Sensitive Spectrophotometric Method for the Determination of Niguldipine in Pharmaceutical Formulations

Chemistry

KEYWORDS : Niguldipine, Methyl orange (MO), Bromothymol blue (BTB), Spectrophotometer, Bulk drugs and tablet formulations

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ABSTRACT

Simple, sensitive and rapid spectrophotometric methods were developed for the determination of Niguldipine in pharmaceutical formulations. Method A was based on the extractive spectrophotometry and the developed color complex due to the formation ion association complex with Bromothymol blue (BTB) at pH 3.7. The formation of a yellow color solution exhibited maximum absorption at wavelength 380 nm. Method B was based on the yellow color formation due to ion association complex between methyl orange (MO) and Niguldipine at PH 3.7 that exhibited maximum absorption at a wave length of 430 nm. Both ion association complexes were extracted into chloroform and Beer's law was obeyed over the concentration ranges 4-25 µgml-1 and 2-12 µgml-1 for methods A and B respectively. Both the methods have been successfully applied for the assay of the drug in pharmaceutical formulations. No interference was observed from common pharmaceutical adjuvants. The reliability and the performance of the proposed methods were established by point and interval hypothesis tests through recovery studies.

INTRODUCTION

Niguldipine, (*R*)-1,4-dihydro-2,6-dimethyl-4-(3-nitrophenyl)-3,5-pyridine di carboxylic acid, 3-(4,4-diphenyl-1-piperidinyl) propyl methyl ester. It is a calcium channel blocker and A_1-adrenergic receptor antagonist (1)**.** The chemical structure of Niguldipine is as shown in **Figure 1.**

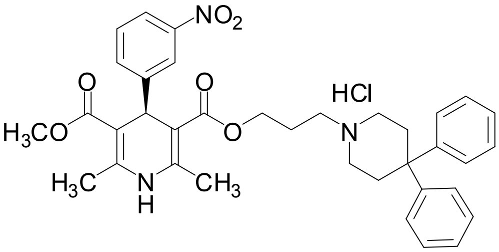

Figure 1. Structure of Niguldipine drug.

Extractive spectrophotometric procedures are more popular in the assay of drugs due to for their high sensitivity. Therefore, ion-pair extractive spectrophotometry has been received considerable attention for quantitative determination of many pharmaceutical compounds. To the best of our knowledge, in the determination of Niguldipine, a few analytical methods like Stripping Voltametric(2) and High performance liquid Chromatography(3) studies have been reported in the literature and no reports were found in the usage of simple spectrophotometric methods. The methods described here are simple and sensitive and has been used for the routine quality control analysis of pharmaceutical formulations containing Niguldipine which involved in the formation of ion association complex with acidic dyes, bromothymol blue (BTB) which is extractable into chloroform and showed absorption at 380 nm (method A), and methyl orange (MO) formed a yellow color solution that exhibited absorption at 430 nm (Method B).

EXPERIMENTAL

Instrumentation

Spectral and absorbance measurements were made with Shimadzu UV/Visible double beam spectrophotometer (model 2450).

Reagents

All the reagents were used of analytical reagent grade. The solutions, bromothymol blue (BTB) (0.2%) for Method-A and methyl orange (MO) (0.1%) for Method-B were prepared daily.

Standard and sample solutions of Niguldipine

A standard stock solution containing 1 mg/mL was prepared by dissolving accurately, 100 mg of drug in 20 mL of methanol. After dissolved the Tablet completely, it was treated with 10 mL of 5.0 N HCl and 4 gm of zinc dust. The working standard solution containing 100 µg/mL was prepared for both A and B methods dilution with methanol.

Assay procedure

Method-A

Aliquots of standard and reduced Niguldipine (1mL = 100 µg/mL) solutions ranging from 0.1-0.5 mL of 100 µg were transferred into a series of 50 mL of separating funnels and then 2 mL of BTB (0.2%) was added to this solution. The total volume of the aqueous phase was made up to 10 mL with distilled water. Approximately, 10 mL of chloroform was added to each funnel, and the contents were shaken for 2 min. and allowed to stand result separate the phases. The absorbance of chloroform layer was measured at 380 nm against the corresponding reagent blank. The amount of Niguldipine present in the sample solution was calculated from its calibration curve **(Figure 2)**.

Method-B

The standard and reduced Niguldipine (1 mL = 100 µg/mL) solutions from 0.5-2.5 mL were taken into a series of 50 mL of separating funnels and then added 2 mL of MO (0.1%). The total volume of the aqueous phase was made up to 10 mL with distilled water. 10 mL of chloroform was added to each funnel, and the contents were shaken for 2 min. and then allowed to stand result separate of the two phases. The absorbance of the chloroform layer was measured at 430 nm using corresponding reagent blank, and the calibration curve was drawn **(Figure 4)**.

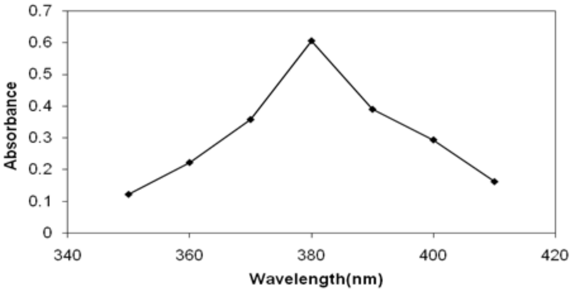

Figure 2. Absorption spectrum of Niguldipine with BTB/$CHCl_3$ system.

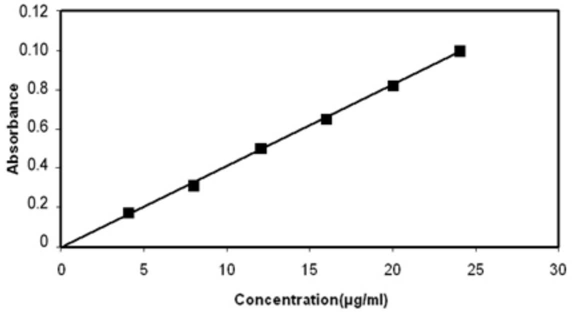

Figure 3. Beer's law plot of Niguldipine with BTB/$CHCl_3$ system.

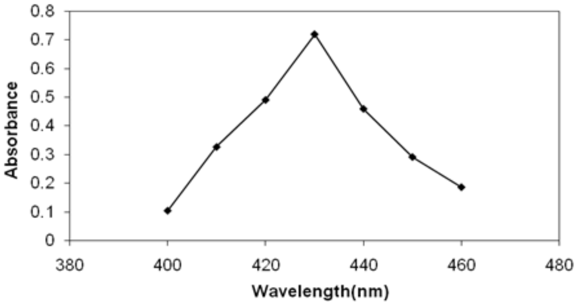

Figure 4. Absorption spectrum of Niguldipine with MO/$CHCl_3$ system.

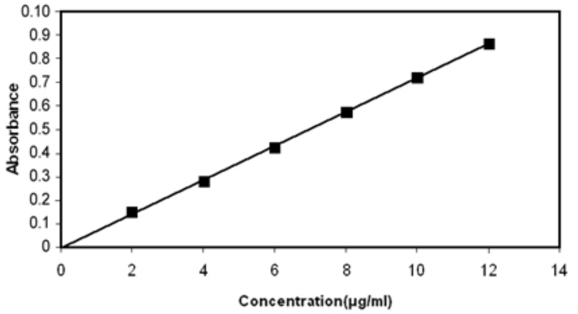

Figure 5. Beer's law plot of Niguldipine with MO/$CHCl_3$ system.

Result and Discussion

Anionic dyes like BTB, MO form ion-association complexes with the positively charged drugs. The drug and dye stoichiometric ratio was calculated by the continuous variation and mole-ratio method and found to be 1:1 both with BTB and MO. The drug dye complex with two positively charged ions behaves as a single unit held together by an electrostatic force of attraction. The possible mechanism for the formation of ion-association complex between drug, and the reagents, BTB and MO were depicted in **Figure 6** and **Figure 7** respectively.

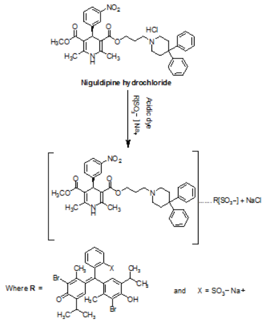

Figure 6. Ion-association complex formation of Niguldipine hydrochloride with BTB

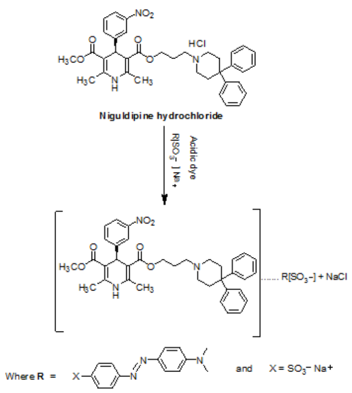

Figure 7. Ion-association complex formation of Niguldipine hydrochloride with Methyl Orange

Optimization of variables

Optimum conditions are necessary for rapid and quantitative formation of colored ion-pair complexes with maximum stability and sensitivity which was established in preliminary experiments. Chloroform was preferred as better solvent for these methods for its quantitative extraction selectively. Optimum conditions were fixed by varying one parameter at a time while keeping other parameters constant and observing its effect on the absorbance at 380 nm for BTB and 430 nm for MO. The optical characteristics such as molar absorptivity, Beer's law range and Sandell's sensitivity are presented in **Table 1**.The regression analysis using the method of least squares was made for the slope (a), intercept (b) and correlation coefficient (r) obtained from different concentrations and the results are summarized in **Table 1**. The relative standard deviations and percent range of error (0.05 and 0.01) confidence limits were calculated for the eight measurements each. The experimental data revealed that method A is more sensitive than method B.

Table 1. Optical Characteristics, precision and accuracy of the proposed methods for Niguldipine.

Parameter	Method-A	Method-B
λmax (nm)	380	430
Beer's law Limits (µg/mL)	4-25	2-12
Molar absorptivity (1 $mole^{-1}$ cm^{-1})	6 x 10^4	6.87 x 10^4
Sandell's sensitivity (µg/cm^2/0.001 absorbance unit)	0.0126	0.01443
Slope (b)	0.0134	0.0446
Intercept (a)	0.018	0.0028
Correlation coefficient (r)	0.9832	0.04472
Standard deviation	2.7285	0.1694
% of Relative standard deviation	0.92	0.79
0.05 level	±0.462	±0.741
0.01 level	±1.14	±0.954

Assay procedures

Aliquots of standard Niguldipine ranging from 0.1-0.5 mL were transferred into a series of 50 mL separating funnels. 2 mL of BTB (0.2%) and MO (0.2%) were added to the aliquots solution and the total volume of the aqueous phase was made up to 10 mL with distilled water. Chloroform (10 mL) was added in three initial amounts to each funnel and the contents were shaken for two minutes. The contents were allowed to stand until two phases are separate clearly and the absorbance of the chloroform layer was measured at 380 nm and 430 nm against the reagent blank. Satisfactory results were obtained for drug analysis in pharmaceutical formulations and the results were reproducible with low R.S.D. values. The average percent recoveries obtained were quantitative, indicating good accuracy of these methods. The results of analysis of the commercial tablets and the recovery studies of drug suggested that there is no interfere for many excipients (such as Strarch, Lactose, Titaniumdioxide, and Magnesium stearate) which are presented in **Table 2**.

Table 2. Assay and recovery of Niguldipine in dosage forms.

S.NO.	Pharmaceutical Formulation	Labelled amount(mg)	proposed method			Found by reference method ±S.D	% recovery by proposed methods ±S.D
			Amount found (mg)	t (Value)	F (Value)		
1	Tablet-1	10	9.97 ± 0.015	0.067	2.169	9.94 ± 0.016	100.2 ± 0.64
2	Tablet-2	10	9.96 ± 0.012	0.075	2.540	9.96 ± 0.091	99.41 ± 0.79
3	Tablet-3	10	9.93 ± 0.095	0.057	2.169	9.99 ± 0.019	100.1 ± 0.85
4	Tablet-4	10	9.91 ± 0.009	0.045	2.474	9.94 ± 0.018	100.9 ± 0.61
5	Tablet-5	10	10.04 ± 0.082	0.262	2.175	10.01 ± 0.00	9.82 ± 0.94
6	Tablet-6	10	10.01 ± 0.008	0.391	2.638	10.04 ± 0.07	99.92 ± 1.04

Conclusions

The proposed methods are simple and sensitive and reproducible for the determination of Niguldipine in any pharmaceutical preparations and did not suffer any interference due to common excipients of tablets like talk starch and magnesium stearate, lactose etc.

REFERENCE

1. Boer R, Grassegger A, schudt C, Glossmann H. Nigludipine binds with very high affinity to Ca2+ channels and to a subtype of alpha1-adrenoceptors. European journal of Pharmacology, Molecular Pharmacology. 1989, 172 (2), 131-45. | 2. Gottfried S, Dagmar O. Determination of trace level Niguldipine in urine and blood by adsoptive stripping voltametry at the hanging mercury, drop electrode. Institute for Analystiche Chemie and Radiochemie. Der Univesrtat Innsbruck, Austria. 1996, 121, 351-356. | 3. Xiao-fei Zhou, Qi Wang, Robert A. Coburn, Marilyn E. Morris., A high performance liquid Chromatographic method for determination of the Nigludipine analogue DHP-014. Journal of Biomed Chromatography, 2006, 20(1), 48-53. |