Academic Sciences International Journal of Pharmacy and Pharmaceutical Sciences

ISSN- 0975-1491 Vol 5, Issue 1, 2013

Research Article

# DEVELOPMENT AND VALIDATION OF METHOD FOR SIMULTANEOUS ESTIMATION OF PYRIDOXINE HYDROCHLORIDE AND DOXYLAMINE SUCCINATE IN TABLET DOSAGE FORM BY FIRST ORDER DERIVATIVE SPECTROSCOPY

**K.S.NATARAJ*[1], Y.SUVARNA[1] AND G. VENKATESWARI[1]**

[1]Shri Vishnu College of Pharmacy, Bhimavaram 534202, West Godavari District, Andhra Pradesh, India. Email: kalakondan@yahoo.com

*Received: 26 Nov 2012, Revised and Accepted: 21 Dec 2012*

**ABSTRACT**

Pyridoxine Hydrochloride and Doxylamine succinate are used in combination for treatment of Morning Sickness.

Objective: To develop a simple spectrophotometric method for simultaneous estimation of Pyridoxine Hydrochloride (PYR) and Doxylamine succinate (DOX) in tablet dosage form.

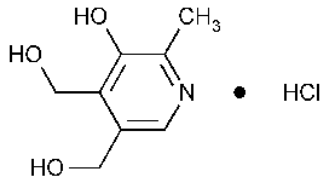
Method: The method employed is first order derivative spectroscopy. Results: For determination of sampling wavelength 10 µg/ml of each of PYR and DOX were scanned in 200-400 nm range and sampling wavelengths were 231.8 nm for PYR where DOX showed zero crossing point and 253 nm for DOX where PYR showed zero crossing point in first order derivative spectroscopy. For this method linearity observed in 1-40 µg/ml for PYR and 2.5-80 µg/ml for DOX. LOD of DOX and PYR were 0.2826 and 0.3571 µg/ml respectively. LOQ of DOX and PYR were 0.4621 and 0.5918 µg/ml respectively.

Discussion: The recovery studies confirmed accuracy of proposed method and low values of standard deviation confirmed precision of method. The method is validated as per ICH guidelines.

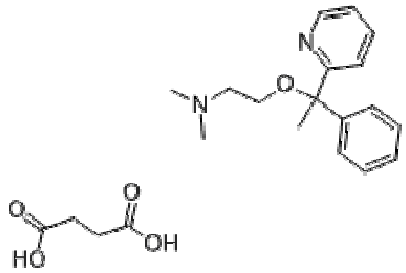
**Keywords:** Pyridoxine Hydrochloride, Doxylamine succinate, first order derivative spectroscopy.

## INTRODUCTION

Pyridoxine Hydrochloride is chemically described as 4,5-Bis(hydroxymethyl)-2-methylpyridine-3-ol Hydrochloride salt[1] (Vitamin $B_6$) assists in the balancing of sodium and potassium as well as promoting red blood cell production and Doxylamine succinate is N,N-Dimethyl-2-[α-methyl-α-2-pyridinyl) benzyloxy] ethylamine Hydrogen Succinate is antihistamine, both in combination used for the treatment of Morning Sickness. Many analytical methods like HPLC [2-3], UV Spectroscopy [4-5] and charge transfer complexation[6] methods were reported for determination of PYR and DOX combination. In this communication we report a new UV spectrophotometric method using derivative spectroscopy.




Pyridoxine Hydrochloride




Doxylamine Succinate

**Fig. 1: Structure of Pyridoxine Hydrochloride and Doxylamine succinate**

## MATERIALS AND METHODS

### Instrument

Spectrophotometric analysis was carried out on a double beam UV-spectrophotometer (UV Lab India) using a 1 cm quartz cell. The instrument settings were zero order and first derivative mode and band width of 1.0 nm in the range of 200–400 nm.

### Chemicals and reagents

The tablets of the said combination were purchased from a local pharmacy (The label claim contained 10mg of PYR and 10 mg of DOX). Distilled water was used as solvent to prepare all solutions.

### Procedure [7-8]

**Preparation of stock solution (1000µg/ml):** Accurately weighed quantity of pure PYR (10mg) and pure DOX (10mg) were transferred into two separate10mL volumetric flasks, dissolved in distilled water and made the volume to 10mL with the same solvent. The stock solution was sonicated for 2min.

**Preparation of working standard solution (100µg/ml):** From the above stock solution 1mL each of PYR and DOX was taken, transferred to separate 10mL volumetric flasks and the volume was made upto 10ml with distilled water.

### Method

In this method, 10µg/ml solution of standard drug PYR and DOX were prepared using distilled water as solvent and scanned from 400nm to 200nm (**Fig 2**). The absorption spectra thus obtained were derivatized from first order to fourth order. First order derivative spectra were selected for analysis of both drugs. From the overlain spectra of both the drugs (**Fig 3 ),** wavelength selected for quantification were 231.8 nm for PYR (zero cross of DOX) and 253

nm for DOX (zero cross of PYR). The calibration curve for PYR and DOX were plotted in the concentration range of 5-25µg/ml at 231.8nm and 253 nm respectively.

**Estimation of Tablet Dosage form**

For the estimation of PYR and DOX in commercial formulations, twenty tablets were weighed and average weight was calculated. The tablets were crushed and tablet powder and powder equivalent to 10 mg of PYR taken and added in 60 ml of Distilled water system sonicated for 10 min after sonication volume was made up to 100 ml. 1ml of this stock solution was diluted to 10 ml to get concentration equal to 10 µg/ml of PYR and 10 µg/ml of DOX. This solution is scanned in range 200-400 nm taking distilled water as blank. The spectrum obtained was converted to first order derivative spectra, absorbances were noted and concentrations were determined from regression equations generated from calibration graph.

**RESULTS AND DISCUSSION**

Sampling wavelengths were determined from scanning individual drug samples in 200-400 nm range(**Fig 2**). Sampling wavelengths were 231.8 nm and 253 nm for PYR and DOX respectively in first order derivative mode(**Fig 3**).

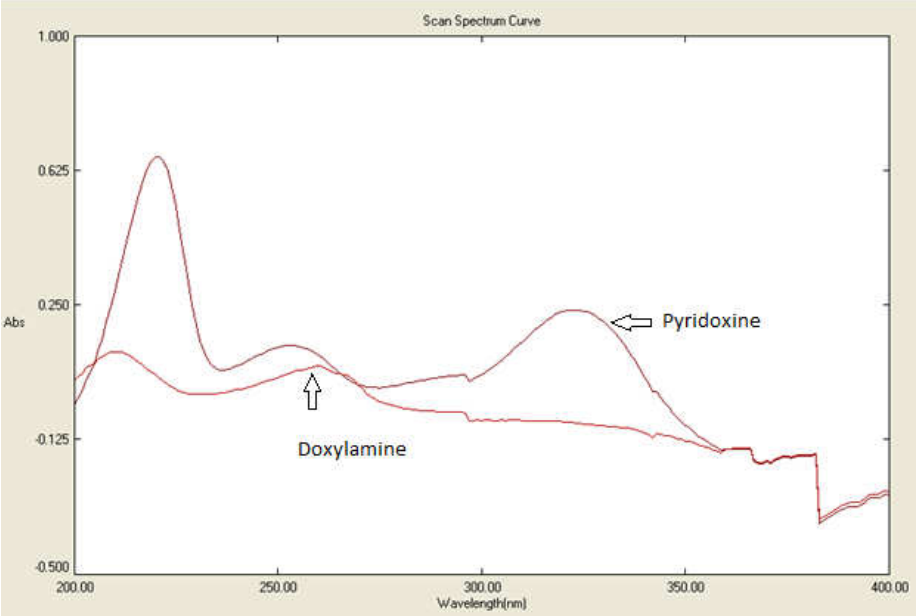



**Fig. 2: Zero order overlain spectrum of PYR and DOX**

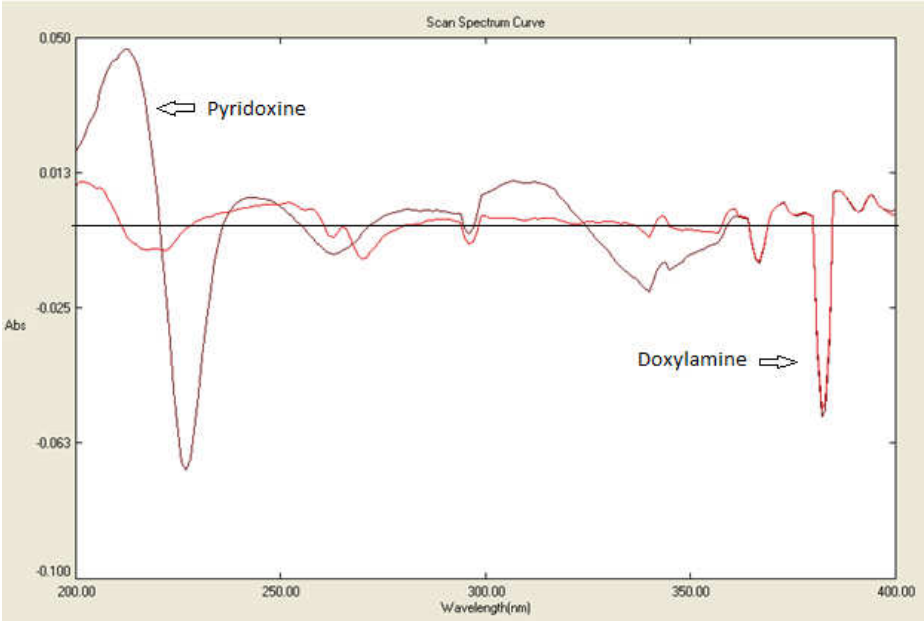



**Fig. 3: First order overlain absorption spectrum of PYR and DOX**

For this method equations generated were Y=0.004x - 0.013 (r2=0.985) and Y=-0.00036x + 0.0004 (0.987) for PYR and DOX respectively. Linearity of proposed method was found to be 1-40 µg/ml for PYR and 2.5-80 µg/ml for DOX. Limits of detection were found to be 0.2826 and 0.3571 µg/ml of PYR and DOX respectively. Limits of quantitation were found to be 0.4621 and 0.5918 µg/ml for PYR and DOX respectively. Results of tablet analysis were reported in table 1, result of precision studies and recovery study reported in table 2 and 3 respectively.

**Table 1: Results of assay of tablet dosage form**

| Analyte | Label claim (mg/tab) | Amount found* (mg/tab) | % label claim*± S.D. |
|---|---|---|---|
| PYR | 10 | 9.91 | 99.1%±0.873 |
| DOX | 10 | 10.14 | 101.4%±0.932 |

*Average of five determinations

**Table 2: Results of Precision studies**

| Analyte | Label claim (mg/tab) | Amount found* (mg/tab) | % label claim*± S.D. |
|---|---|---|---|
| PYR | 10 | 10.11 | 101.1%±0.962 |
| DOX | 10 | 10.18 | 101.8%±0.951 |

*Average of five determinations

**Table 3: Recovery studies data showing average amount of drug recovered from sample solution**

| Analyte | %Recovery (Mean±S.D*) | RSD* |
|---|---|---|
| PYR | 99.5±0.8532 | 0.8575 |
| DOX | 100.13±0.916 | 0.9148 |

## CONCLUSION

The method used is simple and rapid and does not involve the use of complex instrument, low value of Standard deviation showed that the method is precise and high percentage of recovery of as shown in table shows that the method is accurate.

## ACKNOWLEDGEMENTS

The authors are very much thankful to Svizera Health Care (ACE) for providing the standard drugs of Doxylamine and Pyridoxine. The authors are also thankful to Shri Vishnu College of Pharmacy, Bhimavaram for providing the necessary research facilities for carrying out the research work.

## REFERENCES

1. Concise Organic Pharmaceutical Chemistry by Dr.K.R.Mahadik, Nirali Prakashan publishers and distributors, Nov 2008, 13th edition page no. 22.10
2. A.P.Argekar, J.G.Sawanth .Simultaneous determination of Pyridoxine Hydrochloride and Doxylamine Succinate from tablets by ion pair RP-HPLC. Drug Development and Industrial pharmacy, August 1999, Volume 25, Issue 8, Pages 945-950.
3. Ashutosh pathak, Sadhana J Rajput. Simultaneous determination of ternary mixture of Pyridoxine Hydrochloride, Doxylamine Succinate and folic acid by a ratio spectra zero crossing, double divisor-ratio spectra derivative and column HPLC methods. Journal of AOAC International, Volume 91, Issue 5, Pages 1059-1069.
4. M. Abdel-Hady Elsayed, Saied F. Belal, Abdel-Fattah M. Elwalily, Hassan Abdine. Spectrophotometric determination of tolbutamide, thiamine hydrochloride and pyridoxine hydrochloride in combination products. Journal of Pharmaceutical Sciences, June 1979, Volume 68, Issue 6, Pages 739–741.
5. Simultaneous spectrometric determination of folic acid, pyridoxine riboflavin, and thiamine by partial least-squares regression. Analytical letters, 2002, Volume 35, Issue 10, Pages 1677-1691.
6. A.Raza, T.M.Ansari, S B niazi, A.Rehman. Spectrophotometric Determination of pyridoxine Hydrochloride (Vitamin B6) in Bulk and Tablets by Charge-Transfer Complexation with Chloranil. Journal of chemical society of Pakistan, 2007, Volume 29, Issue 1, Pages 33-36.
7. Manaswi Patil, Vrushali Tambe, Vijaya Vichare, Reshma Kolte Validated simultaneous UV Spectroscopic methods for estimation of Ciprofloxacin and Tinidazole in tablet dosage form, March 2012, Volume 4, Issue 3, Pages 182-185.
8. Gajjar Anuradha K.,Shah Vishal D. Simultaneous UV Spectrophotometric estimation of Rosuvastatin and Ezetimibe in their combined dosage forms, 2010, Volume 2, Issue 1, Pages 131-138.