DEVELOPMENT AND VALIDATION OF UV-SPECTROPHOTOMETRIC METHOD OF METRONIDAZOLE QUANTITATIVE DETERMINATION FOR PURPOSES OF FORENSIC AND TOXICOLOGICAL ANALYSIS

Shkarlat G. L., Zhuravel I. A., Klimenko L. Yu., Showkova Z. V., Showkova O. V. The National University of Pharmacy, Kharkiv, Ukraine toxchem@ukrfa.kharkov.ua

Metronidazole is attributed to the group of antiprotozoal medicines and widely used for treatment of infectious diseases, at the same time it is possessed of quite a number of side effects showed by classic symptoms of acute intoxication, especially when interacting with other medicines and alcohol. The concentrations of metronidazole in blood and urine are such high that allow to use UV-spectrophotometry for its quantitative determination. All mentioned above makes actual developing UV-spectrophotometric procedures of metronidazole quantitative determination with the purpose of their further application in chemical and toxicological analysis.

The purpose of this paper is to develop UV-spectrophotometric procedure of metronidazole quantitative determination and validation of the developed procedure using the offered before approaches to the determination procedure and acceptability estimation of linearity, accuracy and repeatability of UV-spectrophotometric methods of analytes quantitative determination in biological liquids applied in forensic and toxicological analysis.

The metronidazole chemical structure supposes its existence in different forms when changing medium pH. The presence of such transformations is confirmed by UV-spectra of metronidazole obtained in different solvents with different values of pH – when increasing the pH value step-by-step shift of substance maximum absorption to the right is observed (277 nm \rightarrow 310 nm \rightarrow 314 nm \rightarrow 319 nm).

The development of procedure of metronidazole quantitative determination was carried out using ethanol as a solvent and wavelength of 310 nm respectively by the following scheme: application of the normalized coordinates (normalization by the reference solution); the application ranges are 25 - 125%, 25 - 150%, 25 - 175%; the number of concentration levels is g = 5, 6 or 7 (depending on the chosen application range) in constant increments of 25%.

The metronidazole concentration in the model solution corresponding to the point of 100% in the normalized coordinates was chosen in the way that the absorbance of this solution was 0.4 - 0.6.

Validation of the developed procedure has been carried out by parameters «linearity», «accuracy» and «repeatability» and its acceptability for further application in forensic toxicology has been shown.