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Q-absorbance ratio spectrophotometric method for the simultaneous estimation of Nimesulide and Chlorzoxazone in dosage form

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ABSTRACT

The present work carries an analytical method development of Nimesulide and Chlorzoxazone in tablet dosage form. The method is based upon Q-absorption ratio method for the simultaneous determination of Nimesulide and Chlorzoxazone. Absorption ratio method is used at two selected wavelengths, one of which is the iso-absorptive point and other being the λmax of other components. Nimesulide and Chlorzoxazone show their iso-absorptive point at 231nm in 0.1M NaOH solution. The second wavelength used is 244 nm which is the λmax of Chlorzoxazone in 0.1M NaOH solution. The linearity was obtained in the concentration range of 3-15 ppm for Nimesulide and Chlorzoxazone. The percentage RSD for precision and accuracy of method was found to be less than 2 %. This method was applied to all pharmaceutical dosage form because there is no excipients interference between them. The results have been validated by recovery studies.

KEYWORDS

Nimesulide;
Chlorzoxazone;
Absorption ratio method;
Validation.

INTRODUCTION

Nimesulide is N-(4-Nitro-2-phenoxyphenyl)-methane sulfonamide. Nimesulide is a relatively COX-2 selective, non-steroidal anti-inflammatory drug (NSAID) with analgesic and antipyretic properties. Its approved indications are the treatment of acute pain, the symptomatic treatment of osteoarthritis and primary dysmenorrhea in adolescents and adults above 12 years old[1]. It is a potent selective Cyclooxygenase-2 inhibitor and is highly effective, with minimum drug-related side effects, in the treatment of various forms of pain and inflammatory conditions. The drug is best with the disadvantage of poor water solubility. It is not official in USP 23 (1995), BP (1998), and IP (1996)[2,3]. Nimesulide is yellowish crystalline powder. It ispractically insoluble in water, freely soluble in acetone, slightly soluble in anhydrous ethanol. Melting point of Nimesulide is 149^0C[4].

Chlorzoxazone is chemically: (5-chloro-2(3H)-benzoxazolone). Molecular formula: $C_7H_4ClNO_2$; Molecular weight: 169.57[5,6]. It is a compound with skeletal muscle relaxant property and used to decrease muscle tone and tension, thus to relieve spasm and pain associated with musculoskeletal disorders[7-10].

Ultraviolet and Visible Spectro-photometry is one of the most frequently employed analytical tools in the pharmaceutical industry. Ultraviolet and visible absorption Spectro-photometry involves the measurement of absorption of monochromatic radiation by solutions of

chemical substances, in the range of 200 nm to 400 nm, and 400 nm to 800 nmof the spectrum, respectively[11]. The amount of absorption depends on the wavelength of radiation and the structure of compound. The absorption of radiation is due to the subtraction of energy from the radiation beam when electrons in orbital of lower energy are excited into orbital of higher energy. The various spectro-photometric methods which are used for estimation of drug in combine dosage form include;

- Simultaneous equation method
- Absorption ratio method (Q-ratio method)
- Geometric correction method
- Orthogonal polynomial method
- Difference spectro-photometry
- Derivative spectro-photometry
- Chemical derivatisation method
- Absorption correction method
- Multi-component method of analysis[12-14].

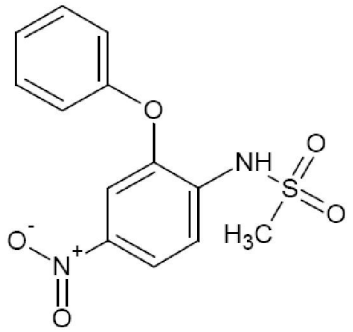

Figure 1 : Nimesulide

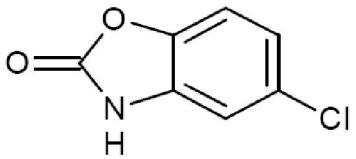

Figure 2 : Chlorzoxazone

Q-absorption ratio method

According to Q-Absorption ratio method, use the ratio of absorption at two selected wavelengths. One is the iso-absorptive point and other being the λmax of other component. Two equations were constructed as described below,

For Nimesulide, $C1 = \frac{Q0 - Q2}{Q1 - Q2} \times \frac{A}{a1}$

For Chlorzoxazone, $C2 = \frac{Q0 - Q1}{Q2 - Q1} \times \frac{A}{a2}$

$$Q0 = \frac{\text{Absorbance of sample at 244}}{\text{Absorbance of sample at 231}}$$

$$Q1 = \frac{\text{Absorptivity of nimesulide at 244}}{\text{Absorptivity of nimesulide at 231}}$$

$$Q2 = \frac{\text{Absorptivity of Chlorzoxazone at 244}}{\text{Absorptivity of Chlorzoxazone at 231}}$$

A= absorbance of sample at iso-absorptive point; Where, a1 and a2 are absorptivity of Nimesulide and Chlorzoxazone respectively at iso-absorptivity point.

MATERIALS AND METHODS

Working standard of Nimesulide and Chlorzoxazone was obtained from well reputed research laboratories. All other chemicals and reagents used were of analytical grade.

Instrument and conditions

A double beam Shimadzu UV-1700 series spectrophotometer was used. Absorption and overlain spectra of both test and standard solutions were recorded over the wavelength range of 200 400nm using 1cm quartz cell at fast scanned speed and fixed slit width of 1.0 nm. All weighing of ingredients were done on Agilent digital weighing balance.

Preparation of standard stock solution

A 10mg amount Nimesulide and Chlorzoxazone of reference substance was accurately weighed and dissolved in 100ml 0.1M NaOH solution in a 100 ml volumetric flask to obtain 100 ppm concentration of stock solution. From stock solution by the serial dilution we prepared required concentrations of 3, 6, 9, 12, 15 ppm for Nimesulide and Chlorzoxazoneseparately. The scanning for solution of Nimesulide and Chlorzoxazone were carried out in the range of 200-400 nm against 0.1M NaOH solution as a blank for obtaining the individual absorption spectra well as overlain spectra that were used in the analysis.

The maximum absorption (λmax) of Chlorzoxazone was found at 244 nm and iso-absorptive point at 231 nm. Absorption and absorptivity for a series of standard solutions were recorded atselected wavelengths.

Methodology

Absorption ratio method uses the ratio of absorptions of two selected wavelength, one of which is iso-absorptive point and other being the λmax of one of the two components. From the overlain spectra of two drugs (as shown in figure 6), it shows that Nimesulide and Chlorzoxazone having iso-absorptive point at 231 nm. The second wavelength used is 244 nm, which is the λmax of Chlorzoxazone. Working standard solutions having concentration 3, 6, 9, 12, 15 μg/ml for Nimesulide and Chlorzoxazone were prepared in 0.1M NaOH and the absorbance at 231 nm (iso-absorptive point) and 244 nm (λmax of Chlorzoxazone) were measured and absorptivity coefficient were calculated using calibrations curve.

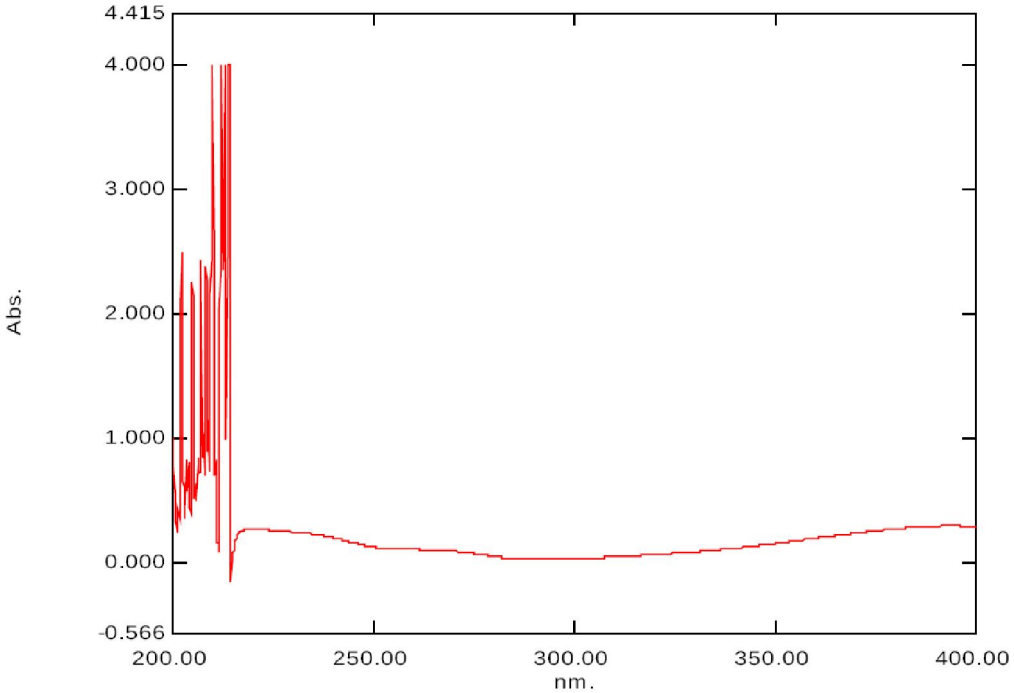

Figure 3 : Spectra of Nimesulide (6ppm)

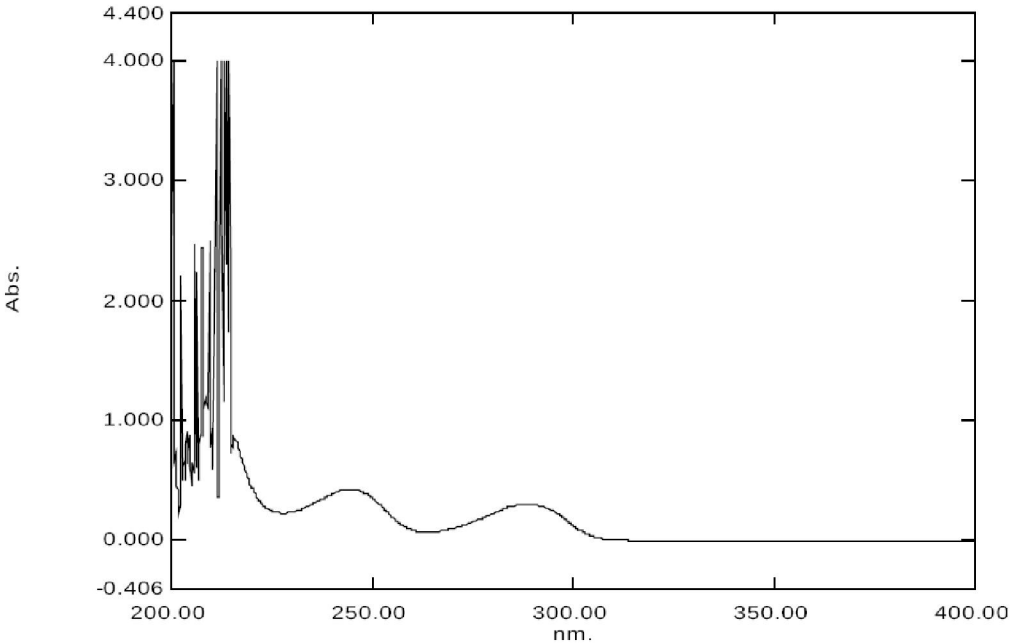

Figure 4 : Spectra of Chlorzoxazone (6ppm)

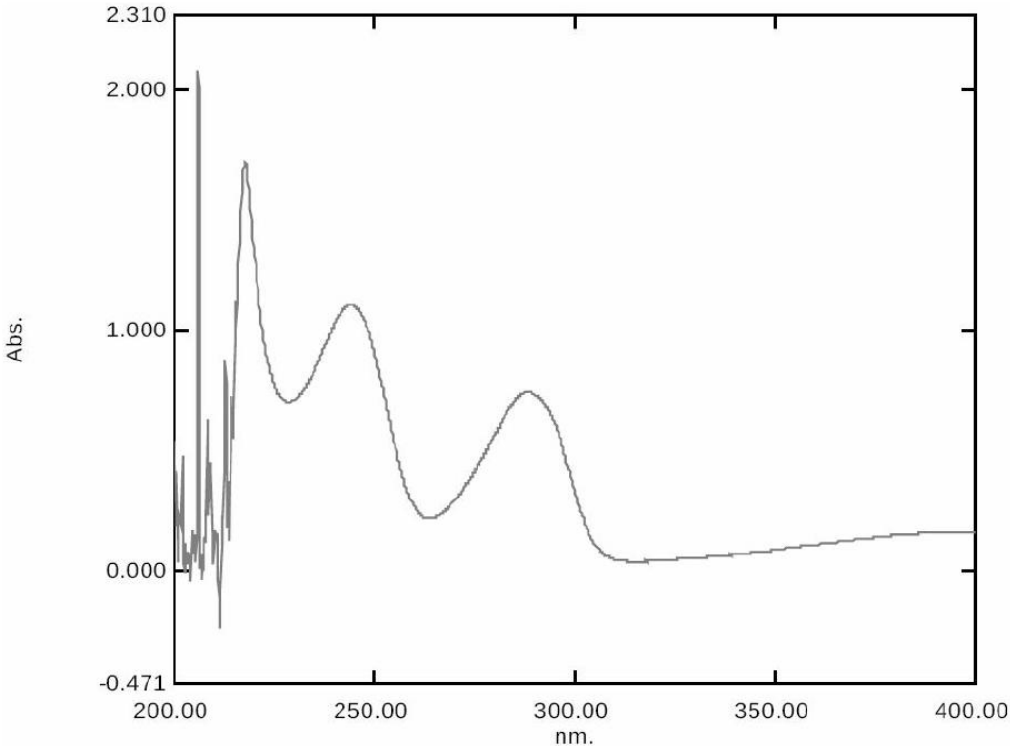

Figure 5 : Spectra of ohysical mixture (3:15)

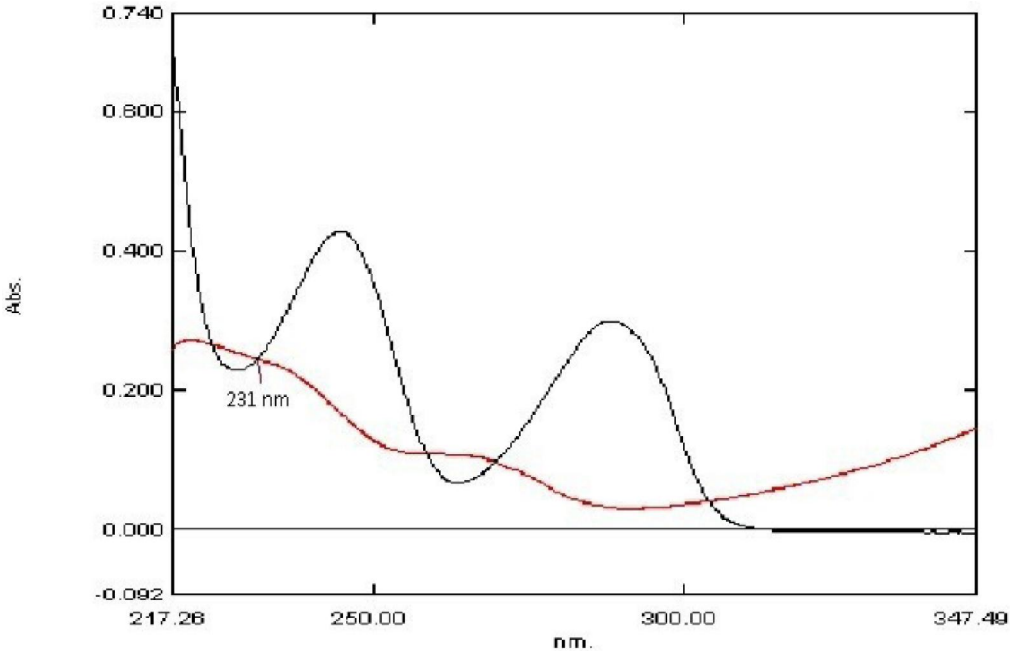

Figure 6 : Overlay spectra of Nimesulide and Chlorzoxazone

Validation of proposed method

(A) Linearity

Linearity was evaluated by preparing different concentration in the range of 3-15 µg/ml for both the drugs and absorption was measured. Each measurement was carried out in triplicate.

TABLE 1 : Linearity of Chlorzoxazone

Concentration (ppm)	Absorption
3	0.255
6	0.376
9	0.468
12	0.581
15	0.687

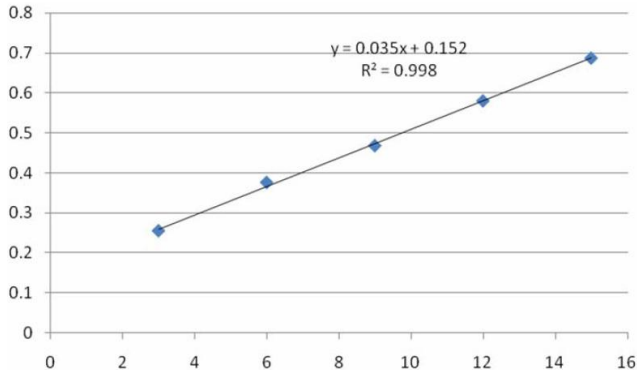

Figure 7 : Linearity graph of Chlorzoxazone

TABLE 2 : Linearity of Nimesulide

Concentration (ppm)	Absorption
3	0.171
6	0.282
9	0.405
12	0.531
15	0.652

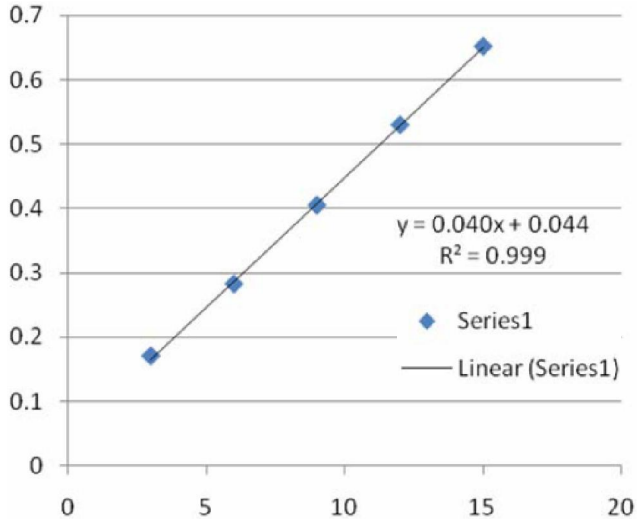

Figure 8 : Linearity of Nimesulide

(B) Precision

The precision of an analytical method is the closeness of replicate results obtained from analysis of the same homogeneous sample. To study precision, Three replicate standard solutions of Nimesulide and Chlorzoxazone were prepared and analyzed using the proposed method. The percent relative standard deviation (% RSD) for peak responses was calculated and it was found to be which is well within the acceptance criteria of not more than 2.0%. Results of system precision studies are shown in TABLE 3.

TABLE 3 : Precision data

Drug	Amount taken (µg/ml)	Intra day Amount found [n=3]		Inter day Amount found [n=3]	
		Mean	% RSD	Mean	% RSD
Nimesulide	3	2.96	1.54	2.97	1.56
	6	5.97	0.51	5.96	0.56
	9	8.91	0.40	9.17	0.61
Chlorzoxazone	3	3.06	0.69	3.10	1.25
	6	5.98	0.33	5.89	0.65
	9	8.98	0.35	8.91	0.86

(C) Accuracy (Recovery studies)

In order to check the accuracy, reproducibility and precision of the proposed method, recovery study was carried out by taking standard mixture solution of both Chlorzoxazone and Nimesulide (as shown in TABLE 4 and 5).

TABLE 4 : Accuracy table for Nimesulide

Sr. No.	Conc. taken in (µg/ml)	Std addition in (µg/ml)	Total conc. found (µg/ml)	% recover ± SD
1	6	4.8	10.81	100.26 ± 0.01
2	6	6	12.01	100.02 ±0.03
3	6	7.2	13.3	101.9 ± 0.008

TABLE 5 : Accuracy table for Chlorzoxazone

Sr. No.	Conc. taken in (µg/ml)	Std addition in (µg/ml)	Total conc. found (µg/ml)	% recover ± SD
1	6	4.8	10.79	99.87 ± 0.04
2	6	6	12.04	100.7 ± 0.01
3	6	7.2	13.3	101.6 ± 0.006

(D) Limit of detection (LOD) and limit of quantization (LOQ)

The LOD and LOQ of Nimesulide and Chlorzoxazone by proposed methodswere determined using calibration standards. LOD and LOQ were calculated as 3.3s/S and 10s/Srespectively, where S is the slope of the calibration curve and s is the standard deviation of response. The results of the same are shown in TABLE 6.

TABLE 6 : Limit of detection and limit of quantification

Parameter	Measured Value	
	Nimesulide	Chlorzoxazone
Limit of Quantification	0.96	1.12
Limit of Detection	0.29	0.37

(E) Tablet assay

The content of 20 Tablet were accurately weight and crushed into fine powder. A quantity of powder equivalent to 100 mg of Nimesulide and 500mg of Chlorzoxazone was transfer in to 100ml volumetric Flask. Dissolve in 100 ml 0.1M NaOH. Dilution are prepare and give the reading in 244 and 231 (iso-absorptive point) λmax. Calculate the equation. Result given in TABLE 7.

TABLE 7 : Analysis of tablet formulation

Formulation	Labeled amount (mg)	Amount recovered (mg) n=5	% Drug recovered	% RSD
Nise-Mr	Nimesulide 100mg	102.95	102.95	1.64
	Chlorzoxazone 500mg	529.40	105.88	0.94

RESULTS AND DISCUSSION

In absorbance ratio method (Q-analysis), the primary requirement for developing a method for analysis is that the entire spectra should follow the Beer's law at all the wavelength, which was fulfilled in case of both these drugs. The two wavelengths were used for the analysis of the drugs were 231 nm (iso-absorptive point) and 244 nm (λmax of Chlorzoxazone) at which the calibration curves were prepared for both the drugs. The overlain UV absorption spectra of Nimesulide (223 nm) and Chlorzoxazone (244 nm) shows iso-absorptive point (231 nm) in 0.1M NaOH.

CONCLUSION

The proposed absorption ratio method was found to be simple, sensitive and accurate method for determination of Nimesulide and Chlorzoxazone in tablet dosage form. In this method the solvent used is easily available and cheap for the analysis of Nimesulide and Chlorzoxazone hence, this method is economic for estimation of tablet dosage form. The common excipients and other additives were used in the formulation doesn't interfere in the analysis of the dosage form. So, we can say that this method can be adopted for combined dosage form in any pharmaceutical dosage form.

REFERENCES

[1] M.J.Oneil, A.Smith, P.E.Heckelman; The Merck Index, White House Station, NJ, USA, 6576 **(2001)**.
[2] F.P.Roberto, S.E.Evarshirman; Rev.Bras.Farm, **76**, 30 **(1995)**.
[3] J.R.Sadhana, R.Gitanjali; Eastern Pharm., **7**, 113 **(1997)**.
[4] British pharmacopoeia 2009, **2**, 1454.
[5] The Merck Index 2001, **13**, 379-380.
[6] L.C.Bailey; Remington, The Science and Practice of Pharmacy, 19th Edition, Mack Publishing Company, Pennsylvania, 1033 **(1995)**.
[7] S.Budavari, J.O.Maryadele, N.A.Smith, E.Patricia, J.F.Heckelman; The Merck Index, Merck Research Laboratories, 13 **(2001)**.
[8] L.George; HPLC methods for pharmaceutical analysis, **2(5)**, 367 **(1999)**.
[9] S.S.Yadav, A.S.Jagtap, J.R.Rao; Scholars research library, Der PharmaChemica, **4(5)**, 1798-1802 **(2012)**.
[10] R.Sharma, P.Gaikwad, R.Joshi; Scholars research library, Der PharmaChemica, **2(3)**, 90-99 **(2010)**.
[11] Indian Pharmacopoeia, Published by Controller of publications, New Delhi, **1**, 112-114 **(2007)**.
[12] A.H.Beckett, J.B.Stenlake; Practical Pharmaceutical Chemistry, Part One. Published by CBS Publishers and Distributors, New Delhi, India, **4**, 133-136 **(2005)**.
[13] A.H.Beckett, J.B.Stenlake; Practical Pharmaceutical Chemistry, Part Two, CBS Published by Publishers and Distributors, New Delhi, India, **4**, 275-306 **(2005)**.
[14] V.P.Nagulwar, K.P.Bhusari; Scholars research library, Der Pharmacia Lettre, **2(1)**, 196-200 **(2010)**.