 10^{-3} \text{ mol l}^{-1}$ 4-nitrobiphenyl in glacial acetic acid were prepared by dissolution of weighed quantities of reagent grade substances (Sigma, USA); other solutions were prepared by dilution. All chemicals were of reagent grade.

Destruction of 4-Nitrobiphenyl

10 ml of $5 \cdot 10^{-3} \text{ M}$ -4-nitrobiphenyl in glacial acetic acid was mixed with 165 mg of powdered Zn and 10 ml of $2 \text{ M-H}_2\text{SO}_4$ and the mixture was stirred for 12 h with a magnetic stirrer. Afterwards, 10 ml of 0.2 M-KMnO_4 was added and the stirring continued for another 12 h.

Checking the Destruction Efficiency by Differential Pulse Voltammetry

Manganese dioxide and residual permanganate in the solution after destruction were reduced with 10 ml of 0.5 M-(COOH)_2 and the solution was made up with water to 50 ml. Both 4-nitrobiphenyl and 4-aminobiphenyl were determined by differential pulse voltammetry using a stationary glassy carbon electrode⁸. A calibration curve was constructed in the concentration range $5 \cdot 10^{-6} - 2 \cdot 10^{-5} \text{ mol l}^{-1}$ corresponding to the degree of destruction 99.5–98.0%; the solutions used contained the same concentrations of acetic, oxalic, and sulphuric acids, zinc and manganese ions as the analysed solution after degradation.

Checking the Destruction Efficiency by High-Performance Liquid Chromatography with UV Detection

Manganese dioxide and unreacted permanganate in the solution after destruction were reduced by adding the necessary quantity of solid ascorbic acid (until decoloration). The solution was partly neutralized by adding about 10 M-NaOH to pH 3–4 and made up to 100 ml with distilled water.

Solutions for construction of the calibration curve were prepared in the concentration range $5 \cdot 10^{-6} - 2 \cdot 10^{-5} \text{ mol l}^{-1}$ and contained the same concentrations of acetic, sulphuric, and oxalic acids, zinc and manganese ions as the analysed solution after degradation. The volume of the solution injected into the column was 20 μl .

RESULTS AND DISCUSSION

Differential pulse polarography was used to study the influence of acetic acid on the efficiency of reduction of 4-nitrobiphenyl with zinc. The relative concentrations of the nonreduced compound after 60 min were as follows:

$[\text{CH}_3\text{COOH}]$, vol. %	10	30	50	70	90
C_t/C_0 , %	0.52	0.45	0.13	0.48	0.49

It can be seen that the medium of 50 vol. % acetic acid is the optimum. The rate of reduction in this medium is illustrated by the following data:

t , min	0	15	30	45	60	75	90
C_t/C_0 , %	100	2.25	0.21	0.17	0.13	<0.1	<0.1

Hence, 4-nitrobiphenyl is practically quantitatively reduced after 90 min to 4-aminobiphenyl, whose formation was substantiated also by comparing the UV spectrum of its $2 \cdot 10^{-5} \text{ mol l}^{-1}$ solution in 1 mol l^{-1} sulphuric acid after reduction with 25 mg powdered Zn for 12 h with the spectrum of $2 \cdot 10^{-5} \text{ mol l}^{-1}$ 4-aminobiphenyl in the same medium. A calibration curve for the determination of 4-nitrobiphenyl was also obtained in the medium considered.

To ensure complete reduction of 4-nitrobiphenyl with zinc and oxidation of the product with permanganate, both reactions were allowed to proceed for 12 h. The unreacted 4-nitrobiphenyl was determined by differential pulse voltammetry on a stationary glassy carbon electrode (from its reduction wave), and the unreacted 4-aminobiphenyl was determined analogously from its oxidation wave⁸. The voltammograms are shown in Fig. 1; it can be seen that at least 99.8% of the starting 4-nitrobiphenyl and of the produced 4-aminobiphenyl was destroyed by the method indicated. Ames' test⁷ showed that the solution thus treated is not mutagenic, hence the products formed are biologically harmless. (From the consumption of permanganate exceeding 5 moles per 1 mol of 4-aminobiphenyl and from the presence of ammonia in the reaction mixture it can be inferred that the oxidation products are mainly water, CO_2 and NH_3 .)

Both substances can be determined rapidly and simply by differential pulse voltammetry using a single electrode, which is an advantage of the method. The sensitivity of the method, whose detection limit is around $10^{-6} \text{ mol l}^{-1}$, and its selectivity is sufficient in most cases for following the efficiency of destruction of the considered carcinogen in laboratory waste water.

If the waste water contains, in addition to 4-nitrobiphenyl, also other compounds, which interfere with the voltammetric determination, it is necessary to use the high-performance liquid chromatography, in which the detection is based on UV absorp-

tion measurement. The chromatograms and calibration curve for 4-nitrobiphenyl are shown in Fig. 2; the characteristic peak corresponds to 7 minutes and 30 seconds.

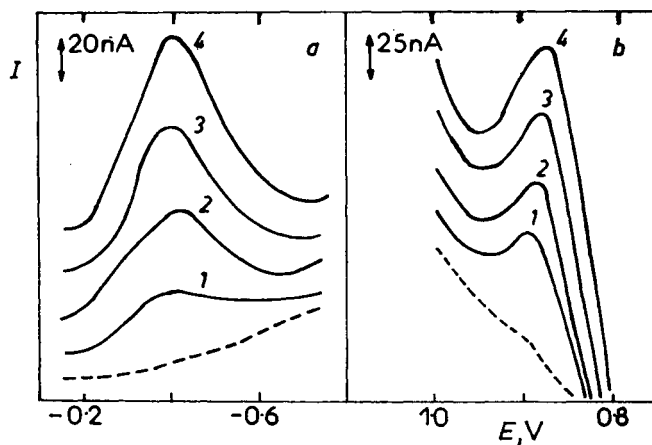


FIG. 1

Differential pulse voltammograms of 4-nitrobiphenyl (a) and 4-aminobiphenyl (b) on a stationary glassy carbon electrode. Depolarizer concentration (mol l^{-1}): $1.5 \cdot 10^{-6}$; $2.1 \cdot 10^{-5}$; $3.15 \cdot 10^{-5}$; $4.2 \cdot 10^{-5}$. Dashed line corresponds to voltammogram after destruction of 4-nitrobiphenyl, whose initial concentration was $1 \cdot 10^{-3} \text{ mol l}^{-1}$

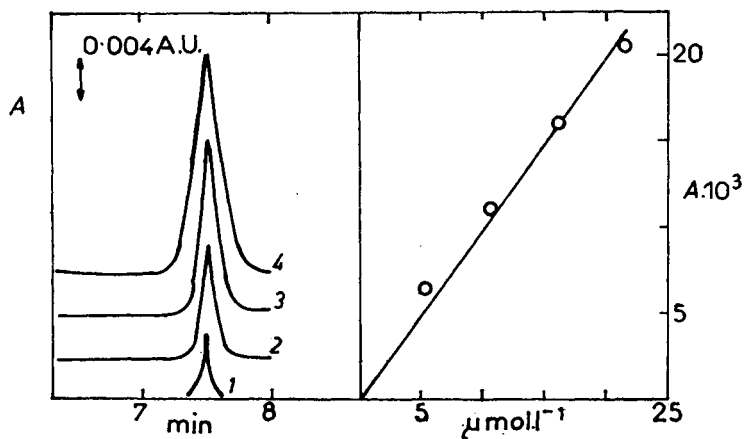


FIG. 2

Calibration curve for determination of 4-nitrobiphenyl by high-performance liquid chromatography with UV detection. Concentration of 4-nitrobiphenyl (mol l^{-1}): $1.5 \cdot 10^{-6}$; $2.1 \cdot 10^{-5}$; $3.15 \cdot 10^{-5}$; $4.2 \cdot 10^{-5}$

The peak for 4-aminobiphenyl was found at 3 minutes. After decomposition, the solution did not give any peak in the interval from 2 to 10 minutes, hence the concentration of the residual 4-nitrobiphenyl or 4-aminobiphenyl was below the detection limit of the method (about $2 \cdot 10^{-6} \text{ mol l}^{-1}$). At the initial concentration of 4-nitrobiphenyl $10^{-3} \text{ mol l}^{-1}$, at least 99.8% of it and of the 4-aminobiphenyl formed is destroyed according to these results. The analytical method described is more universal, but, on the other hand, it is more time-consuming and requires more complicated apparatus.

The method proposed can be used for destruction of both the solid compound and its solutions in solvents miscible with water and inert with respect to permanganate. The solid compound is dissolved in glacial acetic acid and mixed with water and sulphuric acid to obtain a solution of 50 vol. % acetic acid and 1 mol l^{-1} sulphuric acid, the concentration of 4-nitrobiphenyl being around $5 \cdot 10^{-3} \text{ mol l}^{-1}$. Zinc is added in a 100 fold molar excess and allowed to react for at least 12 h during stirring with a magnetic stirrer. Afterwards, permanganate is added in a roughly 50 fold molar excess and allowed to react for another 12 h. Solutions in oxidizable or with water immiscible solvents must be evaporated in a rotating vacuum evaporator. Laboratory dishes and instruments can be decontaminated by washing with acetic acid and proceeding further as above. To liquidate mixtures containing other substances reacting with Zn or permanganate, the quantity of reagents added should be increased so that they remain in excess even after 12 h of reaction. It is advisable in any case to check the efficiency of destruction by a suitable analytical method.

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