DEVELOPMENT AND VALIDATION OF A NEW LC METHOD USING FLUORESCENCE DETECTION FOR DETERMINATION OF OSELTAMIVIR PHOSPHATE IN CAPSULES

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Oseltamivir is used as a neuraminidase inhibitors for the treatment of influenza A and B viruses. In this study, a new sensitive and accurate reversed phase high performance liquid chromatography (RP-HPLC) method was developed and validated for the determination of oseltamivir phosphate (OSP) in capsules. The method was based on the reaction of OSP with 4-chloro-7-nitrobenzofurazan (NBD-C1) in borate buffer solution of pH 8.50 forming highly fluorescent derivatization product. The oseltamivir-NBD derivative was extracted with ethylacetate after acidification and separated by HPLC. It employed a Phenomenex C18 Column (250x4.6mm i.d, 5μm) with an isocratic mobil phase consisting of acetonitrile-10 Mm nitric acid (pH 3, 60+40, v/v) at a flow rate of 1.0 mL/min and monitored by fluorescence detection (λex: 470nm, λem: 541nm). Mexiletine hydrochloride was used as internal standard. The developed HPLC method was validated according to the ICH Guidelines with respect to specificity, linearity, accuracy, precision, and robustness. Linearity over a concentration range of 50-750 ng mL⁻¹ was shown (correlation coefficient= 0.9998). The limit of detection and quantitation of OSP were 0.59 and 6.10 ng mL⁻¹, respectively. The relative standard deviation values for intraday and interday precision were 0.25-0.96 and 0.16-1.03%, respectively. The method was successfully applied to determination of the drug in capsules. The mean % recovery (n=6) was 99.95%. Excipients present in the capsules did not interfere with the analysis. The proposed method is highly sensitive and accurate and can be used for the reliable quantitaion of OSP in capsules.