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# Simultaneous HPTLC estimation of Telmisartan and Amlodipine Besylate in tablet dosage form

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## Abstract

*Method describes an HPTLC method for the simultaneous determination of Telmisartan and Amlodipine Besylate from tablet dosage form. This employes a precoated silica gel 60 $F_{254}$ (0.2 mm thickness) on aluminium sheets and a mobile phase Ethyl acetate: 1, 4 Dioxane: Methanol: 25% Ammonia in the ratio of 15:1.5:3:1.5 v/v, having chamber saturation for 30 min at room temperature. The developing chamber was run upto 8cm. The $R_f$ values was found to be 0.16 and 0.33 for Telmisartan and Amlodipine respectively. The plate was scanned and quantified at 323nm. The linear detector response was observed between 100 µg/ml to 500 µg/ml and 200 µg/ml to 1000 µg/ml for Telmisartan and Amlodipine respectively. The method so developed was validated for its accuracy and precision. The LOD and LOQ were found to be 0.025, 0.0747µg/ml and 0.0236, 0.0714µg/ml, respectively for Telmisartan and Amlodipine. The recovery was carried out by standard addition method. The Average recovery was found to be 100.38% and 100.24% for Telmisartan and Amlodipine respectively.*

**Keywords:** High Performance Thin layer chromatography, Method validation, Amlodipine and Telmisartan.

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## INTRODUCTION

Telmisartan, a nonpeptide molecule, is chemically 40-[(1,4-dimethyl-20-propyl[2,60-1H-benzimidazol]-10-yl) methyl]-[1,10-biphenyl]-2-carboxylic acid and Amlodipine is chemically 2 - [(2 - aminoethoxy)methyl] - 4 -(2 - chlorophenyl) - 3 -ethoxycarboxyl - 5 -methoxycarbonyl - 6 - methyl - 1,4- dihydropyridine. Telmisartan is an angiotensin II receptor antagonist that is highly

selective for type 1 angiotensin II receptor. Angiotensin II is the principle pressor agent of the renninangiotensin system, with effects that include vasoconstriction, stimulation of synthesis and release of aldosterone, cardiac stimulation, and renal reabsorbtion of sodium. Amlodipine is a calcium channel blocker (dihydropyridine) used as an anti-hypertensive and in the treatment of angina. Like other calcium channel blockers, amlodipine acts by relaxing the smooth muscle in the arterial wall, decreasing peripheral resistance and hence reducing blood pressure; in angina it increases blood flow to the heart muscle. Telmisartan and amlodipine combination is used to lower blood pressure (hypertensive agent) [1-5].

The literature survey reveals that several methods were reported for the individual estimation of telmisartan and amlodipine. The methods [6–11] for telmisartan in combination with other drugs in plasma, serum and in tablets by UV and LC and [12–15] for the estimation of amlodipine in combination with other drugs in serum and in tablets by UV and LC.[16-20] None of the reported analytical procedures describe a method for simultaneous determination of telmisartan and amlodipine in combined pharmaceutical dosage form.[20-24]

In the present study attempts were made to develop a rapid, economical, precise and accurate method for the simultaneous estimation of the ingredients of this combination.

## MATERIALS AND METHODS

### Experimental

**Instrumentation and chromatographic conditions:**

Instrumentation and chromatographic conditions are given in the following table

**Table A Instruments and chromatographic conditions**

| **Instrument** | **Description** |
|---|---|
| HPTLC system | Camag HPTLC system |
| Sample applicator | camag linomat IV automatic sample applicator |
| scanner | Camag TLC scanner 3 |
| Software | Camag Wincats software |
| Saturation chamber | Camag Twin-trough chamber (10×10cm) |
| HPTLC plates | Merck HPTLC plates coated with silicagel 60 $F_{254}$ (0.2 mm thickness) on aluminium sheets |
| Syringe | Hamilton syringe (100µl) |

**Table B Reagents required for study**

| Reagents | Manufactured by |
|---|---|
| Ethyl acetate AR grade | S d Fine Chemicals, Mumbai |
| 1, 4-Dioxane AR grade | S d Fine Chemicals, Mumbai |
| Methanol AR grade | S d Fine Chemicals, Mumbai |
| 25% Ammonia AR grade | S d Fine Chemicals, Mumbai |

**Preparation of working standard and sample solutions**

Standard stock solutions of Telmisartan and Amlodipine were prepared by dissolving 100 mg of drug in 100 ml of methanol to get a concentration of 1000µg/ml for each drug seperately. The standard working solutions were prepared by dilution of the stock standard solution with methanol to reach a concentration of 50µg/ml and 100µg/ml for Telmisartan and Amlodipine respectively. Twenty tablets were weighed accurately, finely powdered and powder equivalent to 80 mg of Telmisartan was weighed accurately. The powder was transferred to a 100ml volumetric flask containing 10ml of methanol, shake for 5 min, to it 50ml of methanol was added and the solution was sonicated for 20 min, allowed the solution to cool to room temperature and then volume made up to the mark with methanol, the resulting solution was filtered through Whatmann filter paper ≠41. Required dilutions were made to get 50µg/ml and 100 µg/ml for Telmisartan and Amlodipine respectively.

**Chromatographic conditions and Calibration graphs**

The experiment was performed on a silica gel 60 $F_{254}$ (0.2 mm thickness) HPTLC plates (20×10cm) using a mobile phase Ethyl acetate: 1, 4 Dioxane: Methanol: 25% Ammonia (15:1.5:3:1.5 v/v). The plates were prewashed with methanol and activated at $110^0$C for 30min. prior to chromatography. Samples were applied as bands 4mm long, at 4mm intervals. Ascending development to distance of 8cm was performed in saturated 20×10cm twin trough TLC developing chamber for 30min at room temperature (camag). The plate was scanned and quantified at 323nm.

Aliquots of 10, 8, 6, 4, 2 µl of working standard solution of Telmisartan and Amlodipine were applied on the TLC plate. TLC plate was dried, developed and analyzed photo metrically. The standard calibration curve was generated using regression analysis with Microsoft excel.

**Validation of the method**

The developed method was validated as per ICH guidelines for specificity, linearity, repeatability, Limit of detection, limit of Quantitation and accuracy. (Table I)

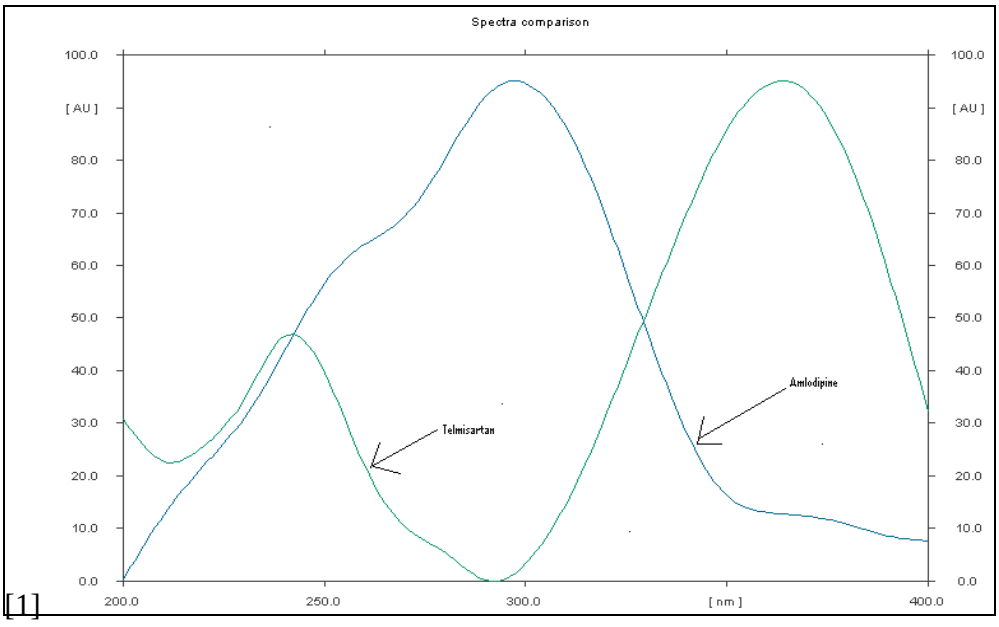
**Assay**

From the prepared sample solution 5µl was spotted in duplicate along with same concentration of standard solution on pre-coated silica gel 60 $F_{254}$ TLC plate. The plate was developed and scanned as mentioned earlier. Analysis was repeated in triplicate. Peak area was recorded and the amount of Telmisartan and Amlodipine present in formulation was estimated. (Table II)

## RESULTS AND DISCUSSION

HPTLC method was optimized with a view to develop a simple, accurate method for estimation of drug in pharmaceutical formulation and in bulk drug. UV scanning at 190-450 nm for both Telmisartan and Amlodipine show that 323 nm is the suitable wavelength for detection of drugs. (Fig.I) The mobile phase of Ethyl acetate: 1, 4-Dioxane: Methanol: 25% Ammonia in the ratio of 15:1.5:3:1.5 v/v. was selected because it gave highest resolution, minimum tailing and $R_f$ values of 0.16 and 0.33 for Telmisartan and Amlodipine respectively. (Fig.II). Telmisartan and Amlodipine showed linearity in the concentration range of 100 µg/ml to 500 µg/ml ($r^2$ =0.9957) and 200 µg/ml to 1000 µg/ml ($r^2$ =0.9990) respectively. For HPTLC method the linearity of calibration graphs and adherence of the system to Beer's law was validated by higher value of correlation coefficient. The LOD and LOQ were found to be 0.025, 0.0747µg/ml and 0.0236, 0.0714µg/ml, respectively for Telmisartan and Amlodipine. Repeatability of measurement of peak area was determined by six replicate spotting and six time measurement of working standard of Telmisartan and Amlodipine and percentage relative standard deviation (%RSD) was found to be 0.40 and 0.69 for Amlodipine and Telmisartan respectively. To confirm the specificity, the solution of formulation was spotted on the TLC plate, developed and scanned. Complete separation of Telmisartan and Amlodipine in presence of common tablet Excipients was noticed, indicating the specificity of the method.

Recovery studies of the drugs were carried out for the accuracy parameter. These studies were carried out at three levels (80%, 100%, and 120%) by standard addition method. after spiking with additional drug afforded recovery of 98-102% and mean recovery was found to be 100.38% and 100.24% for Telmisartan and Amlodipine respectively. (Table III).




[1]

**Fig. I Overlay spectra of standard Telmisartan and Amlodipine**

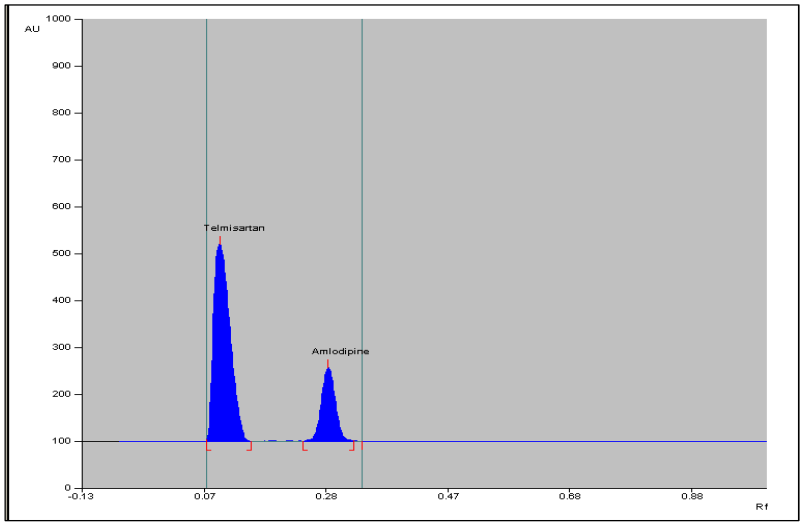



**Fig. II Chromatogram of standard Telmisartan and Amlodipine**

**Table I Method Validation Parameters**

| **Parameters** | Telmisartan | Amlodipine |
|---|---|---|
| Linearity Range | 100-500 (µg/ml) | 200-1000 (µg/ml) |
| Correlation Coefficient | 0.9957 | 0.99903 |
| Limit of Detection (µg/ml) | 0.25 | 0.0236 |
| Limit of Quantitation (µg/ml) | 0.0747 | 0.0714 |
| Precision % RSD | 0.667 | 0.733 |
| Specificity | Specific | Specific |

**Table II Analysis of the marketed formulation**

| Parameters | Amlodipine | Telmisartan |
|---|---|---|
| Label claim (mg/tab.) | 5 | 40 |
| Amount found (mg/tab.) | 5.35 | 40.8 |
| Drug content (%) | 106.8 | 102.22 |
| % R.S.D. | 0.56 | 1.68 |

**Table III Recovery results of Telmisartan and Amlodipine**

| Level of Recovery | Mean of % Recovery | |
|---|---|---|
| | Telmisartan | Amlodipine |
| 80 | 101.40 | 99.84 |
| 100 | 99.19 | 100.95 |
| 120 | 100.55 | 99.94 |

## CONCLUSION

The developed HPTLC method was found to be simple, precise, specific and accurate. Therefore this method can be applied for routine analysis of drugs in formulation and in bulk drug.

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## REFERENCES

[2] ICH, Q2 (R1), Validation of Analytical Procedures: Text and Methodology, International Conference on Harmonization, Geneva, November **1996**, p. 1.
[3] Sethi P.D., HPTLC Quantitative analysis of pharmaceutical formulations, 1st edition, CBS publishers and distributors, New Delhi, **1996**, p. 3-30.
[4] Marydele J. O., Ann S., Patricia E. H., John R. O., Jo Ann R. G., Mary A.D., The Merck Index- An Encyclopedia of Chemicals, Drugs and Biologicals, 13th Edition, Merck Research Laboratories-a division of Merck & Co.Inc., Whitehouse Station, NJ, p. 491, 1628
[5] Indian Pharmacopoeia, 5th edition, Volume 2, Controller of Publications, Govt. of India, Ministry of health and Family Welfare, New Delhi, **2007**, p. 554.
[6] Sweetman S.C., Martindale, the complete drug reference, 34th edition, Pharmaceutical press(a division of Royal Pharmaceutical Society of Great Bretain), **2005**, p. 862, 1010.
[7] Palled M.S., Chatter M., Rajesh P.M., Bhat A.R., *Indian Journal of Pharmaceutical Sciences*, **2006,** 68(5), , p. 685-686.
[8] Lories I., Samah S. A., Fattah L.A., Heba H. R., *Il Farmaco*, **2005**, 60(10), P. 859-867.
[9] Wankhede S.B., Tajne M.R., Gupta K.R., Wadodkar S.G., *Indian Journal of Pharmaceutical Sciences*, **2007**, 69(2), , p.298-300.
[10] Shah N.J., Suhagia B.N., Shah R.R., Shah P.B., *Indian Journal of Pharmaceutical Sciences*, **2007**, 69(2), , p. 202-205.
[11] Bhat L.R., Godge R.K, Vora A.T., Damle M.C., *journal of liuid Chromatography & related technologies*, **2007**, 30(20), p. 3059-3067.
[12] Patil K.R., Rane V. P., Sangshetti J. N., Shinde D.B., *Chromatographia*, **2008**, 67(7-8), , p. 575-582.
[13] Gohil K., Trivedi P., Molvi K.I., *Indian Journal of Pharmaceutical Sciences*, **2005,** 67(3), , p. 376-378.
[14] Singhvi I., Chaturvedi S.C., *Indian Journal of Pharmaceutical Sciences*, **1998**, 60(5), p. 309-310.
[15] Bhat L.R., Vora A.T., Godge R.K., Damle M.C., *Indian Drugs*, **2007**, 44(12), p. 915-919.
[16] Chitlange S.S., Imran M.D., Wankhede S.B., Sakarkar D.M., *Journal of Pharmaceutical research*, **2008**, 7(1), , p. 36-38.
[17] Chitlange S.S., Wankhede S.B., Sakarkar D.M., *Journal of Pharmaceutical research*, **2008,** 7(1), p. 53-55.
[18] Nikalje A.G., Choudhari V.P., Kulkarni R.R., Jagdale S.C., Choudhari V., *Journal of Pharmaceutical research*, **2006,** 5(4), p. 113-115.

[19] Mishra P., Gupta A., Shah K., *Indian Journal of Pharmaceutical Sciences,* **2007,** 69(1), p.110-111.
[20] Rahman N., Singh M., Hoda N., *Il Farmaco*, **2004,** 59(11), P.913-919.
[21] Juval V., Kumar G., Chaudhary M., Gupta R., Kumar P., Mittal P., Agrawal A.C., *Indian Pharmacist*, **2008,** 7(68), p. 77 -79.
[22] Dongre V.G., Shah S B., Karmuse P.P., Phadke M., Jadhav V.K., *Journal of Pharmaceutical and Biomedical Analysis,* **2008,**46(3), P. 583-586.
[23] Klinkenberg R., Streel B., Ceccato A., *Journal of Pharmaceutical and Biomedical Analysis*, **2003,** 32(2), P. 345-352.
[24] Kamat K., Chaturvedi S.C., *Indian Journal of Pharmaceutical Sciences*, **2005,** 67(2), p. 236-239.
Mohammadi A., Rezanour N., Ansari M., Ghorbani F., Hashem M., Walker R., *Journal of Chromatography B,* **2007,** 846(1-2), P. 215-221