

Spectrophotometric Determination of Trimetazidine Dihydrochloride in Pharmaceutical Preparations

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Two simple and sensitive visible spectrophotometric methods (A and B) have been described for the assay of trimetazidine dihydrochloride (TMZ) either in pure form or in pharmaceutical formulations. These methods are based on the oxidation followed by complexation between TMZ and *o*-phenanthroline or potassium ferricyanide in presence of ferric chloride to form a coloured product with λ_{\max} at 510 nm and 760 nm for method A and B respectively. All the variables have been optimized and the reaction sequence is presented. The concentration measurements are reproducible within a relative standard deviation of 1.0%. Recoveries are 97.9-101.75%.

Key words: Spectrophotometric, determination, trimetazidine dihydrochloride, pharmaceutical.

INTRODUCTION

Trimetazidine dihydrochloride (TMZ), chemically 1-[2,3,4-trimethoxy phenyl] methyl] piperazine, is used in angina pectoris and in ischaemia of neurosensorial tissues as in Meniere's¹ disease. TMZ regulates ionic and extracellular exchanges, correcting the abnormal flow of ions across the cellular membrane caused by ischaemia and preventing cellular oedema caused by anoxia. Few physico-chemical methods appeared in the literature for the determination of TMZ in biological fluids. They are based on the HPLC^{2,3} and GC-MS⁴. Only HPLC⁵ and GC⁶ methods have been reported for pharmaceutical preparations.

Although spectrophotometric methods are the instrumental methods of choice commonly used in industrial laboratories, no colorimetric method has been reported so far for the determination of TMZ. Therefore the need for a fast, low cost and selective method is obvious, especially for routine quality control analysis of pharmaceutical formulations, containing TMZ.

Ferric salts (ferric chloride) play a prominent role in the colorimetric determination of organic compounds. Acting as an oxidant a ferric salt converts into ferrous salt. They can be easily detected by the usual reagent for divalent iron, potassium ferricyanide ($K_3Fe(CN)_6$)⁷, *ortho*-phenanthroline (*o*-PTL)⁸, bipyridyl or triazine⁹. We have applied the above two reagents (*o*-PTL-Fe(III) and Fe(III)- $K_3Fe(CN)_6$) for the determination of TMZ in bulk samples and pharmaceutical formulations.

EXPERIMENTAL

All chemicals were of analytical grade and all the solutions were prepared with double distilled water. Freshly prepared solutions were always used. Aqueous

solutions of *o*-PTL (Loba) 0.198%, Fe(III) (S.D. Fine Chem) 0.9%, $K_3Fe(CN)_6$ (Loba) 1.0%, orthophosphoric acid (Qualigens) 0.02 M were prepared. A Systronics model 117 UV-Visible spectrophotometer with 1 cm matched quartz cells was used for all the absorbance measurements.

Standard and sample solutions: TMZ (100 mg) was accurately weighed and dissolved in 100 mL of distilled water (1 mg/mL). This stock solution was further diluted with distilled water to get a working standard solution of $100 \mu\text{g mL}^{-1}$ for both methods.

Analysis of pure samples

Method A: A portion of TMZ solution (1–5 mL, $100 \mu\text{g mL}^{-1}$) was accurately transferred to a 10 mL standard flask. After that 0.5 mL Fe(III) solution and 1.5 mL of *o*-PTL were added and the flasks were allowed to stand for 15 min at room temperature. Then 0.5 mL of orthophosphoric acid was added to each flask and the solution was made up to volume with distilled water. The absorbance was measured at 510 nm during the next 5 min against a reagent blank prepared in a similar manner. The amount of the drug in the simple was computed from Beer Lambert plot.

Method B: An aliquot of TMZ solution containing 0.5 to 2.5 mL ($100 \mu\text{g mL}^{-1}$) was delivered to a series of 10 mL graduated test tubes. Then 1.5 mL of Fe(III) and 1.5 mL of $K_3Fe(CN)_6$ were added and the volume made up to 10 mL with distilled water and kept aside for 40 min at room temperature. The absorbance of the coloured species formed was measured at 760 nm against a reagent blank. The amount of TMZ in the sample was computed from Beer Lambert plot.

Analysis of pharmaceutical formulations

Tablet powder equivalent to 100 mg was taken and the sample solution prepared as described for the standard solution and filtered if insoluble material was present prior to analysis as described for pure samples.

RESULTS AND DISCUSSION

The Beer's law limits, molar absorptivity, Sandell's sensitivity, regression equation and correlation coefficient were obtained by the least square treatment and these results are given in Table-1. The precision of each method was tested by analyzing six replicate samples containing 40, $20 \mu\text{g mL}^{-1}$ of pure drug for methods A and B respectively. The per cent standard deviation and the per cent range of error at 95% confidence level of each method are given in Table-1.

Commercial formulations (Tablets) containing TMZ were successfully analyzed by the proposed methods. The values obtained by the proposed and reference (UV method) for formulations were compared statistically by the t- and f-tests and found not to differ significantly. As an additional demonstration of accuracy, recovery experiments were performed by adding a fixed amount of the drug to the pre-analyzed formulations. These results are summarized in Table-2. The ingredients usually present in formulation of TMZ did not interfere with the proposed analytical methods.

TABLE-1
OPTICAL AND REGRESSION CHARACTERISTICS, PRECISION
AND ACCURACY OF THE PROPOSED METHODS

Parameters	Methods	
	A	B
λ_{\max} (nm)	510	760
Beer's law limit ($\mu\text{g mL}^{-1}$)	10–50	5–25
Molar absorptivity ($\text{L mol}^{-1}\text{cm}^{-1}$)	4.7502×10^3	1.0857×10^4
Sandell's sensitivity ($\mu\text{g cm}^{-2}$ absorbance unit/0.001)	0.0714	0.0312
Regression equation (Y) ^a		
Slope (b)	0.01426	0.0317
Standard deviation on slope (S_b)	0.0502×10^{-3}	0.5999×10^{-5}
Intercept (a)	-0.004	0.0039
Standard deviation on intercept (S_a)	1.6646×10^{-3}	9.9497×10^{-5}
Standard error of estimation (S_e)	1.5874×10^{-3}	9.4868×10^{-5}
Correlation coefficient (r)	0.99989	0.9999
Relative standard deviation (%) ^b	0.5255	0.5119
% Range of error ^b (95% confidence limit)	0.4394	0.4280
Detection limit ($\mu\text{g mL}^{-1}$)	0.8415	0.3691

^aWith respect to $Y = a + bC$, where C is concentration ($\mu\text{g mL}^{-1}$) and Y is absorbance.

^bsix replicate samples.

TABLE-2
ASSAY AND RECOVERY OF TMZ IN PHARMACEUTICAL FORMULATIONS

Pharmaceutical formulations	Labelled amount found (mg)	Amount found ^a (mg) using proposed methods		Found by reference method	% Recovery by proposed methods ^b	
		A	B		A	B
Tablet I	20	19.98 ± 0.14 $t = 0.88$ $F = 1.04$	20.15 ± 0.27 $t = 1.33$ $F = 0.64$	19.90 ± 0.14	99.9 ± 0.38	100.75 ± 0.14
Tablet II	20	19.95 ± 0.17 $t = 0.74$ $F = 1.24$	20.31 ± 0.31 $t = 0.87$ $F = 0.64$	20.01 ± 0.23	99.75 ± 0.56	101.55 ± 0.61
Tablet III	20	20.12 ± 0.25 $t = 1.74$ $F = 1.64$	20.35 ± 0.35 $t = 2.11$ $F = 1.05$	20.09 ± 0.37	100.60 ± 0.29	101.75 ± 0.22
Tablet IV	20	19.96 ± 0.12 $t = 0.65$ $F = 0.85$	19.7 ± 0.38 $t = 1.39$ $F = 1.24$	19.80 ± 0.34	99.80 ± 0.55	98.50 ± 0.62

^aAverage \pm standard deviation of six determinations; the t- and F-values refer to comparison of the proposed method with the reference method. Theoretical values at 95% confidence limits, $t = 2.57$, $F = 5.05$.

Recovery of 10 mg added to the pharmaceutical formulations (average of three determinations).

In both the methods same concentration of Fe(III) was tried and it was found that 0.6–1.0 mL is optimum for method A and 1.0–2.0 mL is optimum for method B. 1.0–2.0 mL of *o*-PTL or $K_3Fe(CN)_6$ was suitable for maximum sensitivity.

Chemistry of coloured species: Methods A and B depend upon the oxidation of TMZ with Fe(III) and subsequent coloured complex formation of the resulting Fe(II) ion with *o*-PTL or potassium ferricyanide. *o*-PTL forms a complex of low tinctorial value with Fe(III) which in turn functions as a better oxidant than Fe(III) itself. The reduction product is a tris complex of Fe(II), well known as ferroin. In conclusion the proposed methods are simple, sensitive, accurate and useful for the routine determination of TMZ in pure samples and in pharmaceutical formulations.

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