Abstract

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Title of master‘s thesis: Analytical evaluation of drugs using HPLC IV.

This thesis describes development of the suitable method for ketoprofen analysis by high performance liquid chromatography. For this aim a LiChro CART® 125-4 Lichrospher® 100 RP-18 (5 μm) column was used as a stationary phase and mobile phase contained mixture of methanol and aqueous KH_2PO_4 buffer (pH 3,4) in a ratio of 60:40 (v/v). The flow rate was set to 1 ml/min, temperature on the column was 25 °C and substances were detected at a wave length of 254 nm. Under these conditions final retention time of ketoprofen was 7,9 minutes. Benetazon was chosen as an appropriate internal standard for the quantitative evaluation. In the next part we were looking for suitable conditions for the chiral separation of the analyzed drug by using normal and reverse mode. Separation of ketoprofen’s enantiomers was carried out on a Chiralcel OD-R, 250 x 4,6 mm, from Daicel Chemical Industries using a mobile phase of heptane and 2-propanol in a ratio of 97:3, with the addition of 0,6 ml formic acid. At a flow rate of 0,7 ml/min, retention times took about 20 minutes. Finally, extraction of ketoprofen from human plasma was performed, as the best method was chosen a liquid-liquid extraction with an average yield of 102,42 %. After the development of the method, these validation parameters were evaluated: precision, accuracy, linearity, selectivity, limit of detection and limit of quantification.