International Journal of All Research Education and Scientific Methods (IJARESM), ISSN: 2455-6211
Volume 10, Issue 4, April-2022, Impact Factor: 7.429, Available online at: www.ijaresm.com

Method Development and Validation of Nimesulide and Racemethionine in Combined Tablet Dosage form by UV-Absorbance Ratio Method

Kalyani S. Nadgauda[*1], D.P. Kardile[1], R.V. Shete [1], V.C. Bhagat[1]

[1]Rajgad Dnyanpeeth's College of Pharmacy, Bhor, Pune Maharashtra

----------**********----------

ABSTRACT

A novel, simple, accurate, precise and reproducible UV-Spectrophotometric method is being build up for the simultaneous estimation of Nimesulide and Racemethionine in tablet dosage form for first time. The stock solutions were prepared in methanol and water. The λmax for Nimesulide and Racemethionine were 228 nm and 226 nm respectively with 215 nm Iso- Absorptive point. The both Nimesulide and Racemethionine complied Beer's law in concentration range of 50-372ppm and 100-750ppm respectively. The Results of analysis of absorbance ratio method were analyzed and validated for various parameters in accordance with ICH guidelines for accuracy, precision, linearity, robustness, LOD and LOQ. The intended method is highly sensitive, precise and accurate, therefore can be used for intended purpose.

Keywords: Nimesulide, Racemethionine, Absorbance Ratio Method, Validation, ICH

INTRODUCTION

Racemethionie i.e. is DL Methionine is one of the sulphur containing essential amino acids. It Outlined chemically as 2-amino-4-(methulthio)butanoic acid. Its use as Antidote.[4] Nimesulide (C13H12N2O5S) is chemically N-(4-Nitro-2-Phenoxyphenyl) methane sulphonamide is a non-steroidal anti-inflammatory drug (NSAID) of Sulfonanilide class having pain medication and fever tumbling properties. Its approved indication of treatment of acute pain, the symptomatic treatment of osteoarthritis, and primary dysmenorrhoea in adolescents and adults more than 12 years old.[1-3] The literature survey for Nimesulide alone and Racemethionine alone make known that lot of methods based on different technique like UV spectrophotometric methods, but no any single method is availed for combination of these two drugs.[5-7] This announcement forms the first report of simple, sensitive, and reproducible methods for the method development and validation of Nimesulide and Racemethionine in combined tablet dosage form by UV- absorbance ratio method.

MATERIALS AND METHODS

Instrumentation

Electronic balance (Mettler Toledo)
Sonicator (Pci- Analytics)
PH Meter (Mettler Toledo)
UV-Visible spectrophotometer (Thermo scientific)

Chemicals and reagents

The pharmaceutically pure chemical sample of Nimesulide was procured from Bajaj healthcares Ltd. Thane and Racemethionine were procured from Prado chemicals. (Mumbai).The Namsafe tablet (Nimesulide 100 mg and Racemethionine 50 mg) was procured from the local drug market. All chemicals and reagents have used was of analytical grade.

Preparation of standard solutions:

1. Preparation of Std. stock Soln.:

50.2 mg of Nimesulide was dissolved in 100 ml of methanol solvent and 100.8mg Racemethionine was dissolved in 100 ml of water solvent to get 1000 µg/ ml of soln. Then further from this prepared solution 10ml of both standard stock solution transferred to another flask of volumetric separately for both drugs and then diluted to 20ml using methanol solvent for Nimesulide and water solvent for Racemethionine .

2. Preparation of sample Soln.

The mean weight of twenty tablets were determined and then that it crushed to fine powder; the amount equal to (powder) 50 mg Nimesulide and 100mg of Racemethionine was diluted to 50ml with Methanol and water respectively with vigorous shaking for 10min. The volume is made to the mark by diluent and filtered by Whatmann filter paper (no. 41) to forms mg/ml of soln. and the this soln. was further utilized to prepare samples of various attentiveness.

Absorption maxima determination of Solution:

The prepared sol$^{n.}$ has scanned against UV range 200- 400 nm.Max. Wavelength for drug of Nimesulide and drug of Racemethionine was found 228 nm and 226 nm respt. And from this got isoabsorptive point at 215nm.

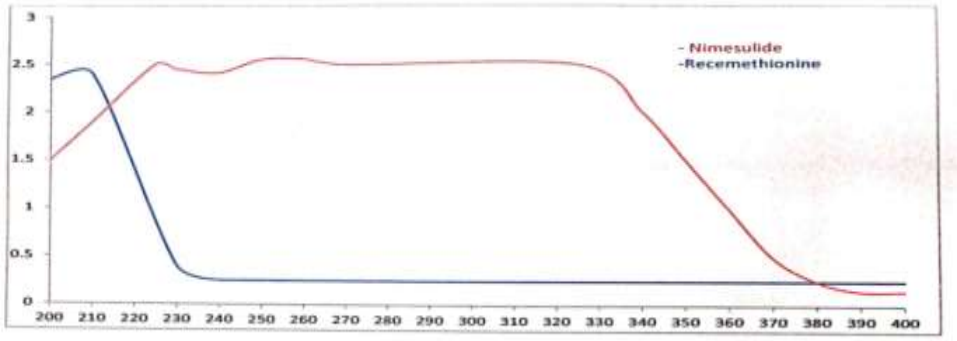

Figure 1: Overlain spectra of Nimesulide & Racemethionine

Absorbance ratio Method:

Through the Nimesulide and Racemethionine overlain spectrum, 2 wavelengths were selected one at 228 nm
(λ maxof Nimesulide) and other at 215nm (Isoabsorptive point). The method shows Q- Values point and the
Attentiveness of drugs in Sol$^{n.}$ of sample received by using following 2 equations.

$$Cx = \{(QM-Qy)/(Qx-Qy)\} (A1/ax1) \text{-------} 1$$

$$Cy = \{(QM-Qx)/(Qy-Qx)\} (A1/ay1) \text{-------} 2$$

Where, A1 and A2 are the absorbance of mixture at
228 nm and 215 nm;
ax1 and ay1 they are absorptivities of Nimesulide and
Racemethionine at 215 nm;
ax2 and ay2 are absorptivities of Nimesulide and Racemethionine at 228 nm;
QM = A2/A1,
Qx = ax2/ ax1,
Qy = ay2/ay1.

METHOD VALIDATION PARAMETERS

Specificity:

Specificity of the method was decided by comparing the absorbance values of standard and sample solution and the absorbance of the sample solution is identical as that of the standard solution.

Precision:

a. System precision: Standard solution prepared as per test method and tested five times.
***b. Method precision*:** Prepared six sample preparations individually using single as per test method.
Accuracy:
The trustworthiness and validity of given methods were done by performing studies of recovery. The recovery of added std. 50 percent, 100 percent & 150 percent was found at 3 attentiveness & 2 duplicate levels. The mean value for statically analysis of % recovery was found satisfactory in range from 100.40 to 100.18 % for Nimesulide drug and 100.40 to 100.18 % for drug of Racemethionine.

Linearity:
The proposed given method was found linear for the range of 50- 375ppm and 100- 750ppm with coefficient of correlation i.e. R^2 is 1 and 1 for Nimesulide drug and Racemethionine drug resp. The curve calibration of Nimesulide and Racemethionine plotted to acquire the calibration graph.

Robustness:
Robustness was performed by changing the preparation process for Nimesulide improves its resilience. Instead of 200ppm concentrations, 250ppm concentration of Nimesulide was used.

RESULTS AND DISCUSSION

System suitability parameter:
Five replicates of standard solution (500ppm) of Racemethionine and (200ppm) of Nimesulide were prepared. The absorbance, SD. and %RSD was detected.

Table 1 Data of Racemethionine and Nimesulide System Suitability

	Injection	Racemethionin Absorbance		Injection	Nimesulide Absorbance
500 ppm Concentration	1	0.825	200 ppm Concentration	1	0.525
	2	0.820		2	0.520
	3	0.834		3	0.534
	4	0.819		4	0.519
	5	0.827		5	0.516
Statistical Analysis	Mean	0.825	Statistical Analysis	Mean	0.523
	SD	0.006		SD	0.007
	% RSD	0.73		% RSD	1.35

Specificity:

Table 2- UV data of Specificity

Solution Type	Drug Name	Absorbance
Standard	Nimesulide	0.524
Solution	Nimesulide	0.521
Solution Type	Drug Name	Absorbance

Standard	Racemethionine	0.831
Solution	Racemethionine	0.833

Precision:

System Precision:

Table 3: Data of Repeatability (System precision) for Nimesulide and Racemethionine

	Injection	Racemethionin Absorbance		Injection	Nimesulide Absorbance
751 ppm Concentration	1	0.830	100 ppm Concentration	1	0.509
	2	0.826		2	0.511
	3	0.823		3	0.508
	4	0.821		4	0.512
	5	0.830		5	0.510
Statistical Analysis	Mean	0.826	Statistical Analysis	Mean	0.510
	SD	0.004		SD	0.002
	% RSD	0.49		% RSD	0.31

(b) Method Precision:

Table 4: Data of Repeatability (Method precision) of Nimesulide

	Injection	Nimesulide Absorbance	% Assay
100 ppm Concentration	1	0.531	104.50
	2	0.542	106.62
	3	0.536	105.44
	4	0.535	104.95
	5	0.527	103.30
	6	0.541	106.39
Statistical Analysis	Mean	0.535	105.20
	SD	0.006	1.237
	% RSD	1.07	1.18

Table 5: Data of Repeatability (Method precision) of Racemethionie

	Injection	Racemethionin Absorbance	% Assay
750 ppm Concentration	1	0.837	102.11
	2	0.840	102.44
	3	0.839	102.31

	4	0.817	99.35
	5	0.822	99.88
	6	0.824	100.45
Statistical Analysis	Mean	0.830	101.09
	SD	0.010	1.359
	% RSD	1.20	1.34

Linearity:

The proposed given method was found linear for the range of 50- 375ppm and 100- 750ppm with coefficient of correlation i.e. R^2 is 1 and 1 for Nimesulide drug and Racemethionine drug

Figure 2: Linearity Plot (ConcentrationVsResponse) of Nimesulide

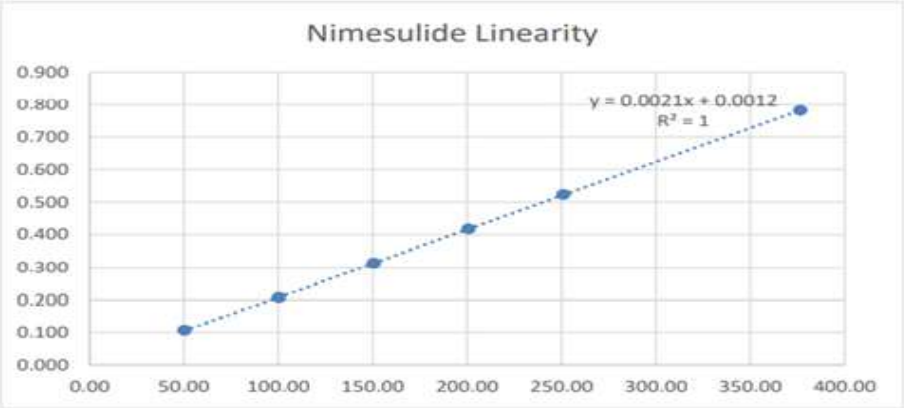

Figure 3: Linearity Plot (ConcentrationVsResponse) of Racemethionine

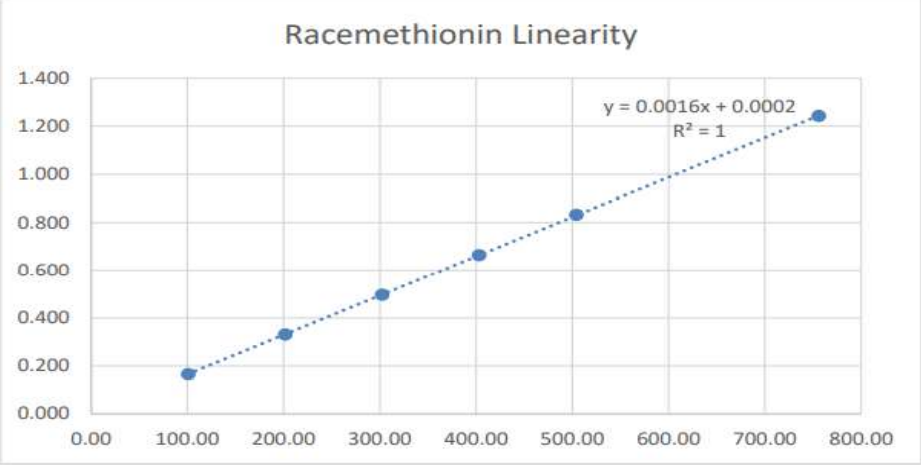

Table 6: Linear regression parameters for Nimesulide and Racemethionine drug by both proposed methods

Sr. no.	Parameters	Nimesulide	Racemethionine
1	Wavelength (nm)	228	226
2	Range of Calibration (ppm)	50-375	100-750
3	Coefficient of Correlation (R^2)	1.0	1.0
4	Slope (m)	0.0021	0.0016
5	Intercept (c)	0.0012	0.0002

Accuracy:

The recovery of added std. 50 percent, 100 percent & 150 percent was found at 3 attentiveness & 2 duplicate levels. The mean value for statically analysis of % recovery was found satisfactory in range from 100.40 to 100.18 % for

Nimesulide drug and 100.40 to 100.18 % for drug of Racemethionine..

Table 7: Data of Accuracy for Nimesulide

Concentration % of spiked level	Absorbance	Amount added (ppm)	Amount found (ppm)	% Recovery	Statistical Analysis of % Recovery (Mean)
50 % Sample 1	1.018	500	501	100.40	**100.40**
50 % Sample 2	1.024	500	501	100.92	
100 % Sample 1	1.530	750	752	100.51	**100.40**
100 % Sample 2	1.529	750	752	100.40	
150 % Sample 1	2.032	1000	1002	100.18	**100.18**
150 % Sample 2	2.047	1000	1002	100.82	

Table 8: Data of Accuracy for Racemethionine

Concentration % of spiked level	Absorbance	Amount added (ppm)	Amount found (ppm)	% Recovery	Statistical Analysis of % Recovery (Mean)
50 % Sample 1	1.018	750	750	100.40	**100.40**
50 % Sample 2	1.024	750	751	100.92	
100 % Sample 1	1.530	1000	1002	100.51	**100.40**
100 % Sample 2	1.529	1000	1002	100.40	
150 % Sample 1	2.034	1250	1253	100.18	**100.18**
150 % Sample 2	2.047	1250	1252	100.50	

Robustness

Table 9: Data For Effect of changing preparation method

	Injection	Racemethionin Absorbance		Injection	Nimesulide Absorbance
500 ppm Concentration	1	0.820	250 ppm Concentration	1	0.539
	2	0.819		2	0.542
	3	0.823		3	0.540
	4	0.820		4	0.543
	5	0.822		5	0.541

Statistical Analysis	Mean	0.821	Statistical Analysis	Mean	0.541
	SD	0.002		SD	0.002
	% RSD	0.20		% RSD	0.29

CONCLUSION

The proposed method of analytical was outlined as per guideline Q2 (R1) of ICH and found that it meets to acceptance criteria. In this executed work new method for UV- Spectrophotometric has developed, successfully, validated for 1st time & the current results of developed study indicate that the given method is rapid, simple, precise, stable and accurate. The developed absorbance ratio (Q-point) method was found appropriate for drug determination so this study can be easily applied in QC laboratory tests in the dosage form.

ACKNOWLEDGEMENT

Rajgad Dnyanpeeth college of Pharmacy

FUNDING SUPPORT
No

CONFLICT OF INTEREST
No

REFERENCES

[1]. Renjitha J R., *et.al*.Simultaneous method development and validation of Nimesulide and camylofin dihydrochloride in tablet dosage form by uv spectrophotometric method, Asian J. Pharm. Hea. Sci. Pharm. Hea. Sci. 2018,8 (3), 1944-1952

[2]. Mr. Jignesh Patel, *et.al.*, Development and Validation of RP-HPLC Method for Simultaneous Estimation of Nimesulide and Diclofenac Sodium in tablet Dosage Form, (IJRMP), 2017,6(5),69-76.

[3]. Prasad R. K., *et.al*. Spectrophotometric Quantitative Estimation and Validation of Nimesulide and Drotaverine Hydrochloride in Tablet Dosage form IJPSDR , 2010,2, (1),67-70

[4]. Shinde Prashanti,et al.. Simultaneous Estimation of DL-Methionine and Pyridoxine Hydrochloride in Tablet Dosage Form by RP-HPLC,IJPS,2014,3(1),105-109.

[5]. Prasanna Reddy., *et.al.* Simultaneous Estimation of Nimesulide and Chlorzoxazone in Pharmaceutical Formulations by a RP-HPLC Method, Int.J. ChemTech Res.2009,1(2), 283-285.

[6]. N.Mamatha., *et.al.* Sensitive and validated u.v.spectrophotometric methods for the estimation of Nimesulide in pharmaceutical and bulk formulations, WJPR.2014,3 (3), 4241-4247.

[7]. Rupali Kirtawade., *et.al.* Simultaneous UV Spectrophotometric Method for Estimation of Paracetamol and Nimesulide in Tablet Dosage Form, Int.J. ChemTech Res.2010,2(2),818-821

[8]. Vilas Arsul., *et.al.* Method Development and Validation of Cetirizine hydrochloride, Phenylephrine hydrochloride and Nimesulide by UV and HPLC, Asian Journal of Pharmaceutical Technology & Innovation, 2016,04 (19); 95 - 109

[9]. Dr. Rashmi Kumari., *et.al.* analytical method and validation for simultaneous estimation of drotavarine hydrochlorideand nimasulide as api in synthetic mixture by rp-hplc method and uv-visible spectroscopy,WJPR.2018, 7(2), 982-992.