Spectrophotometric Determination of Promethazine Hydrochloride in Pharmaceutical Formulations

K. C. RAMESH, B. G. GOWDA[1] AND J. KESHAVAYYA*
Department of Chemistry, Kuvempu University, Shankaraghatta-577 451.
[1]Department of Chemistry, Karnatak University, Dharwad-580 003.

Accepted 30 March 2003
Revised 24 February 2003
Received 5 February 2002

A simple, rapid and selective spectrophotometric method has been proposed for the assay of promethazine hydrochloride in pure and pharmaceutical formulations. The method is based on the formation of a chloroform-soluble red coloured product having an absorption maximum at 516 nm with o-toluidine in presence of N-bromosuccinimide. The reaction conditions were optimized. Beer's law is valid in the concentration range 2-15 μg/ml (r=0.9993) having molar absorptivity of 4.64X10^3 l/mol.cm. The results obtained by the proposed method were compared by means of Student t- test and by the F-test with those of official method and were found to be in good agreement. No interference was observed from common excipients. Moreover, the method is specific for C-2 unsubstituted phenothiazines.

Promethazine hydrochloride (PMH), chemically 10-[2-(dimethylamino)propyl] phenothiazine hydrochloride, is used as an antihistaminic, antiemetic and as a tranquillizer. The drug is common ingredient of a number of pharmaceutical formulations for cough and cold remedies. In view of its extensive applications analytical techniques such as atomic absorption spectrometry,[1] spectrofluorimetry[2], voltammetry[3], non-aqueous capillary electrophoresis[4], FIA with chemiluminescence detection[5], HPLC[6] and AC oscillopolarographic titration[7] have been reported for the assay of PMH. However, these are costly and are not available at most of the quality control laboratories. Spectrophotometric technique continues to be the most preferred method for routine analytical work because of its simplicity and reasonable sensitivity with significant economical advantages. Hence numerous spectrophotometric methods have been proposed for the determination of PMH[8-18]. Most of these methods are based either on coloured complex formation or on oxidative reactions. Some of these require heating for long time[8-12] or long-standing time[13-16] for colour development or involve the use of high concentration of sulphuric acid[16-19]. Spectrophotometric methods based on the formation of coloured radical cations suffer from twin disadvantages of critical acid or oxidant concentration and instability of the coloured species[10,19]; the stability ranging from 15-30 min. The USP method[20] for the assay of PMH involves non-aqueous titration, which is laborious and time consuming.

*For correspondence
E-mail: jkeshavayya@rediffmail.com

The aim of present study is the development of a simple and selective spectrophotometric method for the assay of PMH in pure form and in pharmaceutical preparations. The method is based on the formation of a red coloured product with o-toluidine (OTD) in presence of N-bromosuccinimide (NBS). The use of OTD and NBS as analytical reagents provides an enhanced stability compared with most of the reagents reported earlier. The resulting method is also simple, rapid and quite specific for the determination of PMH.

All spectral measurements were made on a Shimadzu 160 UV/Vis spectrophotometer with 1 cm matched quartz cells. PMH was supplied gratis by Rhone-Poulenc Ltd., Mumbai. Various dosage forms of PMH were obtained commercially. All other chemicals were of analytical reagent or pharmaceutical grade. Quartz-processed high-purity water was used throughout.

A stock solution of PMH was prepared by dissolving 100 mg of pure PMH in distilled water in a 100 ml calibrated

flask. It was diluted as and when required. An aqueous solution of NBS (0.01 %) was prepared in distilled water. A 0.1 % solution of OTD prepared in ethanol was used for the study.

Suitable amounts of an aliquot of PMH (20-150 μg) were transferred into a series of 125 ml separating funnels. To each of these funnels were added 4.2 ml of NBS and 3.2 ml of OTD. The volume of aqueous phase was adjusted to 20 ml and 10 ml of chloroform was added to each of the funnels. The contents were shaken thoroughly and allowed to separate. The organic layer was passed through anhydrous sodium sulphate and transferred to 10 ml calibrated flasks. The absorbance of the red coloured species was measured at 516 nm against a reagent blank prepared identically. A calibration graph was plotted.

Twenty tablets were finely powdered. The powder equivalent to 20 mg of PMH was weighed accurately and treated with distilled water. The mixture was shaken for 15 min and filtered through a Whatman filter paper No. 40. The filtrate and washings were combined, and transferred to a 100 ml volumetric flask. It was diluted up to the mark with distilled water. An aliquot of the solution was analyzed as described for pure drug.

In respect of injection and syrup, requisite volume containing about 10 mg of the drug was transferred to a 100 ml calibrated flask and diluted up to the mark with distilled water. The amount of the drug content was analyzed as described for pure drug.

In case of elixir 10 ml equivalent to 10 mg of drug were transferred into a 250 ml separating funnel and rendered alkaline to litmus paper with 6 M ammonia solution. The mixture was extracted with 3x15 ml portions of chloroform; the chloroform extracts were evaporated to dryness and the residue was dissolved in 0.1 M HCl and made up to 50 ml with distilled water. An aliquot of this solution was treated as described above.

NBS oxidized the PMH to an unstable (2 min) red coloured product of lower sensitivity. When OTD was added to the oxidized product of PMH, an intense green coloured product (λ_{max} 580 nm) was obtained in aqueous medium. But the colour intensity decreased gradually. However, when it was extracted into chloroform, it gave a stable red coloured product with λ_{max} at 516 nm. The probable reaction mechanism for the formation of the coloured product[11,22] may be presented as shown in fig. 1. The optimum reaction conditions were achieved *via* a number of preliminary investigations. It was observed that 4.2 ml of NBS and 3.2 ml of OTD in a total volume of 10 ml were sufficient for achieving a coloured product of maximum intensity and stability. Of the tested water-immiscible organic solvents, chloroform was found to be the most suitable as it was observed that only one extraction was sufficient for quantitative recovery of the coloured product. The order of addition of reagents was found to be critical and hence the sequence suggested in the procedure was followed. It was observed that the coloured product was stable for more than 15 h.

Specificity of the method was checked by examining the effect of the reagents on C-2 substituted phenothiazine derivatives that include trifluoperazine hydrochloride, butaperazine dimaleate, thioridazine hydrochloride, fluphenazine hydrochloride, mepazine hydrochloride and thioproperazine mesylate. These compounds did not yield any red coloured product upon extraction into chloroform indicating the good specificity for C-2 unsubstituted phenothiazines.

The Beer's law limits, molar absorptivity and Sandell's sensitivity were evaluated. Regression analyses of Beer's law plot revealed a good correlation. Graph of absorbance *versus* concentration showed a zero intercept, and is described by the regression equation Y=a+bX (where, Y is the absorbance of a 1 cm layer, b is the slope, a is the intercept

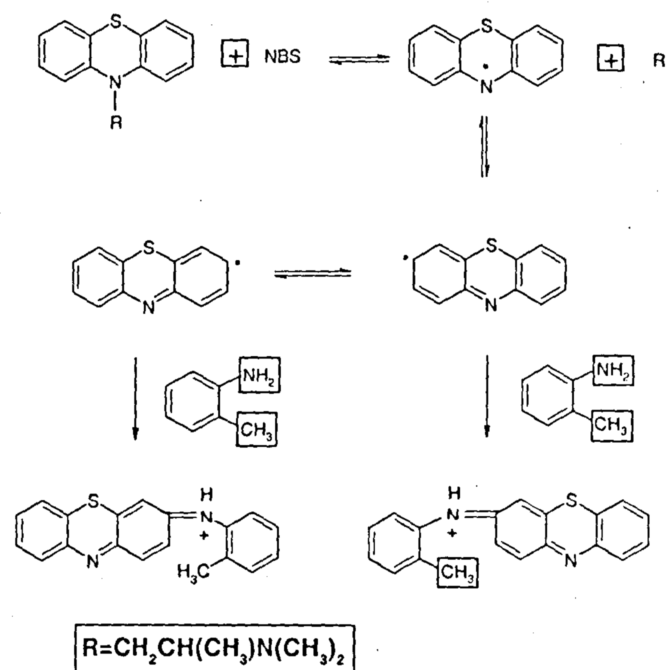

Fig. 1: Reaction scheme.

and X is the concentration of each of PMH in μg/ml) obtained by least-squares method. The results are summarized in Table 1. The precision value of the proposed method was good as indicated from the low relative standard deviation (1.0%) calculated from six replicate analyses of PMH.

The effects of various excipients generally present in dosage forms of PMH were investigated. The results indicated that the talc, dextrose, starch, magnesium stearate and gelatin did not interfere in the assay in amounts far in excess of their normal occurrence in dosage forms.

The recovery technique was applied to judge the suitability of the proposed method. For this, known quantities of pure PMH solution were mixed with definite amounts of preanalysed formulations and the mixtures were analyzed as before. The total amount of PMH was then determined using the proposed method and the amount of the added drug was calculated by the difference. The results were found to be satisfactory.

The applicability of the proposed method was examined by analyzing tablets, syrup, elixir and injection mar-

TABLE 1: OPTICAL CHARACTERISTICS, PRECISION AND ACCURACY DATA.

Parameter	Value
λ_{max} (nm)	516
Beer's law limits (μg/ml)	2-15
Molar absorptivity (l/mol.cm)	4.64×10^3
Sandell's sensitivity (ng cm^{-2})	60.37
Correlation coefficient (r)	0.9993
Regression equation (Y)*	
Slope, b	0.0358
Intercept, a	0.0037
Relative standard deviation (%)**	0.86
% Range of error** (95 % confidence limit)	0.69

*Y=a+bX where X is the concentration of PMH in μg/ml, **For six replicate analysis within Beer's law limits.

TABLE 2: DETERMINATION OF PMH IN PHARMACEUTICAL PREPARATIONS.

Dosage form	Label claim (mg)	USP method[20]	Recovery* ± SD, % and its comparison with the USP method
Tablets			
Avomine[1]	25	99.7±1.07	98.8±1.05 F=1.03; t =1.23
Phenergan[1]	25	99.0±1.23	99.5±0.99 F=1.54; t =1.75
Phena[2]	25	98.4±0.52	99.0±0.58 F=1.24; t =1.15
Romergan[3]	25	99.1±0.52	99.1±0.62 F=1.42; t =1.82
Injection			
Phenergan[1]	25	100±0.67	99.8±0.61 F=1.37; t =1.31
Syrup			
Promethazine[4]	1.5	98.1±1.2	98.3±1.18 F=1.03; t =1.42
Elixir			
Phenergan[1]	5.0	99.1±1.05	99.3±1.10 F=1.09; t =1.12

*Average of five determinations; Marketed by [1]Rhone-Poulenc Ltd., Mumbai; [2]Ind-Swift Ltd., Chandigarh; [3]Uni-UCB Ltd., Mumbai; [4]Jagsonpal Pharma Ltd., New Delhi.

keted under different trade names. The results obtained were compared statistically by Student t-test and by the variance ratio F-test with those obtained by official method[20]. The Student t-values calculated for 95 % confidence level did not exceed the theoretical value indicating that there was no significant difference between the proposed and the official method. It was also observed that the variance ratio F-values calculated for p=0.05 did not exceed the theoretical value indicating that there was no significant difference between the precision of the proposed and official method. The results are tabulated in Table 2.

The proposed method is simple and specific for C-2 unsubstituted phenothiazine like PMH only. The method is more sensitive compared to most of the reported methods and the results are reproducible. The coloured product formed in the present investigation has longer stability, which made the method more practicable over the reported methods. Hence, the proposed method could be used as a better alternative for the assay of PMH in pure and pharmaceutical formulations.

ACKNOWLEDGEMENTS

The authors are indebted to Rhone-Poulenc (India) Ltd., for the gift sample of pure PMH. Thanks are also due to the authorities of Kuvempu University for providing the necessary facilities. One of the authors (KCR) thanks UGC for awarding the fellowship under FIP.

REFERENCES

1. El-Ansary, A.L., El-Hawary, W.F., Issa, Y.M. and Ahmed A.F., **Quim. Anal.**, (Barcelona) 1998, 17, 199.
2. Hornyak, I., Kozma, L., Lapat, A. and Tovari, I., **Biomed. Chromatogr.**, 1997, 11, 99.
3. Uslu, B., Biryol, I., Ozkan, S.A. and Zuhre, S., Turk. **J. Chem.**, 1996, 20, 323.
4. Lu, X. and Wang, R., **Ming-Jia Gaodeng Xuexiao Huaxue Xuebao**, 1999, 20, 856.
5. Xue, Y., He, Y., Feng, M. and Lu, J., **Fenxi Huaxue**, 1999, 427.
6. Saleh, M.I. and Yoong, C.S., **J. Phys. Sci.**, 1993, 4, 55.
7. Liu, C. and Cui, Z., **Huaxi Yaoxue Zazhi**, 1993, 8, 214.
8. Devani, M.B., Suhagia, B.N. and Shah, S.A., **Indian J. Pharm. Sci.**, 1999, 61, 110.
9. Sastry, C.S.P., Tipirneni, A.S.R.P. and Suryanarayana, M.V., **J. Pharm. and Biomed. Anal.**, 1990, 8, 287.
10. Sastry, C.S.P., Tipirneni, A.S.R.P. and Suryanarayana, M.V., **Indian Drugs**, 1987, 26, 351.
11. El-Shabouri, S.R., Youssef, A.F., Mohamed, F.A. and Rageh, A.M.I., **J. Assoc. Anal. Chem.**, 1986, 69, 821.
12. Issa, A.S., Beltagy, Y.A. and Mahrous, M.S., **Talanta**, 1978, 25, 710.
13. Basavaiah, K. and Krishnamurthy, G., **Indian J. Pharm. Sci.**, 1997, 59, 327.
14. Mangala, D.S. and Sastry, C.S.P., **Indian J. Pharm. Sci.**, 1984, 46, 154.
15. El-Sebai, A.I., Issa, A.S., Salam, M.A.A. and Mahrous, M.S., **Talanta**, 1983, 30, 531.
16. Taha, A.M., El-Rabbat, N.A., El-Kommos, M.E. and Refat, I.H., **Analyst**, 1983, 108, 1500.
17. Sane, R.T., Kamat, S.S., Narkar, V.S., Sathe, A.Y. and Mhalas, J.G., **Indian Drugs**, 1980, 19.
18. Zarapakar, S.S. and Athalye, K.R., **Indian Drugs**, 1987, 26, 32.
19. Ramappa, P.G. and Nayak, A.N., **Indian J. Pharm. Sci.**, 1983, 45, 65.
20. United States Pharmacopoeia, XXII, National Formulary XXI, Rockville, MD, 1995, 1311.
21. British Pharmacopoeia, HMSO, London, 1993, 552.
22. Kommos, M.E. and Emara, K.M., **Analyst**, 1988, 113, 1267.