A VALIDATED RP-HPLC METHOD FOR SIMULTANEOUS ESTIMATION OF TIROFIBAN HYDROCHLORIDE IN PURE AND MARKETED FORMULATION

About Authors:
Sukanto Paul, Krishan R Bhadu
Department of Quality Assurance, School of Pharmaceutical Sciences, Jaipur National University, Jaipur, Rajasthan, India.

ABSTRACT
A validated reverse phase high performance liquid chromatography method has been developed for the simultaneous determination of Tirofiban hydrochloride in pure and marketed formulation. Chromatography was carried out on a BDS Hypersil C18 (4.6 mm × 250 mm, 5 μm) using Buffer: Acetonitrile in the ratio of 80:20 (v/v) as the mobile phase at a flow rate of 1.5 mL/min and eluents were monitored at 274 nm using UV detector at ambient temperature. The average retention time of Tirofiban was found to be 9.124 min. The method was validated for linearity, precision, accuracy, specificity, robustness and solution stability. The calibration curve was linear (R²=0.9999) over the range of 12.5-75 μg/mL. Limit of detection (LOD) and Limit of quantitation (LOQ) were 0.11 μg/mL and 0.33 μg/mL respectively. This method can be successfully employed for the quantitative analysis of Tirofiban hydrochloride in bulk drugs and formulations.
INTRODUCTION

Tirofiban hydrochloride (1(S)-2-(butylsulfonamino)-3-(4-[4-(piperidin-4-yl)butoxy]phenyl)propanoic acid, fig. 1), is a white free flowing powder, freely soluble in alcohol and slightly soluble in water. It is a specific non peptide platelet fibrinogen receptor (GPIIb/IIIa)-antagonist and this compound is an antithrombotic and used in treatment of unstable angina[7].

Literature survey indicated that estimation of Tirofiban was done by using LC-MS in biological fluid [8]. It also includes quantitative determination of Tirofiban hydrochloride by UV spectrophotometry. [9] The literature survey does not reveal any validated assay method for the estimation of Tirofiban hydrochloride in bulk and Pharmaceutical dosage form by RP-HPLC method.

The objective of this work was to develop simple and rapid RP-HPLC method which would be accurate and precise.

The methods were validated according to ICH guidelines. The linearity of response, accuracy, and intermediate precision of the described methods has been validated.[1,2,3]

2. MATERIALS AND METHODS:

Tirofiban Hydrochloride of working standard grade was kindly supplied as gift sample by Venus Remedies Ltd., Panchkula, India and was certified to contain 83.93% (w/w), on as is basis. Tirofiban Hydrochloride Active Pharmaceutical Ingredient (API) was kindly supplied as gift sample by Taj Pharmaceuticals, Valsad, India and was certified to contain 99.6% (w/w), on as is basis. Acetonitrile and water used were of HPLC grade and were purchased from Spectrochem Pvt. Ltd. Mumbai, India. The parenteral formulation (Aggrastat injection) containing 5 mg of Tirofiban Hydrochloride was procured from local market and used for analysis of marketed formulation. After several trials taken by changing mobile phase, columns, flow rate, column temperature, the following chromatographic conditions were finalised.

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