

used for treatment of infectious diseases caused by *Trichomonas*, *Lambliia*, *Leishmania*, etc., and also for eradication of *Helicobacter pylori*.

Aim. To study the process of ornidazole extraction from aqueous solutions by organic solvents for further development of the sample preparation procedure for blood and urine.

Materials and methods. Ornidazole was of pharmacopoeial purity. The ornidazole solutions with concentrations of 10, 20, 40 and 70 µg/mL in water, $1 \cdot 10^{-2}$ mole/L and $1 \cdot 10^{-5}$ mole/L hydrochloric acid solutions, $1 \cdot 10^{-2}$ mole/L and $1 \cdot 10^{-5}$ mole/L sodium hydroxide solutions were prepared.

The extraction procedure: 10.00 mL of ornidazole solution was placed into the separating funnel and extracted with 10.00 mL of chloroform or mixture chloroform–isopropanol (8:2). The obtained organic extracts were separated, filtered through the paper filter with 1 g of sodium sulphate anhydrous (wetted with the respective solvent) into the measuring flask with the capacity of 25.0 mL, and diluted to the volume with the same solvent. Two aliquots of the obtained solution (in 10.00 mL each) were used for quantitative determination of ornidazole by the method of UV-spectrophotometry.

All spectrophotometric measurements were carried out using a single beam UV/VIS spectrophotometer SPEKOL®1500 (Analytik Jena AG, Germany).

Results and discussion. Ornidazole extraction from aqueous solutions was carried out with organic solvents immiscible with water; the medium pH was equal to ≈ 2 , 5, 7, 9 and 12 that corresponded to the values commonly used to isolate analytes from body liquids in forensic toxicology. To create pH acid and alkali were used instead of buffer solutions to model the real conditions of sample preparation.

Conclusions. Ornidazole is extracted from aqueous solution in all types of medium (acid, weak acid, neutral, alkaline and alkaline) with effectiveness not less than 30%. The most effective extragent for ornidazole is the mixture chloroform–isopropanol (8:2) at pH ≈ 7 (92%) and pH ≈ 9 (80%).

CHROMATOGRAPHIC METHODS FOR ANALYSIS OF ANTIDEPRESSANTS AND ANXIOLYTIC DRUGS

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Introduction. Antidepressants are the type of psychotropic medicine that acts on the central nervous system and are used to treat depression. Patients with depression tend to abuse psychotropic drugs, take more than one antidepressant at the same time or at higher doses. Buspirone as an anxiolytic drug is sometimes prescribed as additive drug to treat major depression. In this case, there is an increased risk of poisoning, serotonergic syndrome or death. In order to determine which drug a person is poisoned with, it is useful to analyze the most effective ways to extract these drugs from plasma, separate and identify them.

Aim. The aim of this study is to find the best conditions for the solid phase extraction (SPE) of escitalopram (ESCI), fluoxetine (FLU), paroxetine (PAR), buspirone (BUS) from blood plasma and find the best HPTLC and HPLC conditions for separation and identification of ESCI, FLU, PAR, BUS.

Materials and methods. The SPE method (1 cm³ 30 mg Oasis HLB reversed phase extraction columns, Waters, USA) was used to isolate the analytes from blood plasma. Standard solutions were made by diluting ESCI, FLU, PAR, BUS separately in water to 0.1mg/ml concentrations each. Water-drugs working solution was made by mixing all standard solutions to equal parts. Plasma solution was made by mixing plasma and each standard solution to equal parts. Extraction from blood plasma was performed on SPE. Conditioning step included 1 ml methanol and then 1 ml water, loading step included 1 ml plasma-drug solution, washing step included 1ml 5% methanol solution, eluting step was performed with methanol, propanol, trichlorometan, acetonitrile, acidified/alkalined ethanol, different concentrations of ethanol. Separation and identification of analytes from plasma were obtained with HPLC (Shimadzu Nexera X2 LC-30AD, Japan), using ACE C18 column (Pennsylvania, USA). The binary solvent system of the mobile phase consist of solvent A (0.1 TFA in water) and solvent B (acetonitrile). The following linear gradient elution

profile was used: 98% A/2% B–0 min, 98% A/2% B–1 min, 2% A/98% B–20 min, 2% A/98% B–23 min, 98% A/2% B–24 min and 98% A/2% B–30 min. The flow rate was 1 mL/min and injection volume was 10 μ L. The column temperature was constant 30 °C. The chromatograms were recorded at 250 nm

HPTLC separation (CAMAG) was done using silica gel plates specifically HPTLC Silica gel 60 F₂₅₄(10*10cm). We used a mixture of acetonitrile : methanol : 25% ammonia solution (85:10:5 v/v/v) as a mobile phase. Migratin distance of mobile phase was over a path 80 mm. The determination of compounds was made with UV detector with wavelength 254 nm.

Results and discussion. The conditions of method were found experimentally. Best eluents from blood plasma for buspirone was acetonitrile (extraction output 90,1%), ethanol (84%); for fluoxetine was ethanol (84,96%), propanol (80%); for escitalopram was ethanol (87,12%), propanol (77,65%); for paroxetine was methanol (88,5%), ethanol (80,17%). Therefore the most appropriate eluent for the best separation and identification of the analytes ESCI, FLU, PAR, BUS, was found to be 80% ethanol in water solution, 2% acidified with concentrated formic acid (Figure 1 B). . The retention times of BUS, ESCI, PAR, FLU are 12.14, 13.82, 14.72 respectively.

The image of HPTLC (Figure 1 A). The obtained chromatogram by main spots corresponds to the typical reference solutions spots. R_f values were determinated for all four main substances. It was 0.92, 0.60, 0.72, 0.65, for BUS, PAR ESCI, FLU respectively in reference solutions and test sample of blood plasma. These results show an effective application of the methodology for BUS, PAR, ESCI, FLU analysis.

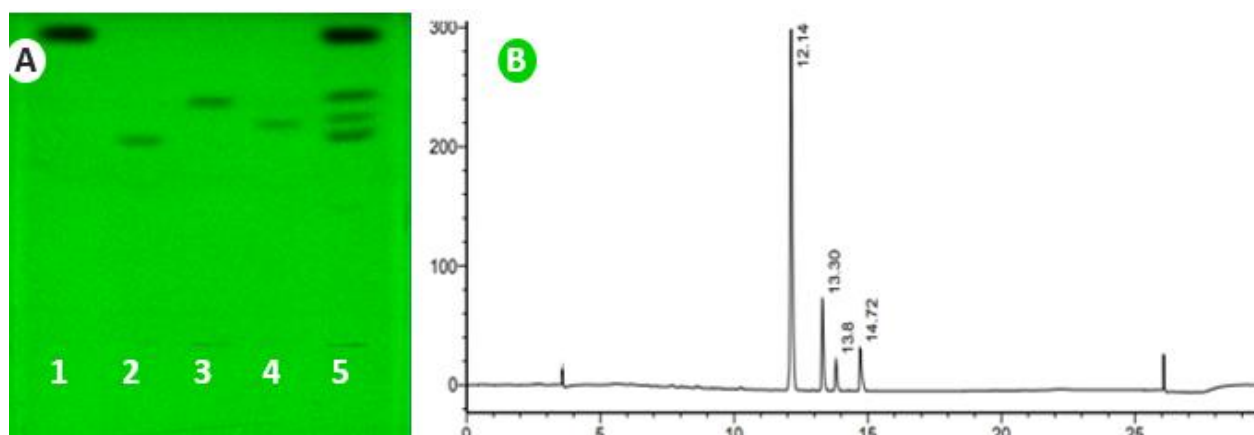


Figure 1 (A). HPTLC plate with 1-4 reference solutions of BUS, PAR, ESCI and FLU respectively and 5-test sample of blood plasma. **(B).** HPLC chromatogram of compounds, extracted from blood plasma with 80% ethanol in water solution, 2% acidified with concentrated formic acid.

Conclusion. We developed sensitive and specific methods of HPLC and HPTLC with prior solid phase extraction step for determination of SPE BUS, PAR, ESCI and FLU in blood plasma.

MODERN ANALYTICAL METHODS OF IDENTIFICATION SUBSTANCES FOR PHARMACEUTICAL APPLICATION IN LEADING WORLD PHARMACOPOEIAS

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Introduction. Methods of identification of substances for pharmaceutical use based on new advances in chemistry, physics, biology, pharmacy and other natural sciences are rapidly developing and used in a comprehensive manner. A specialist in the pharmaceutical industry, choosing a particular method of analysis, it is necessary to correctly assess their selectivity, specificity, sensitivity and informativeness.