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ANALYTICAL METHOD DEVELOPMENT AND VALIDATION OF FELODIPINE IN BULK AND TABLET DOSAGE FORM BY USING RP-HPLC TECHNIQUES

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Abstract:

This paper describes the analytical method suitable for validation of Felodipine by reversed Phase High Performance Liquid Chromatography (RP-HPLC) method. The method utilized RP-HPLC (Water 2695 with PDA detector) model and a column ODS C18 (4.6 x 150mm, 5µm). The mobile phases were comprised with Acetonitrile and Water (80:20 V/V) at a flow rate of 1.0 ml/min. UV detection at 305 nm MTS were eluted with retention times of 3.155min. The method was continued and validated accordance with ICH guidelines. Validation revealed the method is rapid, specific, accurate, precise, reliable, and reproducible. Calibration curve plots were linear over the concentration ranges 15-75 µg/mL (R^2 = 0.9998). Limit of detection (LOD) was 0.19µg/ml and limit of quantification (LOQ) was 0.6µg/mL. The method showed good recoveries (98.9 - 100.4%). Statistical analysis was proves the method is suitable for the analysis of Felodipine as a bulk, in tablet dosage form without any interference from the excipients. It was also proved study for degradation kinetics. It may be extended for its estimation in plasma and other biological fluids.

Keywords: *Felodipine, RP-HPLC, Method Development and Validation.*

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INTRODUCTION:

Felodipine is chemically 3-Ethyl 5-methyl 4-(2,3-dichlorophenyl)-2,6-dimethyl-1,4-dihydro-3,5-pyridinedicarboxylate [1]. Felodipine is used to treat high blood pressure and stable angina [2, 3]. CYP3A4 inhibitors, which increase the amount of felodipine available per dose, include cimetidine, erythromycin, itraconazole, ketoconazole, HIV protease inhibitors, and grapefruit juice [2, 4].

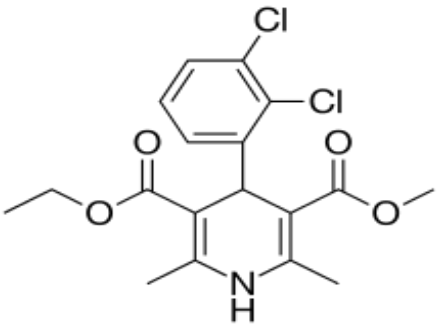

Fig. 1: Chemical structures of Felodipine

In the scientific literature, analysis of Felodipine has been reported by RP-HPLC [5] and UV-Visible Spectroscopy [6, 7]. No other chromatographic methods are found for analysis of Felodipine in a tablet dosage form. The method described is rapid, economical, precise, and accurate and can be used for routine analysis of tablets. It was validated as per ICH guidelines [8-11].

MATERIALS AND METHOD

Apparatus:

The analysis was performed by using the analytical balance G285 (Mettler Toledo), the HPLC used is of Water 2695 with PDA detector. Column used in HPLC is ODS C18 (4.6 x 150mm, 5µm) with a flow rate of 1.0 ml/min. The mobile phase consists of ACN: Water (80:20% v/v) which is degassed in a sonicator for about 10 minutes the injection volume is 10µl and the ultra violet detection was at 305 nm.

Reagents and solutions:

Pure sample of Felodipine and other ingredients such as Acetonitrile and water used were of HPLC and milli-q grade. Optimized chromatographic conditions are listed in table no.1.

Preparation of standard solution:

Accurately weigh and transfer 10 mg of Felodipine working standard into a 10ml of clean dry volumetric flasks add about 7ml of ACN and sonicate to dissolve and removal of air completely and make volume up to the mark with the same ACN.

Further pipette 0.45ml of the above Felodipine stock solutions into a 10ml volumetric flask and dilute up to the mark with ACN.

Sample preparation:

Weigh and powder the 20 tablets. An accurately weigh and transfer the powder equivalent to 10 mg of Felodipine into 10 ml volumetric flask. Add about 10 ml of mobile phase and sonicate for 20 minutes with occasional swirling to dissolve. Cool it and makeup to the volume with mobile phase. Centrifuge the solution at 3000 rpm for 15 minutes.

Transfer the 0.45 ml of the above supernatant solution into 10 ml volumetric flask, dilute to the volume with mobile phase and mix well. Filter the solution through the 0.45 mm filter.

Linearity & Range:

The Linearity of detector response is established by plotting a graph to concentration versus area of Felodipine standard and determining the correlation coefficient. A series of solution of Felodipine standard solution in the concentration ranging from about 15-75 µg/ml level of the target concentration (45µg/ml of Felodipine) was prepared and injected into the HPLC system.

Accuracy:

Accuracy for the assay of Felodipine tablets is determined by applying the method in triplicate samples of mixture of placebo to which known amount of Felodipine standard is added at different levels (50%, 100%, and 150%). The sample were filtered through 0.45mm membrane filter and injected into the chromatographic system.

Precision:

The precision of the analytical method was studied by analysis of multiple sampling of homogeneous sample. The precision expressed as %RSD. The % RSD was found to be 0.74% in the results of precision.

Robustness:

The analysis was performed in different conditions to find the variability of test results. The following conditions are checked for variation of results. .

Effect of Variation of flow conditions:

The sample was analyzed at 0.9ml/min and 1.1ml/min instead of 1ml/min, remaining conditions are same. 10µl of the above sample was injected and chromatograms were recorded.

Effect of Variation of mobile phase organic composition:

The sample was analyzed by variation of mobile phase i.e. ACN: Water was taken in the ratio and 75:25, 85:15 instead of 80:20, remaining conditions are same. 10µl of the above sample was injected and chromatograms were recorded.

RESULTS AND DISCUSSION:

Felodipine standard having concentration 45 µg/ml was scanned in UV- region between 200-400 nm. λmax of Felodipine was found to be at 305 nm.

The Felodipine peak in the sample was identified by comparing with the Felodipine standard and the Retention time was found to be around 3.155 minutes.

The estimation of Felodipine tablets was carried out by RP-HPLC using Mobile phase having a composition of ACN: Water (80:20% v/v). Then finally filtered using 0.45µ nylon membrane filter and degassed in sonicator for 10 minutes. The column used was ODS C18 (4.6 x 150mm, 5µm). Flow rate of Mobile phase was 1.0 ml/min, System suitability parameters such as RSD for six replicate injections was found to be less than 2%, theoretical plates - 5918, and tailing factor - 1.33.

The acceptance criteria of System Suitability is RSD should be not more than 2.0% and the method show System Suitability 0.31% which shows that the method is repeatable.

The acceptance criteria of Method Repeatability is RSD should be not more than 2.0% and the method show Method Repeatability 0.74% which shows that the method is precise.

The validation of developed method shows that the drug stability is well within the limits. The linearity of the detector response was found to be linear from 15-75 µg/ml of target concentration for Felodipine standard with a correlation coefficient value is greater than 0.999. The correlation coefficient of (R^2) = 0.9998, the Felodipine LOD is 0.19µg/ml and LOQ is 0.6µg/ml, which shows that the method is capable of producing good response in PDA detector.

The Accuracy limit is the % recovery should be in the range of 98.9 - 100.4%. The validation of developed Method shows that the accuracy is well within the limit, which shows that the method is capable of showing good accuracy.

Table 1: Optimized chromatographic conditions

Parameters	Method
Stationary phase (column)	ODS C18 (4.6 x 150mm, 5µm)
Mobile Phase	ACN: Water (80:20% v/v)
Flow rate (ml/min)	1.0
Run time (minutes)	8.0
Column temperature (°C)	35
Volume of injection loop (µl)	10
Detection wavelength (nm)	305
Drugs RT (min)	3.155

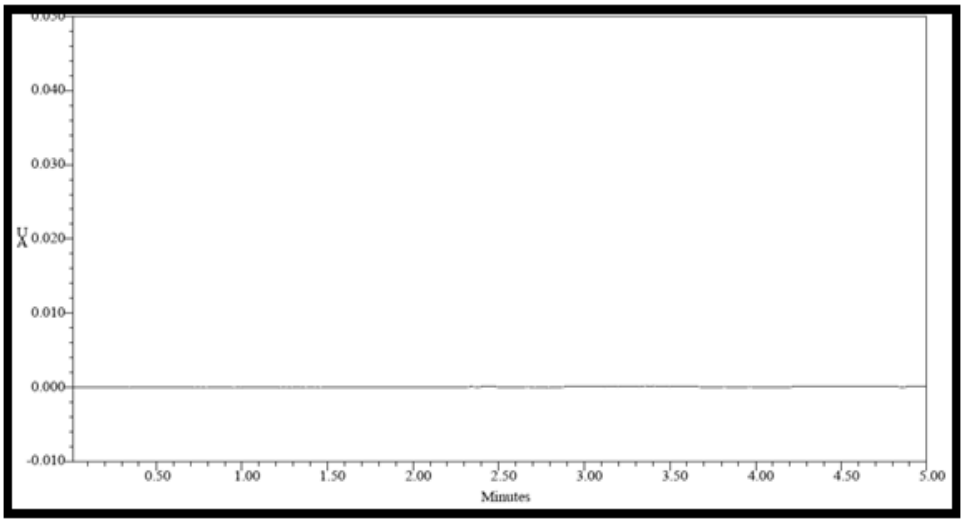

Fig. 2: Chromatogram showing blank (with Mobile Phase)

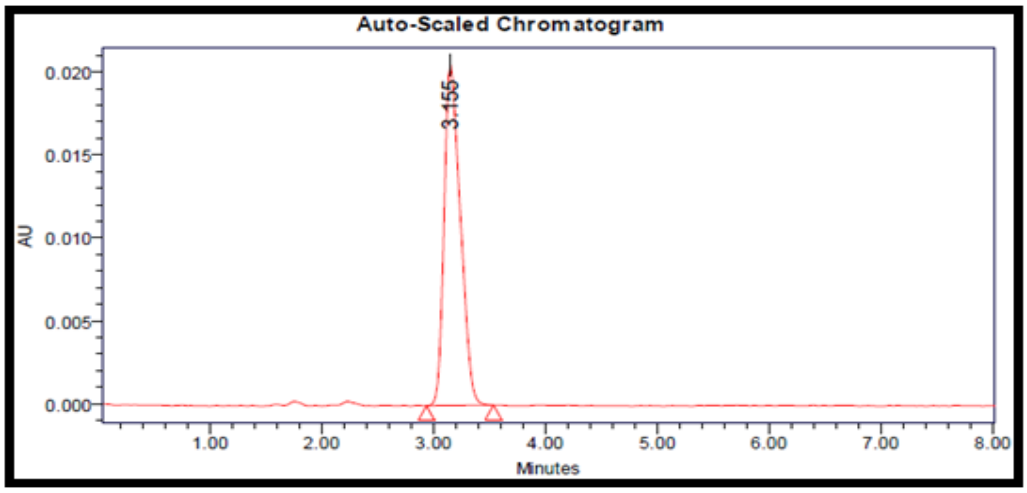

Fig. 3: Chromatogram of Felodipine at 305nm

Table 2: Data for Linearity study

Concentration µg/ml	Average Peak Area
15	38455
30	71755
45	102086
60	135415
75	164313

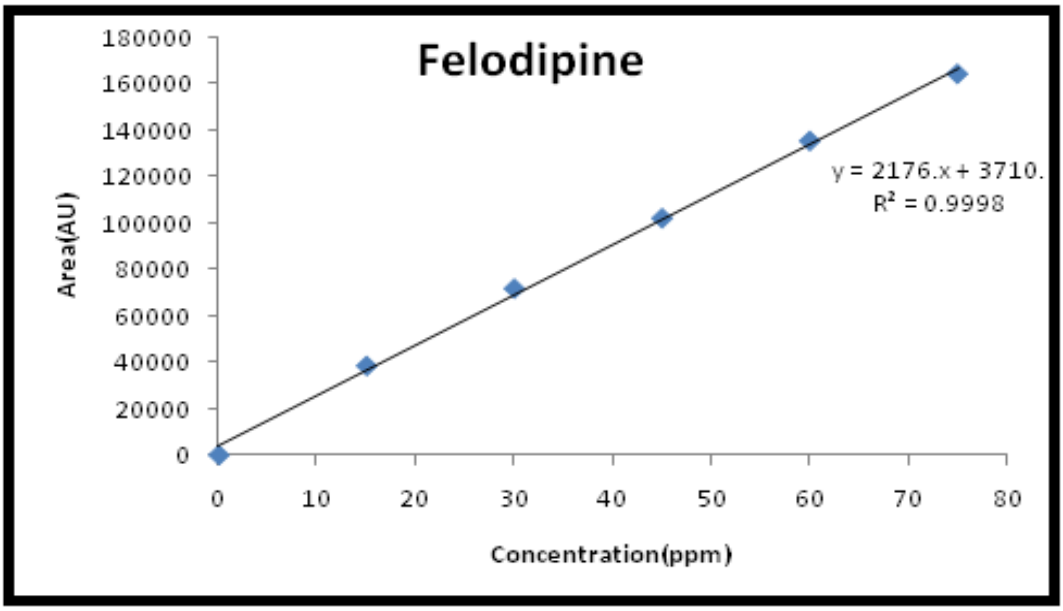

Fig. 4: Linearity Curve of Standard Felodipine

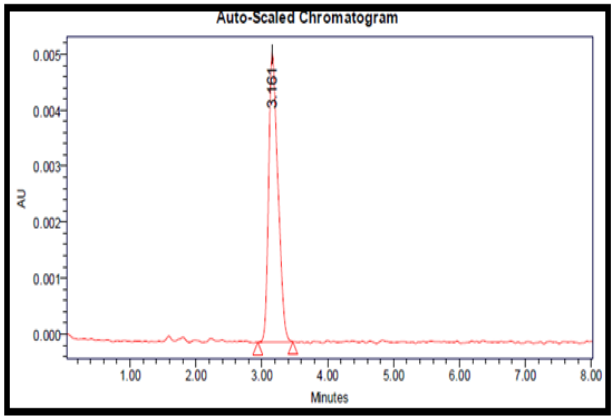

Fig. 5: Chromatogram showing linearity level-1

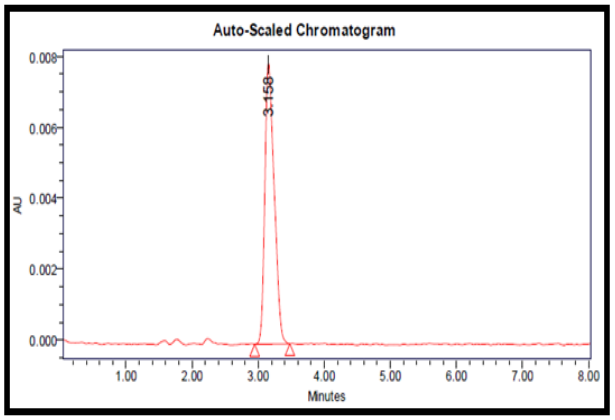

Fig. 6: Chromatogram showing linearity level-2

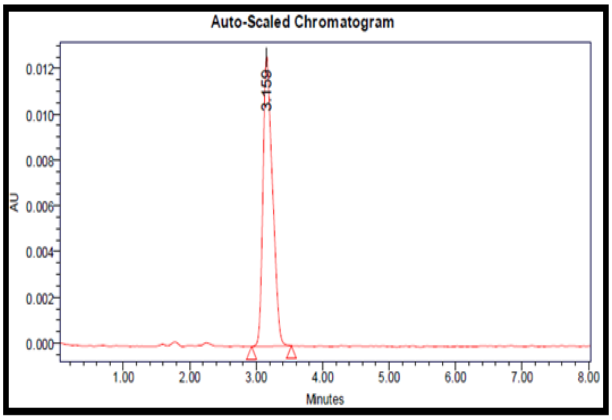

Fig. 7: Chromatogram showing linearity level-3

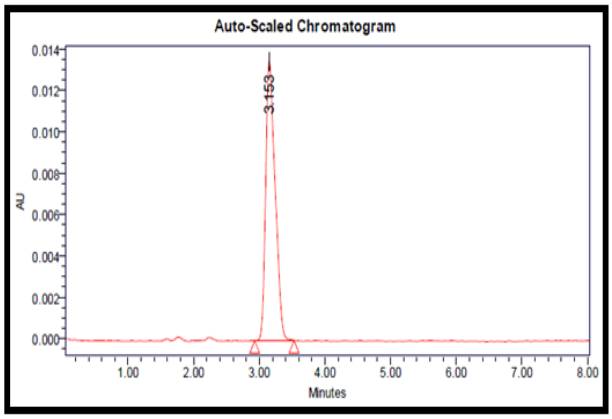

Fig. 8: Chromatogram showing linearity level-4

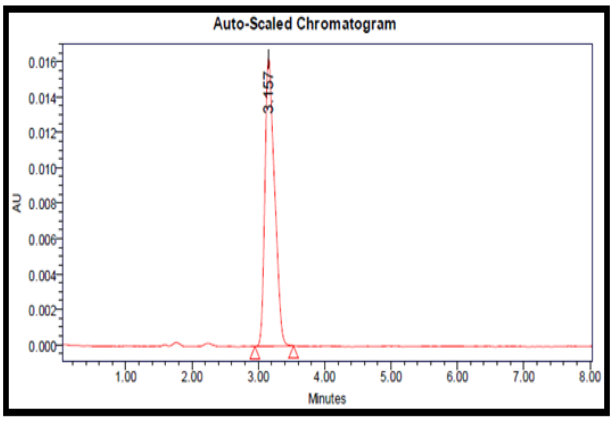

Fig. 9: Chromatogram showing linearity level-5

Table 3: Results for Robustness

Parameter used for sample analysis	Peak Area	Retention Time	Theoretical plates	Tailing factor
Actual Flow rate of 1.0 mL/min and Mobile Phase (80:20 ACN: Water)	126086	3.155	4245	1.33
Less Flow rate of 0.9 mL/min	139530	3.488	5372	1.3
More Flow rate of 1.1 mL/min	114279	2.877	3656	1.4
Less organic phase (75:25)	116384	4.705	5362	1.4
More organic phase (85:15)	113480	2.090	6251	1.2

Table 4: System suitability parameters

Parameter	Felodipine
Calibration range (µg/ml)	15-75
Theoretical plates	5918
Tailing factor	1.33
Correlation Coefficient(r2)	0.9998
% Recovery	98.9 - 100.4%
System Suitability %RSD	0.31%
Method Repeatability %RSD	0.74%
LOD (µg/ml)	0.19
LOQ (µg/ml)	0.6

CONCLUSION:

A new RP-HPLC method described in this manuscript provides a simple, convenient and reproducible approach for the estimation and quantification of Felodipine in routine quality control analysis.

REFERENCES:

1.https://www.drugbank.ca/drugs/DB01023.
2.Felopidine UK label. UK Electronic Medicines Compendium. 15 September 2015.
3.Felodipine US label. FDA. October 2012.
4.Jawad Kiani, Sardar Z Imam. Medicinal importance of grapefruit juice and its interaction with various drugs. Nutr J 2007;6(33):33.
5.B. Raja and A. Lakshmana Rao. Development and Validation of RP-HPLC Method for Simultaneous Estimation of Ramipril and Felodipine in Pharmaceutical Dosage Forms**.** Asian J Res in Chem 2013;6(11): 1018-1022.
6.Akash A. Vadangale, Pawar SS, Pande VV, Arote SR and Musmade Deepak S. Development and validation of UV spectrophotometric method for estimation of regioisomeric impurity in Felodipine bulk and formulation. Int J Pharm Chem 2015;5(04):128-133.
7.Alexandra Filareta Neagu, Ioana Clementina Constantinescu, Angela Nedelcu. The Assay of Felodipine by Second Derivative Spectrophotometry. Farmacia 2016;64(1):143-146.
8.Code Q2(R1)-Text on Validation of Analytical Procedure Step-3 Consensus Guideline. ICH Harmonised Tripartite Guideline, 2005.
9.Code Q2A-Text on Validation of Analytical Procedure Step-3 Consensus Guideline. ICH Harmonised Tripartite Guideline, 1994.
10.Code Q2B-Validation of Analytical Procedure Methodology Step-4 Consensus Guideline. ICH Harmonised Tripartite Guideline, 1994.
11.International Conference on Harmonization. Photo stability testing of new drug substance and products Q1B. International Conference on Harmonization, IFPMA, Geneva, 1996.