Spectrophotometric Determination of Pyridoxine Hydrochloride (Vitamin B6) in Bulk and Tablets by Charge-Transfer Complexation with Chloranil

[1]A. RAZA, [1]T. M. ANSARI*, [1]S. B. NIAZI AND [2]A. REHMAN
[1]*Department of Chemistry, Bahauddin Zakariya University Multan, 60800, Pakistan.*
[2]*Analytical Laboratory, Bio Fine Pharmaceuticals (Pvt.) Ltd. Multan, Pakistan.*

(Received 15th August, 2006, revised 28th September, 2006)

Summary: A rapid, simple and sensitive spectrophotometric method has been developed for the determination of Pyridoxine Hydrochloride (Vitamin B6) in pure and tablet formulations. The method depends on the charge–transfer complexation between pyridoxine as n-electron donor in basic medium with chloranil as π-acceptor in acetonitrile medium to give a colored complex that absorbs maximally at 550 nm. Beer's law is obeyed in the concentration range 0.5-38 μg ml^{-1} with molar absorptivity of 1.9×10^3 L $mole^{-1}$ cm^{-1}. The proposed method is precise, accurate and specific for the quantitative determination of the drug in bulk and tablet formulations.

Introduction

Pyridoxine hydrochloride (3,4-pyridinedimethanol, 5-hydroxy-6-methyl-, hydrochloride) may be used prophylactically to prevent, or to treat, peripheral neuritis in patients treated, with isoniazid. It has been claimed that the vitamin controls the nausea and vomiting of pregnancy or of radiation sickness. It is medically effective in treating the carpal-tunnel syndrome [1].

Several analytical procedures are available for the determination of pyridoxine hydrochloride (individual and combined with other vitamins in pharmaceutical formulations) e.g., liquid chromatography [2-9] titrimetry [10], first derivative spectrofluorimetry [11-12], differential pulse voltammetry [13], gas chromatography [14], GC-MS [15], spectrophotometry [16-19], capillary electrophoresis [20], graphical and multivariate calibration-prediction method [21]. However, most of these methods are costly, tedious and time-consuming. Therefore, the need for a rapid, economical and selective method is obvious, especially for a routine quality control analysis of pharmaceutical products containing pyridoxine hydrochloride.

The aim of this study was to develop a fast, sensitive, economical and easy UV-Visible spectrophotometric method for the determination of pyridoxine hydrochloride in raw and pharmaceutical tablet formulations.

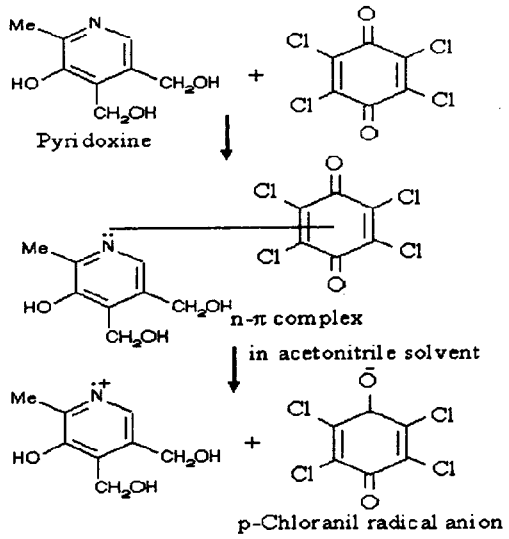
Results and Discussion

The charge-transfer (CT) reactions have been widely studied recently. Many drugs have been studied by spectrophotometric method based on color charge-transfer complexes formed with electron acceptors [22, 23]. Some hydrochloride salts of amines do not react with δ and π acceptors because they do not possess a lone pair of electrons. Similarly, pyridoxine hydrochloride does not react with chloranil owing to the non-availability of lone pair of electrons. In order to prepare the assay procedure of pyridoxine hydrochloride, it is necessary to convert pyridoxine hydrochloride solution into pyridoxine base solution. Some workers have reported the extraction method for the conversion of acidic drug into basic form [24, 25]. But these methods are time- consuming and tedious. In this study, an easy and fast method for the conversion of acidic drug into basic form was adopted.

In this method, pyridoxine hydrochloride was reacted with 2M sodium bicarbonate solution, which changed the solution to basic medium. The addition of chloranil solution to pyridoxine base solution (n-donor), resulted in the instantaneous formation of a charge transfer complex of the n-π type, which showed absorption maximum at 550 nm. The stoichiometric ratio of the pyridoxine base to chloranil was determined by Job's method [26] and was found to be 1: 1 (Fig. 1). This confirmed the presence of one donation center in the pyridoxine base.

A possible reaction between pyridoxine base and chloranil based on literature review is presented in the Scheme-1. The effect of the volume of chloranil solution was studied by taking 1 ml of 10

*To whom all correspondence should be addressed.



Scheme-I

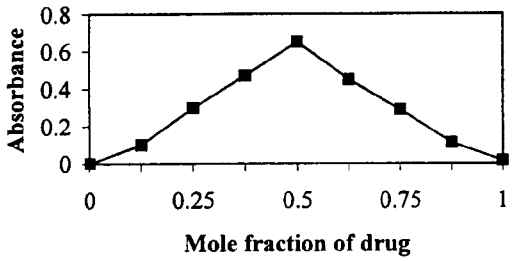


Fig. 1: Stoichiometric ratio of the complex.

μg ml^{-1} pyridoxine base to different volumes of 0.2 % chloranil solution. The reaction mixture was diluted to 10 ml with acetonitrile and the absorbance was recorded. The results showed that the highest absorbance was obtained with 2.5 ml of 0.2 % chloranil reagent (Fig. 2).

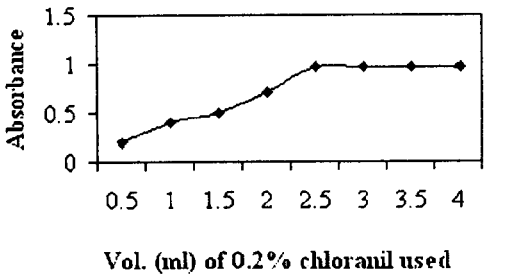


Fig. 2: Effect of color producing reagent concentration.

The effect of various excipients, frequently used in formulations, was evaluated using the proposed method. It was found that commonly used excipients do not interfere with the determination (Table-1). Under the optimum experimental conditions, absorbance responses the linear relation to the concentration of pyridoxine hydrochloride over the range 0.5-38 μg ml^{-1} (Fig. 3).

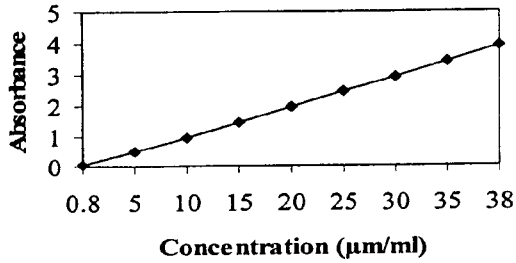


Table-1: Determination of pyridoxine hydrochloride in the presence of different excipients.

S. No.	Excipients	Amount taken (μg ml^{-1})	% Recovery ± RSD (n = 5)
1	Talc	50	99.7 ± 0.30
2	Microcrystalline cellulose	300	99.6 ± 0.25
3	Hydroxry propyl methyl cellulose	200	99.5 ± 0.25
4	Lactose	50	99.4 ± 0.45
5	Magnesium stearate	200	99.9 ± 0.25
6	Starch	50	99.1 ± 0.30

Table-2: Optical characteristics and statistical data for the regression equation of the proposed method.

Parameters	Values
λ_{max}	550 nm
Beer's law limit (μg ml^{-1})	0.8-38
Molar absorptivity (L mole^{-1} cm^{-1})	1.996×10^3
Sandell's sensitivity (μgml^{-1} per 0.001 A)	1.02×10^{-2}
Regression equation (Y*)	
Slope (b)	1.002×10^{-1}
Intercept (a)	3.7×10^{-2}
Correlation coefficient (r)	0.9987
RSD** (%)	0.852
Limit of detection (μg ml^{-1})	0.37
Limit of quantitation (μg ml^{-1})	1.25

Y* = a + bC

Where C is the concentration of analyte (μg ml^{-1}) and Y is absorbance unit.

** = Calculated from five determinations

Optical characteristics and statistical data for the regression equation of the proposed method are given in Table-2. Commercial formulations were successfully analyzed for pyridoxine hydrochloride by the proposed method and compared with the reference method [27]. Results are shown in Table-3. The calculated student's t-values and F-values did not

Table 3: Determination of pyridoxine hydrochloride formulations by the proposed and reference method

Formulation	Company	Proposed method Recovery* (%)	RSD (%)	Reference method [27] Recovery *(%)	RSD (%)	t-test	F-test	Significantly different?
Pyro-6	Medera, Islamabad	99.91	0.71	100.25	0.49	0.668	2.125	No
Vita-6	Mendoza, Karachi	100.05	0.66	101.14	0.37	0.412	3.021	No
V-Six	Zesion, Islamabad	99.86	0.68	100.09	0.47	0.872	2.194	No

*Average of 5 independent analyses

exceed the theoretical values that indicated the absence of any difference between the methods compared.

The proposed method showed good results for pyridoxine hydrochloride in raw and pharmaceutical tablet formulations. However, it cannot be applied for the estimation of pyridoxine hydrochloride in multi-vitamin pharmaceutical samples and clinical samples (i.e. blood and urine) because chloranil may also react with other vitamins and amines containing in the samples.

Experimental

Apparatus

A Hitachi UV-Visible spectrophotometer (model U 1100), with 1 cm silica cells, was used throughout this work.

Chemicals and Reagents

All the chemicals and reagents were of analytical reagent grade. Pure sample of Pyridoxine hydrochloride was provided by Bio Fine Pharmaceuticals (Pvt.) Ltd. Multan, Pakistan. Pyridoxine hydrochloride tablets were purchased from the local market. Double distilled water was used throughout this work.

A 100 μg ml^{-1} solution of pyridoxine hydrochloride was prepared by dissolving 100 mg of pure drug in least amount of water in 100 ml measuring flask and the volume was made up to mark by acetonitrile. A 0.2 % chloranil solution was freshly prepared by dissolving appropriate weight in acetonitrile. A 2M aqueous sodium bicarbonate solution was used.

Proposed Procedure

To an aliquot (0.5-38 μg ml^{-1}) of the drug solution in 10 ml measuring flask, 1 ml of 2M sodium bicarbonate solution was added and shaken for one minute, followed by the addition of 2.5 ml of chloranil solution to it. A colored product was produced instantaneously. The absorbance was measured after 5 minutes at 550 nm against reagent blank.

Procedure for the Determination of Pyridoxine Hydrochloride in Tablets

An accurately weighed portion of powdered tablets equivalent to 100 mg of pyridoxine hydrochloride was stirred well with 20 ml water. The residue was filtered on Whatman filter paper #1 and washed with acetonitrile. The filtrate and washings were diluted to 100 ml with acetonitrile using a measuring flask and then subjected to the proposed procedure for determination.

Conclusion

The proposed method is rapid, simple, economical and fairly sensitive. It can be used in routine analysis of pharmaceutical tablet formulations of pyridoxine hydrochloride in quality control laboratories. Moreover, the present method can be directly applied to the pharmaceutical sample without prior separation or treatment.

Acknowledgements

The authors are thankful to the Department of Chemistry, Bahauddin Zakariya University Multan, Pakistan and Bio Fine pharmaceuticals (Pvt.) Ltd. Multan, Pakistan for providing necessary research facilities to carry out this work.

References

1. A. R. Gennaro (Ed.), "*Remington, The Science and Practice of Pharmacy*", 19th edition, Mack Publishing Company, Easton, Pennsylvania (1995).
2. B. K. Ayi, D. A.Yuhas and N. J. Deangelis, *J. Assoc. Off. Anal. Chem.*, **69**, 56 (1986).
3. M. C. Walker, B. E. Carpenter and E. L. Cooper, *J. Pharm. Sci.*, **70**, 99 (1981).

4. E. Din, G. Kokdil, F. Onur, *J. Pharm. Biomed. Anal.*, **22**, 915 (2000).
5. A. El-Gindy, *J. Pharm. Biomed. Anal.*, **32**, 277 (2003).
6. P. F. Chatzimichalakis, V. F. Samanidou, R, Verpoorte and I. N. Papadoyannis, *J. Sep. Sci.*, **27**, 1181 (2004).
7. A. El-Gindy, S. Emara and A. Mostafa, *IL Farmaco*, **59**, 713 (2004).
8. K. Li, *Biomed. Chromatogr.*, **16**, 504 (2002).
9. F. L. Lam, I. J. Holcomb and S. A. Fusari, *J. Assoc. Off. Anal. Chem.*, **67**, 1007 (1984).
10. B. Jayaram and M. N. M. Gowda, *J. Assoc. Off. Anal. Chem.*, **69**, 47 (1986).
11. A. Molina-Diaz, N. Ramos-Martos, A. Navalon and L. F. C. Vallvey, *J. Assoc. Off Anal. Chem.*, **85**, 861 (2002).
12. H. H. Abdine, A. A. Gazy and M. H. Abdel-Hay, *J. Pharm. Biomed. Anal.*, **15**, 1113 (1997).
13. B. Uslu, B. T. Demircigil, S. A. Ozkan, Z. Senturk and H. Y. Aboul-Enein, *Pharmazie*, **56**, 938 (2001).
14. L. T. Sennello and C. Argoudelis, *J. Anal. Chem.*, **41**, 171(1969).
15. L. J. N. Vergara,. J. A. Squella,. J. C. Sturm, H. Baez and C. Camargo, *J. Pharm. Biomed. Anal.* **26**, 929 (2001).
16. D. Ozdemir and E. Dinc, *Chem. Pharm. Bull.*, **52**, 810 (2004).
17. S. C. Sharma, R. C.Saxena and S. K Talwar, *J. Pharm. Biomed. Anal.*, 7,321(1989).
18. M. A. Elsayed, S. F. Belal, A. M. Elwalily and H. Abdine, *J. Pharm. Sci.*, **68**,739 (1979).
19. J. P. Xu, B. R Xiang and D. K. An, *Yao Xue Xue Bao.*, **25**, 77 (1990).
20. L. Fotsing, M. Fillet, I. Bechet, P. Hubert and J. Crommen, *J. Pharm. Biomed .Anal.*, **15**, 1113 (1997).
21. R. D. Bautista, A. I. Jimenez, F. Jimenez and J. Arias, *J. Pharm. Biomed. Anal.*, **15**, 183 (1996).
22. Z. A. El-Shreif, A. O. Mahamed, M. I. Walash and F. M. Tarras, *J. Pharm. Biomed. Anal.*, **22**, 13 (2000).
23. A. A. M. Moustafa, *J. Pharm.. Biomed. Anal.*, **22**, 45 (2000).
24. N. Rehman and S. N. Hejaz-Azmi, *Anal. Sci.*, **16**, 1353 (2000).
25. N. Rehman, A. K. Nadeen and S. N. Hejaz-Azmi, *Anal. Sci.*, **20**,1231(2004).
26. W. Likussar and D. F. Boltz, *Anal. Chem.*, **43**, 1265 (1971).
27. *British Pharmacopoeia*, Her Majesty's Stationery Office, London (2003).