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Letter to the Editor

Electrochemical determination of 2,6-diisopropylphenol after high-performance liquid chromatography of extracts from serum

Sir,

Propofol (2,6-diisopropylphenol) is a rapid-acting intravenous anesthetic. Its determination in blood samples from patients is of interest with regard to pharmacokinetic studies of the drug. Present methods include fluorimetric assays [1,2], ultraviolet (UV) detection following derivatization with Gibbs reagent [3], and a direct UV method [4]. In this paper, we outline an electrochemical method for the determination of propofol in serum which is rapid, accurate and sensitive.

Blood samples (2–3 ml) were allowed to clot and then centrifuged. To an aliquot of the resulting serum was added chlorprothixene (0.2–0.4 μg per ml of serum) as an internal standard. To the serum sample (0.5 ml) containing internal standard was added ethyl acetate (0.5 ml), followed by careful extraction on a slow rocking apparatus for 30 min. The mixture was centrifuged at 2500 *g* for 5 min, and an aliquot (20 μl) of the upper ethyl acetate phase was removed for direct injection into the chromatographic apparatus. Analysis was performed on a Brownlee C_8 column (100 mm$\times$3.2 mm I.D., 3 μm particle size), using an eluent containing 0.082 *M* sodium acetate adjusted to pH 4.0 with orthophosphoric acid–acetonitrile (45:55, v/v). The detector consisted of an ESA Coulochem Model 5100 A controller with a ESA 5010 analytical cell. Samples were pumped through the two porous graphite in-line electrodes using a Spectra-Physics Isochrom LC pump. Electrode 1 was set at +0.20 V to ensure a smooth baseline, and cell 2 was run at +0.65 V, the optimum voltage for oxidation of both propofol and the internal standard. Ethyl acetate was used in preference to cyclohexane [1–4] for extraction of propofol from serum because it removes far less lipid material and gives clearer extracts. Recovery of both propofol and chorprothixene via ethyl acetate extraction was 98–99%, and internal standards were always added to serum samples. Serum concen-

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trations of propofol were between 100 and 2000 ng/ml in pharmacokinetic studies.

The hydrodynamic voltammogram for the electrochemical oxidation of propofol in 0.082 *M* sodium acetate–phosphate (pH 4.0) (45%, v/v) and acetonitrile (55%, v/v) at room temperature (18–22°C) is shown in Fig. 1a, which indicates an optimum voltage of +0.65 V. Chlorprothixene was also found to have an optimum potential of +0.65 V. A linear standard curve was obtained for propofol extracted with ethyl acetate from drug-free serum samples previously loaded with the drug, with a reproducible lower limit of detection equivalent to 80 ng of propofol per ml of serum. This value compares favourably with previous analyses of propofol in blood following coupling with Gibbs reagent (200 ng ml^{-1}) or direct UV detection (100 ng ml^{-1}) [4], but is higher than that of a fluorimetric method (2 ng ml^{-1}) [1].

The separation of propofol and the internal standard and their electrochemical detection is shown in Fig. 1b. Both compounds elute within 10 min. A gradual decrease in response of the graphite electrodes during the electrochem-

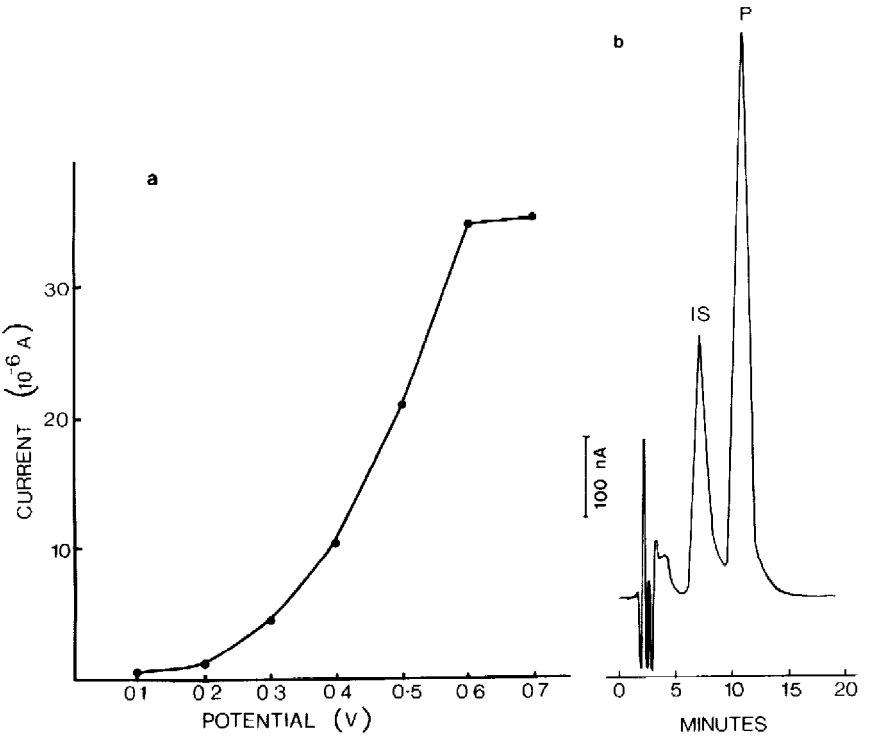

Fig. 1. (a) Hydrodynamic voltammogram for the electrochemical oxidation of propofol. Details of buffers and the cell-electrode system are given in the text. (b) Separation of propofol (P) and the internal standard (IS). Conditions for chromatography are given in the text.

ical oxidation of propofol samples was corrected for by reference to the internal standard.

The electrochemical method described in this report offers an alternative method to those in use [1–4] for the determination of propofol in blood. It is quick and compares satisfactorily with the methods in use [3,4].

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