RESEARCH PAPER

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Differential Pulse Polarographic Determination of Alprazolam

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Electrochemical behaviour and analysis of alprazolam has been studied by differential pulse polarography over pH range 2.0 to 12.0. Both standard addition and calibration methods are used. The optimum pH range for the analysis of the alprazolam in different pharmaceutical formulation is found to be 4-6. The lower detection limit is 1.25×10^{-7}M

SOME reviews[1-3] have been published on the determination of 1,4-benzodiazepine and their metabolities in formulations and biological fluids using direct current polarography. The purpose of this work is to describe an analytical procedure for the determination of alprazolam, a 1,4-benzodiazepine in its dosage forms by using differential pulse polarography, a highly sensitive technique compared to d.c. polarography.

EXPERIMENTAL

The differential pulse polarograms were obtained with a Metrohm E-506 polarecord connected to 648 VA combistand and E-608 VA controller. A three electrode combination was employed consisting of a droping mercury electrode (area 0.0223 cm^{-2}), Ag/Ag (s) C1 electrode and a platinum electrode as an auxilary electrode. Model LI 120 Elico digital pH meter was used for pH measurements. All the experiments were performed at 28+1 C.

Reagents and Solutions: Universal buffers of pH 2.0 to 12.0 were used as supporting electrolytes for the present investigation and they were prepared by using 0.2M boric acid 0.05M citric acid and 0.1M tri sodium orthophosphate[4]. All the chemicals were of analar grade. A stock solution (1.5×10^{-5} M) of alprazolam was prepared in dimethylformamide (DMF).

Reference Standard: Pure alprazolam was obtained from Torrent laboratories, Ahmedabad and was used without further purification.

Procedure: A 0.5 ml volume of the stock solution of alprazolam was placed in the polarographic cell and 9.5 ml of the appropriate buffer solution of selected pH was added and the solution was purged with oxygen free nitrogen for 15 minutes prior to each run. The differential pulse polarograms of alprazolam were obtained in each of the buffers of the entire pH range 2.0 to 12.0.

Preparation of Calibration graphs: A stock solution of alprazolam (10^{-5} M) was prepared in DMF and solutions containing various concentrations of alprazolam were obtained by dilution of the stock solution with an appropriate buffer to give the selected pH. The polarograms of the final sample solutions were obtained after deaeration for 15 minutes. A measured diffusion current was plotted against concentration and the detection limit is evaluated using the following equation[5]

$$dl = \frac{3\,sd}{m}$$

where dl = detection limit
sd = standard deviation
m = slope of the calibration plot

Table 1: Assay of Alprazolam dosage form by differential pulse polarography in pH 4.0 Pulse amplitude : 50 mV, Drop time : 2 sec.

Sample	Labelled amount mg	Amount found mg	Recovery %	Standard deviation
Alprax	25	24.89	98.89	0.022
Alzolam	25	24.85	98.74	0.025
Alzopax	10	9.98	99.92	0.016
Trika	10	9.97	99.82	0.018

Recommended analytical procedure: Stock solution ($1.0x10^{-5}$M) is prepared by dissolving the appropriate amount of the alprazolam in DMF. 1.0 ml of unknown solution is transferred into a polarographic cell and made up with 9 ml of the supporting electrolyte and then deoxygenated with pure nitrogen gas for 10 minutes. After recording the polarograms, small increments (0.2ml) of standard solution are added and the polarograms are recorded after each addition under similar conditions. The unknown concentrations of the substance was calculated using the equation.[6]

$$C\mu = \frac{C_s \times V}{V_t \times i_2} \times i_1$$

Where i_1 = the observed maximum current of the differential pulse polarogram (in μA) of the unknown solution (V), i_2 = the maximum current of differential pulse polarogram after adding a volume of unknown concentration, $C\mu$ = concentration of unknown solution, C_s = concentration of standard addition, V_t = total volume of the solution (V+v).

RESULTS AND DISCUSSION

ANALYSIS: Alprazolam is found to give a single well defined peak throughout the pH range 2.0 to 12.0. The electrode processes are found to be diffusion controlled without any absorption complications as shown from the linear plots of i vs.$t^{2/3}$ which are observed to be passing through the origin.

The polarographic waves obtained in pH range 4-6 are well resolved and are used for the analysis employing both calibration and standard addition methods. Using the calibration method, the peak height is found to be linear over the concentration range $1.0x10^{-5}$ M-$1.5x10^{-7}$ M for alprazolam and the lower detection limit obtained is $1.25x10^{-7}$M.

The relative standard deviation and correlation coefficient values (for 10 replicants) are found to be 1.4% and 0.997 for alprazolam. The optimum conditions for the analytical determination of alprazolam are found to be a drop time of 2 sec. and a pulse amplitude of 50mV.

Analysis of Pharmaceutical dosage forms:

The above described procedure is successfully utilised for the determination of alprazolam in various pharmaceutical formulations (Alprax, Alzolam, Alzopax and Trika) without any prior separation. The assay results of the various dosage forms investigated are given in **Table I.** The recoveries, which are in the range 98.74% to 99.92% indicate its accuracy and reproducibility.

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