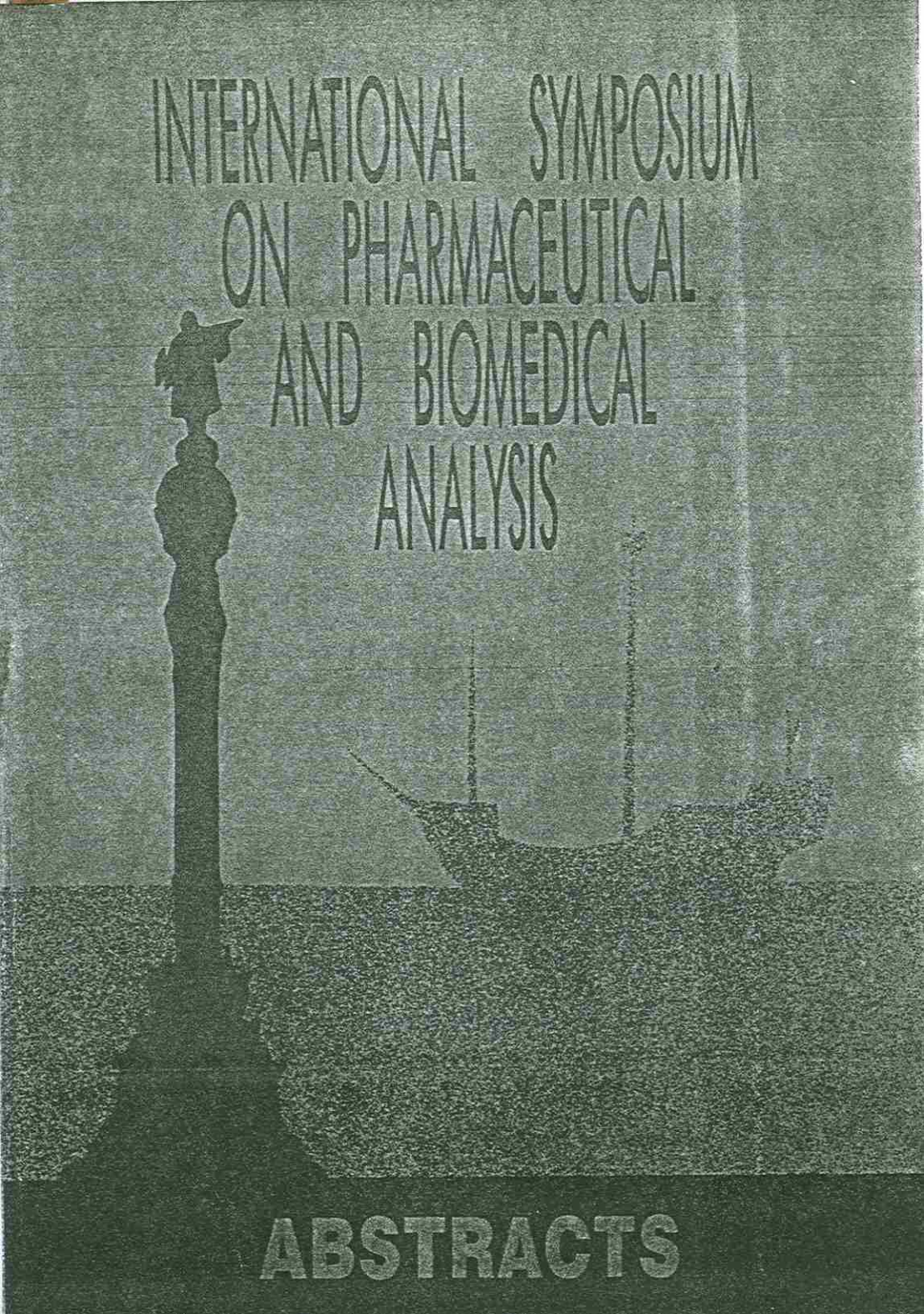
INTERNATIONAL SYMPOSIUM ON PHARMACEUTICAL AND BIOMEDICAL ANALYSIS



ABSTRACTS

ADSORPTIVE STRIPPING VOLTAMMETRIC ASSAY OF FOLIC ACID IN HUMAN SERUM.

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Folic acid (pteroylglutamic acid), a part of the vitamin B complex , is administered in the profilaxis and treatment of megaloblastic anaemia and other diseases. Present communication describes an electrochemical assay useful for monitoring the folic acid concentration in serum samples in the presence of reduced folates.

Experimental conditions affecting the adsorptive stripping behaviour of the folic acid have been examined in a previous paper (1). Alternating current (AC) voltammetry yields the most sensitive stripping signal, allowing 10^{-11} M folic acid to be determined. Suitable conditions for adsorptive preconcentration of the molecule in the presence of a large excess of surface active materials have also been established.

Folic acid can be determined in human serum by AC adsorptive stripping voltammetry (AC-AdSV) after removing of proteins . Sample clean-up was accomplished by reverse-phase extraction using C_{18} cartridges (Sep-Pak, Waters).

1 mL of serum diluted with 10 mL of pH 5.0 acetate buffer were extracted, eluting the retained materials with 2.0 mL of methanol. The dry extract was redissolved in 10 mL of background electrolyte (pH 5.0 acetate buffer) and, after degassing , the AdSV measurement was carried out. Instrumental setting: preconcentration for 70 s. in open circuit and quiescent solution, amplitude modulation of 20 mV, frequency of 75 Hz. and potential scan rate of 10 $mV\,s^{-1}$. Curves were scanned between -0.350 V and -0.750 V, recording the folic acid stripping peak at -0.630 V (SCE). Measurements were made in the sampled current mode. 5-methyltetrahydrofolic acid, the naturally occurring folate in serum (2), does not interfere when present in a 50-fold amount, as well as 5-formyltetrahydrofolic acid.

Serum samples spiked with folic acid at concentrations ranging from 2.00×10^{-8} M to 2.00×10^{-6} M give rise to a linear calibration plot (r=0.9997). The overall assay precision at the 1.00×10^{-7} M concentration level was found to be 9.9% (relative standard deviation, n=7) with a mean recovery of 56.7% .

(1) J.M. Fernández, A. Costa, A.J. Miranda and P. Tuñón, J. Electroanal. Chem., in press (1987)

(2) V. Herbert, A.R. Larrabee and J.M. Buchanan, J. Clin. Invest., 41 , 1134 (1962)