Abstract

Charles University in Prague, Faculty of Pharmacy in Hradec Králové

Department: Pharmaceutical Chemistry and Pharmaceutical Analysis

Candidate: Mgr. Iva Kopecká

Tutor: PharmDr. Pavla Pilařová, Ph.D.

Name of Degree Paper: Chromatographic evaluation of amiodaronu and its active metabolite

The main purpose of this thesis was to optimize the existing HPLC method used in University Hospital Hradec Králové for determination of amiodarone and its active metabolite in biological material. For this HPLC method was used Waters Symetry C18 column, 150 x 4,6 mm, 5 µm, mobile phase: acetonitrile: 25 mM phosphate buffer (55:45), flow rate 1,4 ml/min, at 45 ° C, injection 5 µl, UV detection at 242 nm. This method was subsequently transferred to a UHPLC using Phenomenex Kinetex C18 column, 100 x 2,1 mm, 1,7 µm, mobile phase: acetonitrile: 25 mM phosphate buffer (55:45), flow rate 0,3 ml/min, at 45 ° C, injection 5 µl, UV detection at 242 nm. This UHPLC method was modified for eventual detection by mass detector using same UHPLC column, mobile phase: acetonitrile: 0,1% formic acid (55:45), flow rate 0,3 ml/min, at 45 ° C, injection 5 µl, UV detection at 242 nm.

At the same time plasma, resp. serum sample isolation was optimized. The best result was achieved by adding 4 µl of 10% zinc sulfate to the plasma sample followed by precipitation by adding 200 µl of acetonitrile with internal standard dronedarone, followed by shaking and centrifugation.

The found methods meet validation parameters (linearity, selectivity, stability, limit of detection and quantitation, accuracy, precision, robustness). The methods were applied to real sample of patients and compared with the values measured in the University Hospital.