ANALYSIS OF NICOTINE IN ANTISMOKING PHARMACEUTICAL PRODUCTS BY DIFFERENTIAL PULSE POLAROGRAPHY AND VOLTAMMETRY

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A fast and simple differential pulse polarographic method was developed for analysis of nicotine in various pharmaceutical formulations (chewing gum, tablets (drops) and patches). This method requires a simple liquid–liquid extraction procedure for chewing gum and patches, or a direct dilution in supporting electrolyte for tablets before polarographic analysis. The polarographic analysis was done in a Britton–Robinson buffer (pH 6.2) as supporting electrolyte. The multimode electrode from Metrohm was used as working electrode (dropping mercury electrode). This method was applied to the determination of the nicotine content in chewing gum, tablets and patches by using the standard addition method. The results are in good agreement with the content declared by the manufacturer. The method is fast, simple and reliable, and it is a complementary method to the chromatographic method being used today for quantitative analysis of nicotine in pharmaceutical formulations. The limit of quantification is assumed to be far below 0.1 mg/l in the polarographic vessel. The method uses simple dilution and/or extraction procedures for sample preparation before polarographic analysis. It is also shown that it is possible to use a glassy carbon electrode with a mercury film (MTFE electrode) for the determination of nicotine in antismoking pharmaceutical products.

Keywords: Nicotine; Chewing gum; Patches; Tablets; Differential pulse polarography; Mercury film electrode; Electroanalysis; Electrochemistry.

Nicotine (1-methyl-2-(3-pyridyl)pyrrolidine) is a drug obtained from the plant *Nicotiana tabacum*. It is a colourless to pale yellow oily liquid with a tobacco-like odour. It is a highly addictive alkaloid found in all parts of the tobacco plant[1]. Nicotine chewing gum, nicotine patches and nicotine tablets have become popular products among people who want to stop smoking[1,2]. Nicotine replacement therapy, administered by chewing gum,

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patches or nicotine tablets, can help motivated smokers to abstain from tobacco use.

Quantitative determination of nicotine in antismoking pharmaceutical products is mainly done by chromatographic methods such as HPLC [1–3] and gas chromatography[4]. There are no voltammetric methods for determination of nicotine in such products. But there is reported a polarographic method and another electrochemical method for determination of nicotine in tobacco and tobacco smoke[5–8]. There is also one study dealing with the determination of nicotine in antismoking pharmaceutical products using an inhibition biosensor[9], and a voltammetric study of nicotine oxidation at boron-doped diamond electrodes[10].

In the present study, a differential pulse polarographic (DPP) method has been developed for determination of nicotine in chewing gum, tablets (drops) and patches. Our main objective was to develop a method that could be a complementary method to the chromatographic methods commonly used for this type of analyses.

In addition, we have also used a glassy carbon electrode with a mercury film (MTFE electrode) for the determination of nicotine in antismoking pharmaceutical products.

EXPERIMENTAL

Instrumentation

Polarographic measurements were carried out using a 797 VA Computrace instrument from Metrohm (Metrohm Ltd, CH-9101 Herisau, Switzerland). The instrument was connected to a personal computer (IBM Thinkpad), in which a software package from Metrohm called Metrodata was installed. All measurements and calculations were done using this software. The multimode electrode from Metrohm (Metrohm Ltd, CH-9101 Herisau, Switzerland) was used as working electrode (dropping mercury electrode). The result was obtained using the differential pulse mode with a pulse amplitude of 25 mV. Potentials were measured versus a silver|silver chloride|potassium chloride (3 mol/l) reference electrode, using a three-electrode system. The third electrode was a platinum wire. The drop time was 0.5 s and the drop size was 4 (0.30 mm^2). The scan rate was 2 mV/s. All samples were degassed with nitrogen before the polarographic analysis and the whole analysis was done at room temperature (20 ± 1 °C).

For the voltammetric measurement, a glassy carbon electrode (Metrohm Ltd, CH-9101 Herisau, Switzerland) was used. A mercury film was formed on the glassy carbon electrode. The surface of glassy carbon was prepared by polishing with alumina slurry using a polishing kit from Metrohm (Metrohm Ltd, CH-9101 Herisau, Switzerland). The mercury deposition was achieved by holding the electrode for 60 s at –1.5 V in a 0.001 M mercuric nitrate in 0.1 M HCl. The result was obtained using the differential pulse mode with a pulse amplitude of 25 mV. Potentials were measured versus a silver|silver chloride|potassium chloride (3 mol/l) reference electrode, using a three-electrode system. The third electrode was a glassy

carbon electrode rod (Metrohm Ltd, CH-9101 Herisau, Switzerland). The scan rate was 2 mV/s. All the samples were degassed with nitrogen before voltammetric analysis. The analyses were performed at 20 ± 1 °C.

Solid electrodes without mercury film were also tested with the same procedure; however, no voltammetric peak was observed for these experiments. A static mercury drop electrode was also tested using the same experimental conditions as for the dropping mercury electrode.

Materials

Nicotine (Lot 093K4121) was obtained from Sigma (Sigma–Aldrich CO. St Louis, MO, U.S.A.), n-hexane (95%, HPLC grade) from Lab-Scan (Lab-Scan Ltd, Dublin, Ireland), and acetic acid (96%), phosphoric acid (85%), boric acid and sodium hydroxide, all of analytical grade, from Merck (Darmstadt, Germany). High-purity water was supplied with a Milli-Q purification system (Millipore, Bedford, MA, U.S.A.). Mercuric nitrate was supplied by Sigma–Aldrich (Schnelldorf, Germany). The nitrogen used was 99.999% pure (AGA, Oslo, Norway), whilst the mercury was 99.9999% pure (Merck suprapur, Darmstadt, Germany).

Britton–Robinson buffer was made by dissolving 46 g of phosphoric acid (85%), 25 g of acetic acid (96%) and 25 g of boric acid in 500 ml of sodium hydroxide solution (2 mol/l) in a 1000-ml volumetric flask and made up to 1000 ml with water. pH of the buffer was 6.2.

Standard nicotine solution was made by dissolving 0.1110 g in 100 ml of water and stored in a dark container. This stock solution was diluted before use as standard addition solution. Polarograms were recorded from –1.0 V and a calibration curve was drawn. The unknown concentration of the samples was calculated by standard addition.

Nicotine gum containing 2 and 4 mg of nicotine, nicotine tablets (drops) containing 2 mg of nicotine and nicotine patches containing 16.6 mg of nicotine (all Nicorette, Pfizer Oslo, Norway) were supplied by the local pharmacy. Nicotine tablets (drops) release nicotine, which is absorbed through the mouth tissue.

Sample Preparation

Nicotine gum was cut into 8 pieces and placed in a 250-ml separatory funnel. The funnel was charged with 50 ml of n-hexane and 45 ml of Britton–Robinson buffer and shaken until the gum was dissolved. The supernatant was decanted into a 100-ml volumetric flask and additional 45 ml of Britton–Robinson buffer was used to rinse the gum residues. The rinse was added to the volumetric flask and the volume was adjusted to 100 ml with Britton–Robinson buffer and diluted 1:10 with the same buffer. The sample (10 ml) was filtered through an 0.45-μm nylon filter attached to a plastic syringe (both Millipore, Bedford, MA, U.S.A.) and added to the polarographic cell. The polarogram was recorded and to the polarographic cell was added 100 μl of 111 mg/l standard nicotine solution twice (standard addition).

To a nicotine tablet (2 mg/unit) in a 100-ml volumetric flask, Britton–Robinson buffer was added to the mark, shaken until it dissolved and diluted 1:10 with the same buffer. The sample (10 ml) was filtered through a 0.45-μm nylon filter attached to a plastic syringe and added to the polarographic cell, the polarogram was recorded.

Nicotine patches (16.6 mg/unit) without the protecting liners was cut into 8 small pieces and placed in a 250-ml separatory funnel, extracted and analysed as described for nicotine gum above.

RESULTS AND DISCUSSION

Figure 1 shows a differential pulse polarograms of a gum extract containing 2.06 mg/l of nicotine using double standard addition. Figure 2 shows a voltammograms of a tablet extract containing 40.96 mg/l of nicotine using an MTFE electrode using double standard addition.

The mechanism of the reduction and analysis of nicotine at the Hg electrode are described by Thomas et al.[5]. The instrumental parameters and the supporting electrolyte used in this study were also taken from this literature. Nitrogen heterocyclic compounds, such as nicotine, are reduced in a two-electron step.

The sample preparation procedure used in this study for analysis of gum and patches is a modification of the method described by Tambwekar et al.[2] for HPLC analysis of nicotine in pharmaceutical products. In this study, we used Britton–Robinson buffer (used also as supporting electrolyte) instead of phosphate buffer in the extraction with hexane. Calibration plots were drawn and were found to be linear over a broad concentration range from 0.1 to 20 mg/l of nicotine in the polarographic vessel with a correlation coefficient of 0.9997. The limit of quantification is assumed to be far below 0.1 mg/l.

The results obtained in the analysis of different antismoking pharmaceutical products are shown in Table I. This result is in a good agreement with the manufacturer's declaration (nicotine claimed) and the % RSD values are good for all the products.

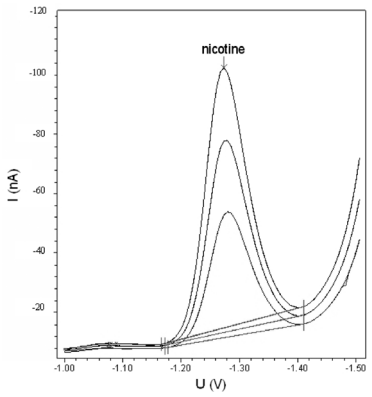

FIG. 1
Differential pulse polarogram of nicotine gum extract (10 ml) with double standard addition (100 µl of standard nicotine solution, 111 mg/l). The sample concentration was 2.06 mg/l

In Table II, the results of analysis of spiked samples of gum, tablets and patches are shown. All the samples were spiked with nicotine by adding 2 ml of standard nicotine solution (1.11 mg/ml) to the separatory funnels containing gum and to the volumetric flasks containing tablet. The same results were obtained by adding 6 ml of standard nicotine solution (1.11 mg/ml) to the separatory funnels containing patches before the extraction. The method shows good recovery after the standard addition procedure.

TABLE I
Nicotine content in antismoking pharmaceutical products using differential pulse polarography (dropping mercury electrode)

Product	Nicotine claimed mg/unit	Nicotine found mg	Recovery %	% RSD $n = 6$
Nicotine gum	2	2.03	101.5	2.56
Nicotine gum	4	3.91	97.8	2.77
Nicotine tablet	2	2.06	103.0	2.57
Nicotine patch	16.6	17.51	105.5	4.39

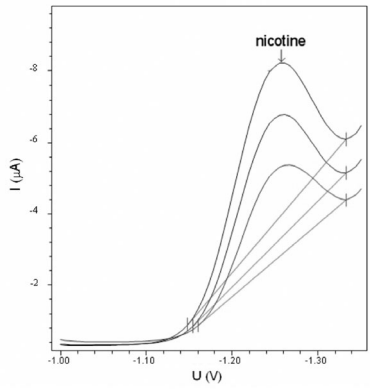

FIG. 2
Differential pulse voltammograms (MTFE electrode) of nicotine tablet extract (10 ml) with double standard addition (200 μl of standard nicotine solution, 1065 mg/l). The sample concentration was 40.96 mg/l. The supporting electrolyte was Britton–Robinson buffer of pH 4.5. The sample was obtained by adding 1 nicotine tablet to 50 ml of supporting electrolyte and shaken until it dissolved, then 10 ml was filtered through a 0.45-μm nylon filter

TABLE II
Nicotine content in antismoking pharmaceutical products after nicotine addition using differential pulse polarography (dropping mercury electrode)

Product	Nicotine claimed mg/unit	Nicotine added mg/unit	Nicotine found mg	Recovery %	% RSD $n = 5$
Nicotine gum	2	2.22	4.19	99.3	3.51
Nicotine gum	4	2.22	6.24	100.3	2.74
Nicotine tablet	2	2.22	4.16	98.6	3.02
Nicotine patch	16.6	6.66	23.65	101.7	1.89

This study also shows that it is possible to use MTFE electrodes for the determination of nicotine in antismoking pharmaceutical products. MTFE electrodes have a lower hydrogen overpotential and narrower voltage range than a pure mercury electrode[5]. That will give the MTFE electrode lower sensitivity than the dropping mercury electrode.

The analysis clearly shows that a dropping mercury electrode has a higher sensitivity than both a static mercury drop electrode and an MTFE electrode. Static mercury drop electrode and the MTFE electrode have a similar sensitivity which was approximately 10 times lower than for the dropping mercury electrode.

In summary, a simple differential pulse polarographic method has been developed for the determination of nicotine in antismoking pharmaceutical products. The significant advantage of this method is that it comprises a liquid–liquid extraction step or a simple dilution and a filtration step before analysis in the polarographic vessel. The method is reliable, simple and fast. It is a method of choice for quantitative determination of nicotine in gum, patches and tablets complementary to the chromatographic methods commonly used. It is also shown that a glassy carbon electrode with a mercury film can also be used for the determination of nicotine in antismoking pharmaceutical products.

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