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Simple Stability Indicating Methods for Metaxalone by RP-HPLC

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Abstract

A novel stability-indicating high-performance liquid chromatographic assay method was developed and validated for Metaxalone in the presence of degradation products generated from forced decomposition studies. Metaxalone is a skeletal muscle relaxant. The method was based on the separation of Metaxalone on Hypersil BDS C_{18} (150 mm x 4.6 mm, 5 µm) column using isocratic mobile phase Methanol: water (90:10, v/v) with a flow rate of 1 ml/min and the UV detection at 279 nm. And the column temperature was maintained at ambient condition. The linearity of the method was found to be 10-60 µg/ml the mean % RSD was found to be 1.67 and 1.86. Forced degradation studies have been carried out for metaxalone. Sample was exposed to various stress conditions like, treatment with acid (0.1 N HCl), alkali (0.1 N NaOH), heat (60°C for 30 min) and peroxidation (H_2O_2) effects has been determined and degradation products were studied. Sample was degraded to 75-85 % in acid, alkali and peroxidation treatment. But found to be stable for heat treatment. The method was validated as per ICH guidelines. All the validation parameters were found to be within the predetermined limits. This shows that the proposed method was sensitive and selective for the determination of stability indicating assay of MET in bulk and its tablet dosage form.

Keywords: Metaxalone, ICH guidelines, stability-indicating and Methanol.

Contents

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1. Introduction

Metaxalone, chemically 2-[(3, 4-dimethylphenoxy) methyl]-2-oxazolidinone is centrally acting muscle relaxant.[1] Figure 1 shows the chemical structure of Metaxalone. The mechanism of action of Metaxalone in humans has not been established, but may be due to general central nervous system depression [2]. Metaxalone has no direct action on the contractile mechanism of striated muscle, the motor end plate, or the nerve fibre. There is very limited or inconsistent data regarding the effectiveness and safety of Metaxalone [3]. Metaxalone is one of the commonly used muscle relaxant therapies for acute low back pain [4].

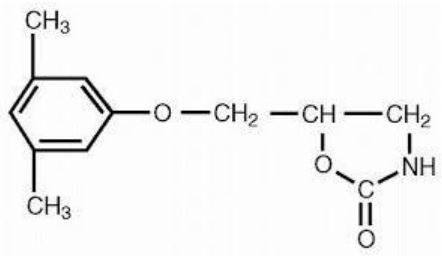

Figure 1: Chemical structure of Metaxalone

Literature survey revealed that, RP-HPLC [5–9], LC–MS [10], HPTLC [11] and UV spectrophotometric methods [12-14] are available for the estimation of MET alone or in combination with diclofenac sodium. This present study reports for the estimation of Metaxalone by HPTC in bulk drug and in tablet dosage form. The proposed method is validated as per ICH guidelines.15-18.

2. Materials and Method

Chemicals and reagents:

Pharmaceutical grade Metaxalone (MET) was gifted by MSN Laboratories, Hyderabad, India. Methanol (HPLC grade) was purchased from Merck Chemical Company, India. HPLC grade water was prepared in house using Milli-Q water filtration system. Sodium hydroxide, HCl and H_2O_2 was purchased from SD Fine chem., Mumbai. All the reagents were of AR grade.

Preparation of Mobile Phase: 900 ml of HPLC grade Methanol was placed in a 1000 ml volumetric flask. And 100 ml HPLC grade water was added to it. The solution was mixed, filtered through the membrane filter and sonicated for 5 min. Mobile phase was freshly prepared daily. And the same was used as diluents.

Preparation of stock solution:

100 mg of pharmaceutical grade MET was weighed and dissolved in mobile phase in a 100 ml volumetric flask. The final volume was made up to the mark with the same to get 1 µg/ml primary stock solution. This solution was sonicated for 5 min. . From the above stock solution 3 ml was further diluted with the mobile phase in a 100 ml volumetric flask. The final volume was made up to the mark with the same. The required working standards for linearity were prepared from the primary stock solution.

Forced decomposition studies

Forced degradation studies were performed on MET bulk drug. Intentional degradation was attempted to stress conditions of UV light (279 nm), heat 30 ^{0}C, acid (0.1N HCl), base (0.1N NaOH) and oxidation (H_2O_2) to determine the ability of the proposed method to separate MET from its impurity and degradation products generated during forced decomposition studies. For heat and light studies, study period was 10 days whereas for acid, base and oxidation it was 48 hrs. Peak purity test was carried out on the stressed samples by using DAD. Assay was also calculated for bulk sample by spiking with MET at the specification 99.98.

Method validation: The analytical method validation was carried out as per ICH method validation guidelines. The following validation parameters were addressed: specificity, precision, linearity, accuracy, limit of detection, limit of quantization, robustness and stability of Metaxalone in diluents.

3. Results and Discussion

Degradation was not observed in MET samples under stress conditions thermal exposure. The drug degradation was observed when MET was exposed in solid state to acid, base, and oxidation.

System suitability: System suitability test was an integral part of method development and has been used to ensure adequate performance of the chromatographic system. Retention time (Rt), capacity factor (k), number of theoretical plates (N) and tailing factor (T) were evaluated for six replicate injections of the drug at a concentration of 30 µg/ml. The results were within the acceptable limits .The Results were shown in Table 1.

Table 1: Results of System Suitability

Parameter	Result	Acceptance Limit
Retention time (Rt)*	3.01 min	--
Resolution factor*	NA	--
Number of theoretical plates (N)*	11,315	More than 3000
Tailing factor (T)*	1.079	Less than 2
Similarity factor*	1.012	0.9-1.2
* Number of injections: 6 replicates		

Linearity:
MET showed linearity in the concentration range of 10–60 µg/ml. The regression equation obtained was Y=3841x+69745 and r^2 = 0.998. Where Y is peak area and X is concentration of MET (µg/ml). This equation was used to determine the amount of MET present in the stress induced samples.

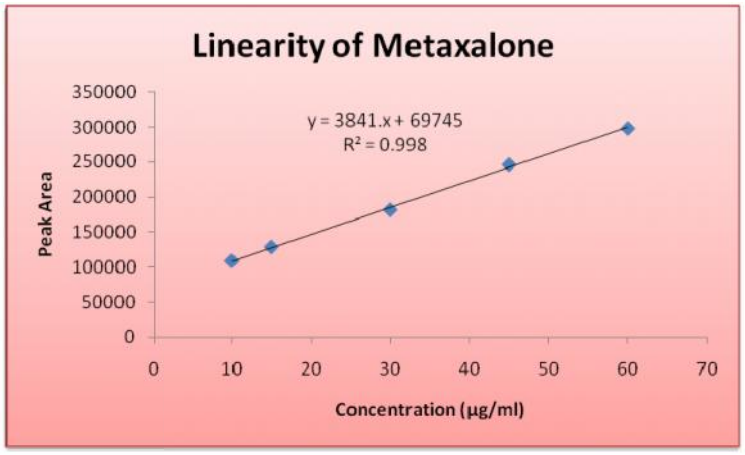

Figure 2: Linearity of Metaxalone

Limits of detection and quantification:
The LOD was defined as the lowest concentration of MET resulting in a signal-to-noise ratio of 3:1 and LOQ was expressed as a signal-to-noise ratio of 10:1. The LOD and LOQ obtained were 0.75 and 2.48 µg/ml, respectively.

Accuracy:
Accuracy of the method was determined by performing the recovery experiments. Known amount of the standard at 50%, 100% and 150% levels was fortified to the degradation sample. Peak area of the standard was calculated by the difference of peak area between fortified and unfortified samples. Six replicate samples of each concentration level were prepared and the percentage recovery at each level (n=6) was determined For MET, the results obtained are in good agreement with the added amounts. The results were shown in table 2.

Table 2: Results of Accuracy

Concentration (µg/ml)	Peak area	µg/ml added	µg/ml found	Recovery	% Mean Recovery	Mean Recovery
15	78879.25	14.92	14.72	98.65	97.56	100.96
	78266.37	14.92	14.61	97.88		
	77832.20	14.92	14.53	97.34		
	77814.40	14.92	14.52	97.32		
	77870.18	14.92	14.53	97.39		
	77377.38	14.92	14.44	96.77		
30	161539.80	29.84	30.15	101.02	102.52	
	166618.95	29.84	31.09	104.19		
	163668.84	29.84	30.54	102.35		
45	243834.60	44.77	45.51	101.65	102.79	
	241926.56	44.77	45.15	100.86		
	240678.50	44.77	44.92	100.34		
	253911.05	44.77	47.39	105.85		
	249537.41	44.77	46.57	104.03		
	249547.30	44.77	46.57	104.03		

Precision:
Intraday and inter day precision was evaluated by injecting six different concentrations (10, 30, 50, and 80 mg/ml) of MET. For intra-day variation, sets of six replicates of the optimized concentrations was analysed on the same day; for inter- day variation, six replicates was analysed on six different days. The intra-day and inter-day precision (% RSD) was found to be less than 2%. The results was shown in table 3, indicating that the method was precise.

Table 3: Results of Precision

	Peak area	
	Interday	**Intraday**
	159254.15	159144.15
	165387.78	163287.78
	161616.72	161616.72
	157449.09	154549.09
	160787.42	160787.42
	160016.55	160001.55
Mean	160751.95	159897.79
SD	2680.79	2980.79
%RSD	1.67	1.86

Specificity:

Specificity is the ability to measure accurately and specifically the analyte of interest in the presence of other components that may be expected to be present in the sample matrix. The specificity of the HPLC method was illustrated in fig. 3 to 6, where complete separation of MET was noticed in the presence of degradation products.

Forced Degradation studies:

MET was subjected to forced degradation in acid, alkali and oxidation it was observed that the sample was found to be sensitive and the degradation products were eluted with sample, shown in the fig. 3 to 5. The sample was found to be stable under heat and light treated samples, shown in fig.6.

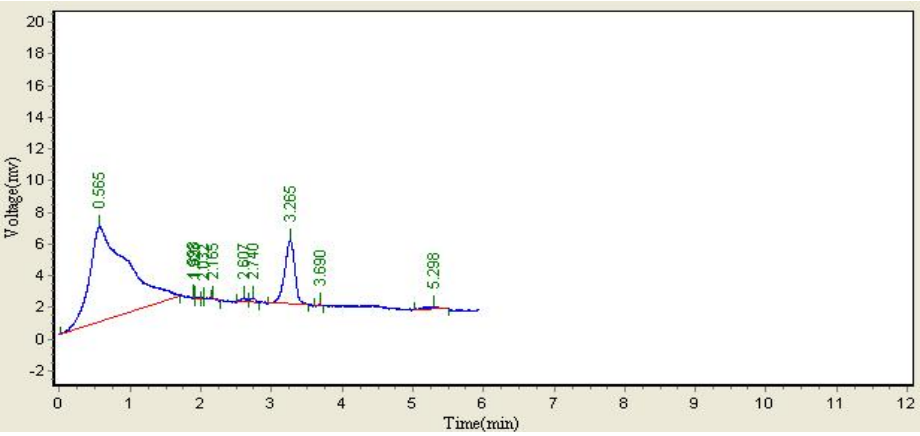

Figure 3: Chromatogram of Acid treated sample

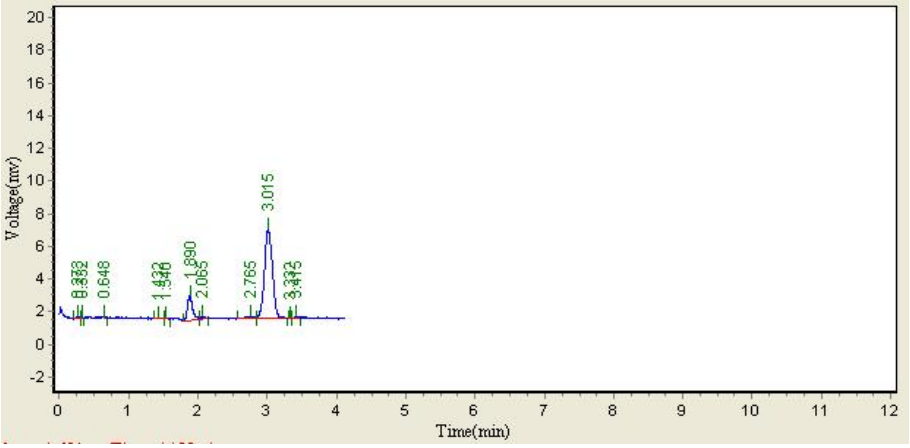

Figure 4: Chromatogram of Alkali treated with sample

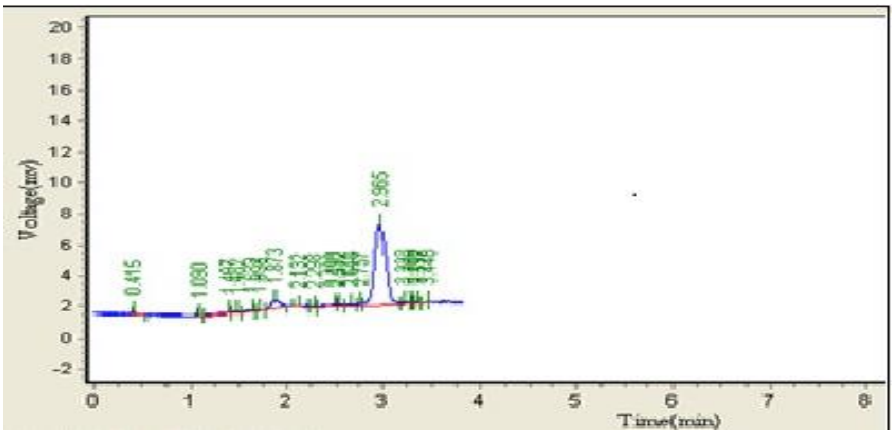

Figure 5: Chromatogram of oxidising agent treated with sample

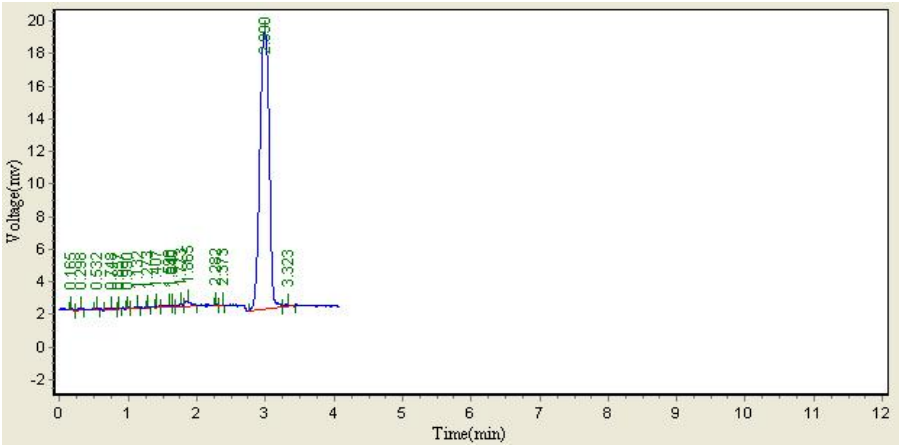

Figure 6: Chromatogram of heat treated with sample

4. Conclusion

A stability-indicating HPLC assay method was developed for the quantization of Metaxalone along with its degradation products. Method was found to be stable for Heat treatment, where as the degradation products were co-eluting along with the peak of the drugs in acid, base and oxidation samples. This indicates that the method was sensitive to acid, base and peroxidation treatment.

5. References

1. Gabhane KB, Kasture V, N. Shrikhande, N. Barde and P. Wankhade simultaneous spectrophotometric determination of metaxalone and diclofenac Potassium in combined tablet dosage form. Int. J. Chem. Sci.: **2009,** 7(1): 539-545.
2. Gruszecki A.C, Kloda S, Simmons G.T, Daly T.M, Hardy R.W, and Robinson C.A. Polydrug fatality involving metaxalone. J. Forensic Sci. **2003**, 48:432-434.
3. Nissen D, Ed. Mosby's Drug Consult & Trade, 13th ed. Mosby, Inc., St. Louis, MO, **2003**.
4. Physicians' Desk Reference, 58th ed. Medical Economics Co., Montvale, NJ, **2004**, pp. 2181.
5. Sahu P.K, Annapurna M.M, Sahoo D.K, Development and validation of stability indicating RP-HPLC method for the determination of Metaxalone in bulk and its pharmaceutical formulations, E-J.Chem. **2011**, 8(S1): S439–S447.
6. Nagavalli D, Sankar A.S.K, Anandkumar K, etal., Estimation of Metaxalone in bulk and in tablet dosage form by RP-HPLC, Res. J.Pharm.Technol. **2010**, 3: 409–411.
7. Cacae J, Reilly E.E, Amann A, Comparison of dissolution of Metaxalone tablets (skelaxin) using USP apparatus 2 and 3, AAPS Pharm.Sci.Tech.5(2004).
8. Gabhane K.B, Simultaneous estimation of Metaxalone and diclofenac potassium in combined dosage form by high performance liquid chromatography method, Int. J.Chem.Sci. **2010**, 8: 253–260
9. Nirogi R.V.S, Kandikere V.N, Shukla M, etal., Quantification of Metaxalone in human plasma by liquid chromatography coupled to tandem mass spectrometry, J.Anal.Toxicol. **2006**, 30(4): 245–251.

10. Rajput M. P, Bharekar V.V, Yadav S.S,etal., Validated HPTLC method for simultaneous estimation of diclofenac potassium and Metaxalone in bulk drug and formulation, Pharm. Globale, **2011,** 2(12): 1–4.
11. Gabhane K.B, Kasture A.V, Shrikhande V.N, etal., Simultaneous spectrophotometric determination of Metaxalone and diclofenac potassium in combined tablet dosage form, Int .J. Chem. Sci. **2009**, 7(1): 539–545.
12. Rao N.R, Bn A.K, Laxmi A, etal., Method validation of Metaxalone drug by using UVspectroscopy, Int. J. Pharm. Bio. Sci. **2011,** 1(3): 284–302.
13. Mahipal reddy N.V, Mohammed Ishaq B, Rajan V.S. T, Gopinath C, Analytical Method development and validation of Metaxalone in bulk and its pharmaceutical formulation by UV spectroscopic method, Int. Res. J. Pharm. **2013**, 4(3).
14. Panda S. S, Patanaik D, Ravi Kumar B.V.V, New stability- indicating RP-HPLC method for determination of diclofenac potassium and Metaxalone from their combined dosage form, Sci. Pharm. **2012**, 80(1): 127–137.
15. ICH, Q2 (R1) Validation of Analytical Procedure, Test and Methodology, International Conference on Harmonization, Geneva, **2005**.
16. Mohammed Ishaq B, Abdul Ahad H, Shaik Muneer, Parveen S, Fahmida B, Analytical Method Developement and Validation for the Estimation for Temozolomide in Phsophate Buffer pH 2.0 as a solvent by UV Spectroscopy Int. Res. J. Pharm. **2014**, 5(1).
17. Muneer S, Jose Gnana Babu C, Ruksana Hakeem, Sai Sumanth K, Development And Validation of Rp-Hplc Method For Estimation Of Rufinamide In Bulk And Its Pharmaceutical Dosage Form Int. J. Pharm. Res. Anal. **2012**, 2(1).
18. Mohammed Ishaq B, Abdul Ahad H, Shaik Muneer, Parveen S, Fahmida B, Development and validation of UV spectrophotometric method for quantitative estimation of Temozolomide in 0.1 N HCl as a solvent J. Pharm. Biomed. Anal. Lett. **2014**, 2(1).