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Research Article

# STABILITY INDICATING RP-HPLC METHOD FOR THE DETERMINATION OF TELMISARTAN IN PURE AND PHARMACEUTICAL FORMULATION

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**ABSTRACT**

A simple, selective, precise and stability indicating RP High Performance Liquid Chromatographic (HPLC) method of analysis of Telmisartan in pure and pharmaceutical dosage form was developed and validated. The chromatographic conditions comprised of a reversed-phase $C_8$ column (4.6 x 150mm, 3.5 µm, Make: XTerra), with a mobile phase composed of Buffer and Methanol (40:60v/v, Adjusted the pH to 3.0 with ortho Phosphoric acid). Flow rate was 0.5 mL / min. Detection was carried out at 230 nm. The retention time of Telmisartan was 2.6 min. The linear regression analysis data for the calibration plots showed good linear relationship in the concentration range 20-100 µg/ml. The values of correlation coefficient, slope and intercept were, 0.9998, 2.326and-6.708, respectively. The method was successfully validated in accordance to ICH guidelines acceptance criteria for specificity, linearity, precision, recovery, ruggedness and robustness. The drug undergoes degradation under acidic, basic, Peroxide and thermal degradation conditions. All the peaks of degraded product were resolved from the active pharmaceutical ingredient with significantly different retention time. As the method could effectively separate the drug from its degradation product, it can be employed as a stability-indicating one.

**Keywords:** Telmisartan, RP-HPLC, Degradation studies.

## INTRODUCTION

Telmisartan is chemically described as 4'-[(1,4'-dimethyl-2'-propyl [2,6'-bi-1H-benzimidazol]-1'-yl)methyl]-[1,1'-biphenyl]-2-carboxylic acid. Its empirical formula is $C_{33}H_{30}N_4O_2$, its molecular weight is 514.63. The objective of this work was to develop an analytical HPLC procedure, which would serve as stability indicating assay method for Telmisartan. A thorough literature survey revealed that the reported analytical procedures describing a stability indicating HPLC method for Telmisartan were more economical.

The Objective of this study was to develop the method with less economical, precise, simple and sensitive and determination of Telmisartan in the presence of its degradation products. Here direct use of the mobile phase as diluent for formulations in quantitative analysis minimizes errors that occur during tedious extraction procedures. From the best of our knowledge via literature search, this is the first known RP-HPLC method that can separate all the related compounds of Telmisartan from each other and from Telmisartan with less economical and is therefore suitable to conduct stability studies of Telmisartan.

## MATERIAL AND METHODS

### Materials

Telmisartan was supplied by Anant Labdhi Private Limited and Product Name: Micardis (80mg). Methanol (HPLC grade) purchased from Rankem Ltd., New Delhi, India. High purity water was prepared by using Millipore Milli-Q plus water purification system.

### Instrument used

The HPLC used was WATERS HPLC with photodiode array detector and Empower software. The column used was XTerra® RP 8, 4.6 x 150mm, 3.5 µ. Thermal Stability studies were performed in a dry air oven (Thermo labs, India).

### Methodology

### Chromatographic conditions

Chromatographic separation was achieved at ambient temperature on a reversed phase column. The mobile phase consisted of Methanol-Phosphate buffer solution (60:40v/v) at a flow rate of 0.5 ml/min. Monobasic potassium phosphate solution was prepared by dissolving 7 gms KH2PO4 in 1000ml double distilled water. Final pH of the mobile phase was adjusted to 3.5 with orthophosphoric acid. The mobile phase so prepared was filtered through 0.22 µm nylon membrane filter and degassed by sonication. Detection was carried out at 230 nm. The injection volume was 20 µL for assay and degradation level.

### Standard preparation

Accurately weigh and transfer 20mg of Telmisartan Working standard into a 10 mL volumetric flask add about 7 mL of Diluent and sonicate to dissolve it completely and make volume up to the mark with the same solvent. (Stock solution).A series of standard solutions in the concentration range of 20, 40, 60, 80, 100µg/ml were prepared followed by a suitable dilution of stock solution with the mobile phase.

### Sample preparation

Weigh 20 Telmisartan Tablets and calculate the average weight. Accurately weigh and transfer the sample equivalent to 20 mg of Telmisartan into a 10 mL volumetric flask. Add about 7 mL of diluent and sonicate to dissolve it completely and make volume up to the mark with diluent. Mix well and filter through 0.45µm filter.

### Method validation

### Linearity

The linearity response was determined by preparing and injecting solutions with concentrations of about 20, 40, 60, 80, 100µg/ml of Telmisartan.

### Precision

Precision was measured in terms of repeatability of application and measurement. Repeatability of standard application was carried out using six replicates of the same standard concentration (30 µg/mL for standard application). Repeatability of sample measurement was carried out in six different sample preparations from same homogenous blend of marketed sample (30 µg / mL for sample application). It showed very low % relative standard deviation (% RSD) of peak area of Telmisartan.

### Accuracy

Accuracy (Recovery) study was performed by spiking 50, 100 and 150% of Telmisartan working standard to a preanalysed sample. The accuracy of the analytical method was established in triplicate across its range according to the assay procedure.

**Ruggedness and robustness of the method**

To evaluate the intermediate precision (also known as Ruggedness) of the method, Precision was performed on different day by using different make column of same dimensions. Method Robustness was carried by a deliberate change in the Flow rate and change in Mobile Phase composition, was made to evaluate the impact on the method.

**Forced degradation studies**

**Acid degradation**

Accurately weighed and transfered 10mg of Telmisartan Working standard into a 100mL volumetric flask.To it 10mL of 0.1N Hcl was added and sonicated for 5minutes. Refluxed under heat at 60 degrees in a heating mantle for 2 hours. The sample solution was neutralized using 0.1N NaOH and diluted up to the mark with Mobile phase. Further pipette 1 ml of the above solution into a 10ml volumetric flask and diluted up to the mark with Mobile phase. Mixed well and filter through 0.45µm filter and injected into HPLC system.

**Base degradation**

Accurately weighed and transfered 10mg of Telmisartan Working standard into a 100mL volumetric flask. To it 10mL of 0.1N NaOH was added and sonicated for 5minutes. Refluxed under heat at 60 degrees in a heating mantle for 2 hours. The sample solution was neutralized using 0.1N Hcl and diluted up to the mark with Mobile phase.Further pipette 1 ml of the above solution into a 10ml volumetric flask and diluted up to the mark with Mobile phase. Mixed well and filter through 0.45µm filter and injected into HPLC system.

**Thermo degradation**

Accurately weighed and transfered 10mg of Telmisartan Working standard into a 100mL volumetric flask and oven under heat at 105 degrees for 12 hours. Further pipette 1 ml of the solution into a 10ml volumetric flask and diluted up to the mark with Mobile phase. Mixed well and filter through 0.45µm filter and injected into HPLC system.

**Peroxide degradation**

Accurately weighed and transfered 10mg of Telmisartan Working standard into a 100mL volumetric flask. To it 10mL of 3% Hydrogen Peroxide ($H_2O_2$) and sonicated for 5minutes and Refluxed under heat at 60 degrees in a heating mantle for 2 hours.

Further pipette 1ml of the solution into a 10ml volumetric flask and diluted up to the mark with Mobile phase. Mixed well and filter through 0.45µm filter and injected into HPLC system.

**RESULTS AND DISCUSSION**

**Method of development**

The chromatographic conditions were optimized with a view to develop a stability- indicating assay method. Two different columns were tried as under chromatographic conditions namely, XTerra® RP $C_8$, 4.6 x 150mm, 3.5 µ. (water, Ireland) and Luna $C_8$ (Octylsilane), 150 x 4.6 mm, 3.5 µ (Phenomenax, USA). XTerra® RP $C_8$ column had given a good peak shape with response at affordable retention time than Luna $C_8$. The chromatographic conditions finally comprised of Methanol: Potassiumdihydrogen phosphate solution (60:40 v/v) at a flow rate of 0.5 ml/min using XTerra® RP $C_8$ column at 230 nm.

**Validation of the method**

**Linearity**

These results indicate that the response is linear over the range of 20, 40, 60, 80, and100 µg/ml of Telmisartan. The results were shown in **Table: 1.**

**Table: 1 Regression characteristics of the proposed RP-HPLC method**

| S.NO. | Regression characteristics | Telmisartan |
|---|---|---|
| 1. | Range (µg/ml) | 20-100 |
| 2. | Detection wave length(λmax) | 230 |
| 3. | Mean '$R^2$' value | 0.9998 |
| 4. | Slope (m) | 2.326 |
| 5. | Intercept (c) | -6.708 |
| 6. | Run time(min) | 5 |
| 7. | Retention time(min) | 2.6 |
| 8. | Theoretical plates(N) | 3025 |
| 9. | Tailing factor | 1.14 |

**Precision**

Method Precision was evaluated by injecting the standard solution of 30 µg mL-1 six times and %RSD was 0.33%. System precision (repeatability) was evaluated by performing six consecutive injections of the 30 µg mL-1 standard solution, giving a low R.S.D. value of 0.16% and no change in retention time of the drug. The Telmisartan contents were found in the tablet formulations using the proposed method. The low R.S.D. values indicate that the proposed method is precise.

**Ruggedness and robustness of the method**

Method robustness and ruggedness was determined by analyzing same sample at normal operating conditions and also by changing some operating analytical conditions such as column make, mobile phase composition, flow rate and analyst. The deliberate aforementioned changes in parameters alter the result of Telmisartan 0.01% to method precision study, which is not a significant change. The robustness and ruggedness of the method shows assay value less than ±2.0%. Table: 2 represent the ruggedness and robustness of the method.

**Table: 2 Ruggedness and robustness of Telmisartan**

| Parameters | Normal (Original) | Changed conditions |
|---|---|---|
| Column make | XTerra RP-$C_8$;4.6 x 150mm; 3.5 µm | Luna RP-$C_8$ 150 x 4.6 mm; 3.5 µ |
| Flow rate | 0.5 | 0.4 |
| Mobile phase Composition | Buffer and Methanol (40:60v/v) | Buffer and Methanol (30:50v/v) |
| Analyst | Sujana.K | Bala souri.O |
| % assay of Telmisartan | 99.25% | 99.22% |

**Limit of detection (LOD) and Limit of Quantitation (LOQ)**

The S/N Ratio values of LOD and LOQ concentrations were found to be 2.88 and 9.62 respectively.

**Accuracy**

The accuracy of the method was established by recovery studies. Results indicate that the individual recovery of Telmisartan ranges from 100.3% to 101.9% with mean recovery of 100.9% and % relative standard deviation of 0.37%. The recovery of Telmisartan by proposed method is satisfactory as % relative standard deviation is not more than ± 2.0% and mean recovery between 99.0 - 102.0%. Table: 3 represent the accuracy of method.

**Analysis of the marketed formulation**

The drug content was found to be 99.22% with a % RSD of 0.87%. It was noted that no degradation of Telmisartan had occurred in the marketed formulation that was analyzed by this method. The low RSD value indicated the suitability of this method for routine analysis of Telmisartan in pharmaceutical dosage form.

**Table: 3 Recovery of Telmisartan**

| %Concentration (at specification Level)* | Area | Amount added (mg) | Amount found (mg) | % Recovery* |
|---|---|---|---|---|
| 50% | 1477896 | 10.3 | 10.5 | 101.9 |
| 100% | 2817481 | 20.0 | 20.07 | 100.3 |
| 150% | 4386755 | 31.0 | 31.2 | 100.6 |
| Mean | | | | 100.9 |
| ± Standard deviation | | | | 0.85 |
| % Relative standard deviation | | | | 0.84 |

*Average of three determinations

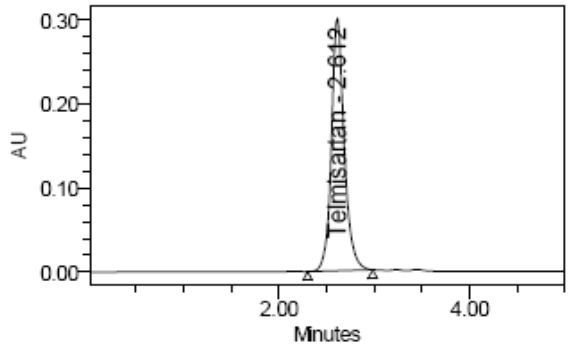


**Fig. 1: The simple chromatogram of standard Telmisartan**

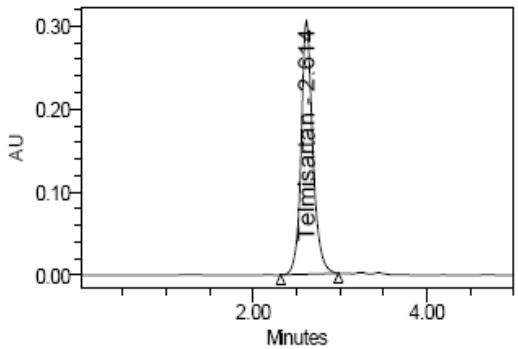


**Fig. 2: The simple chromatogram of test Telmisartan**

**Stability- indicating property**

The chromatogram of no stress treatment sample (as control) showed no additional peak (**Figure: 1& 2**). The retention time (RT) of standard and sample were 2.612 min and 2.614 min respectively

The chromatogram of acid degraded sample showed no additional peaks. The chromatogram of alkali degraded sample showed no additional peaks. The chromatogram of thermal degraded sample showed no additional peaks The chromatogram of hydrogen peroxide degraded sample showed additional peak at RT of 2.90 min (**Fig: 3**). and the values were shown in **Table: 4**.

**Detection of the related impurities**

The sample solution showed no additional peak other than principal peak. Hence, related impurities are not present in the market sample.

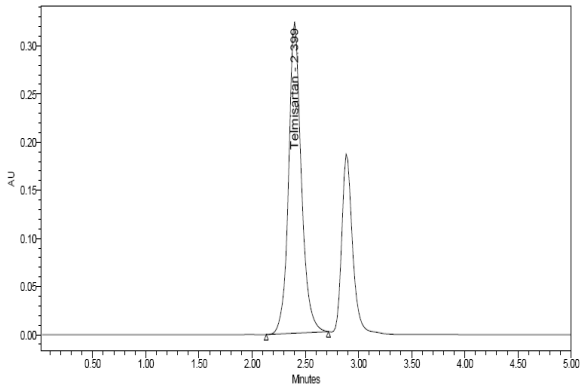


**Fig. 3: The simple chromatogram of Hydrogen Peroxide degraded sample.**

**Table 4: Stressed study data of Telmisartan**

| S. No | Condition | Time(hrs) | % assay of Telmisartan | Retention time of drug | % Degradation | Mass balance (%assay + %degradation products) |
|---|---|---|---|---|---|---|
| 1. | No stress treatment | - | 101.0 | 2.612 | Nil | - |
| 2. | Acid | 2 | - | - | Nil | - |
| 3. | Alkali | 2 | - | - | Nil | - |
| 4. | H202 | 2 | 101.4 | 2.399,2.90 | 1.003 | 102.4 |
| 5. | Thermal | 12 | - | - | Nil | - |

**CONCLUSION**

The developed HPLC technique is precise, specific, accurate and stability-indicating. Statistical analysis proves that the method is suitable for the analysis of Telmisartan as bulk drug and in pharmaceutical formulation without any interference from the excipients. This study is a typical example of a stability-indicating assay, established following the recommendations of ICH guidelines. The method can be used to determine the purity of drug available from various sources by detecting any related impurities. The method has been found to be better than previously reported methods, because of use of a less economical and readily available mobile phase, lack of extraction procedures, no internal standard, and use of the same mobile phase for washing of the column. All these factors make this method suitable for quantification of Telmisartan in bulk drugs and in pharmaceutical dosage forms. It can therefore be concluded that use of the method can save much time and money and it can be used in small laboratories with very high accuracy and a wide linear range.

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**REFERENCES**

**6.** ICH, Q1A (R2): Stability Testing of New Drug Substances and Products, International Conference on Harmonization, Geneva. 2003.

7. Carstensen JT, Rhodes CT. A rational approach to stability testing and analytical development for NCE, drug products marketed product stability testingIn: In: Drug Stability: Principles and Practices, Wolfgang G,, Ed, New York, Marcel Dekker, 2000; pp 415–81.
8. Patil KR, Rane VP, Sangshetti JN and Shinde DB, A Stability-Indicating LC Method for the Simultaneous Determination of Telmisartan and Ramipril in Dosage Form, Chromatographia, 2008, 67, 575-582.
9. Kurade VP, Pai MG and Gude R; Development and Validation of a Reverse Phase Liquid Chromatographic Method for Quantitative Estimation of Telmisartan in Human Plasma, Indian Journal of Pharmaceutical Sciences, 2009, 71(2), 148-151.
10. Kabra V, Agrahari V and Trivedi P, Springer Berlin Heidelberg, Development and Validation of a Reverse Phase Liquid Chromatographic Method for Quantitative Estimation of Telmisartan in Human Plasma, 2009, Volume 23, 1297-1300.
11. Rane VP, Sangshetti JN and Shinde DB, Simultaneous high-performance liquid
12. Chromatographic determination of telmisartan and hydrochlorothiazide in pharmaceutical preparation, Chromatographic science 2008 Nov-Dec; 46(10):887- 91.
13. Bankey S, Tapadiya GG, Saboo SS, Bindaiya S, Jain D and Khadbadi SS, simultaneous Determination of Ramipril, Hydrochlorothiazide and Telmisartan by Spectrophotometry, International Journal of ChemTech Research, 2009, Vol.1 , 183-188.
14. Chitra Prabhu, Ganesa Sundararajan Subramanian,Arumugam Karthik, Suvarna Kini, Mallayasamy Surulivel Rajan and Nayanabhirama Udupa, Determination of telmisartan by HPTLC a stability indicating assay, Journal of Planar Chromatography 2007, December, Volume 20.
15. FDA, Draft Guidance for Industry: Stability Testing of Drug Substances and Drug Products, FDA, Rockville 1998.
16. Snyder L R, Kirkland J J and Glajch J L, Practical HPLC method development, 2nd Edn., Wiley-interscience Publication, John Wiley & Sons, Inc, 1997, 709.