STUDY OF EFAVIRENZ EXTRACTION FROM AQUEOUS SOLUTIONS

Slabiak O.I., Ivanchuk I.M., Klimenko L.Yu., Mykytenko O.Ye.
Ivano-Frankivsk National Medical University, Ivano-Frankivsk, Ukraine
National University of Pharmacy, Kharkiv, Ukraine
lina_klimenko@nuph.edu.ua

Efavirenz is a non-nucleoside reverse transcriptase inhibitor and attributed to the group of antiretroviral medicines used for treatment of HIV infection.

Our work is devoted to research of the efavirenz behaviour under the conditions of a number of methods commonly used for sample preparation of biological liquids and tissues in chemical toxicological analysis.

We proposed the procedure of extraction studies – extraction by amphiphylic solvents followed by the separation of the organic layer under the conditions of aqueous phase saturation with electrolyte (2-propanol, acetonitrile and ethanol were used as amphiphylic solvents; ammonium sulphate has been applied as an electrolyte). The medium pH was created with 6 M and 0.1 M solutions of hydrochloric acid, 25% ammonium hydroxide solution or 10% sodium hydroxide solution.

10.00 mL of distilled water or 10.00 mL of distilled water acidified with 6 M hydrochloric acid solution to pH = 2, or 10.00 mL of distilled water acidified with 0.1 M hydrochloric acid solution to pH = 5, or 10.00 mL of distilled water alkalified with 25% ammonium hydroxide solution to pH = 9, or 10.00 mL of distilled water alkalified with 10% sodium hydroxide solution to pH = 12 were placed into the separation funnel, 1.00 mL of efavirenz standard solution was added and the mixture was shaken. 10.00 mL of acetonitrile, 2-propanol or 96% ethanol were added to the aqueous solution and the mixture was shaken for 15 minutes. Then the mixture was salted-out by adding ammonium sulphate till stopping its dissolution. Top organic layer was separated, filtered through the paper filter with 1 g of sodium sulphate anhydrous (wetted with the organic solvent) into the measuring flask with the capacity of 25.0 mL, and diluted to the volume with the organic solvent (organic extract).

In all cases, extraction recovery was determined for a single processing of aqueous solution with an organic solvent for 15 minutes at the concentration levels of efavirenz corresponded to the points of 25%, 50% and 100% in the normalized coordinates. The amount of extracted medicine was determined by two methods using 96% ethanol and 0.1 M sodium hydroxide solution and measuring the absorbance at λ = 247 nm and 267 nm respectively.

Processing the aqueous solutions with amphiphylic solvents followed by «salting out» with ammonium sulphate in all cases allows to extract sufficiently high amounts of efavirenz in all media (not less than 80%). For application of acetonitrile the highest value of recovery is observed in the strong acid medium (≈98%) and then gradually decreases to 80%. For application of 2-propanol and ethanol the highest values of recovery are observed in the strong acid medium (≈95%) and then decreases to 85% (at pH = 5) and the same values are fixed in the neutral and alkaline medium.

The high extraction efficiency in the strong acid medium allow to develop the procedures of sample preparation of biological objects with processing at pH < 2 that provide obtaining of the purer extracts than in the neutral and alkaline medium.