ACKNOWLEDGEMENTS

One of the author (DBW) is grateful to Dr. S. N. Umathe and Mr. M. R. Tajne for providing necessary help.

REFERENCES

1. Dai, J.D., Jin, D.J. and Liu, Y.L., **Yaowu Fenxi Zazhi,** 2001, 21, 36.
2. Zhong, W.Z. and Williams, M.G., **J. Pharm. Biomed. Anal.,** 1996, 14,465.
3. Yamashita, W.Z., Murakami, H., Okuda, T. and Motohashi, M., **J. Chromatogr.,** 1996, 677, 141.
4. United State Pharmacopoeia, United State Pharmacopoeial Convention Inc., Rockville, MD, 1998, 1923.

Simultaneous Spectrophotometric Estimation of Nalidixic Acid and Metronidazole in Combined Dosage Forms.

VAISHALI NAGULWAR*, P. A. UPADHYE, N. K. MEHTA, KANCHAN UPADHYE, SUPARNA BAKHLE, SHILPA DESHPANDE AND GOURI DIXIT.
J. L. Chaturvedi College of Pharmacy, 846, New Nandanvan, Nagpur-440 009

Accepted 2 April 2005
Revised 26 October 2004
Received 14 January 2004

Nalidixic acid and metronidazole in combination are routinely used as antidiarrhoeal. The present investigation attempts to develop estimation method for these two drugs in combined dosage forms. It was found that both the drugs follow the Beer's Law from 2.0-10.0 µg/ml and 2.0-12.0 µg/ml at 257 and 277 nm, respectively. The absorptivity (1%, 1 cm), values at the specified wavelengths for nalidixic acid was found to be 1232.84 and 211.05, respectively and for metronidazole 263.65 and 376.18, respectively. Thus, the present method is simple, rapid and accurate and is based on the principle of simultaneous UV spectrophotometric determination of binary mixture.

The combination of nalidixic acid and metronidazole is marketed as tablet and suspension formulations and is used as antidiarrhoeal. Chemically, nalidixic acid is 1-ethyl-1,4-dihydro-7-methyl-4-oxo-1,8-naphthyridine-3-carboxylic acid and official in IP[1]. It is an antibacterial agent active against enterobacteriaceae and is given orally to treat urinary tract infections[2]. Chemically, metronidazole is 2-(2-methyl-5-nitro-1H-imidazol-1-yl)-ethanol and is also official in IP[3]. It is an antimicrobial drug, active against obligate anaerobic micro-organisms both bacteria and protozoa[4]. Though both the drugs have overlapping UV absorption spectra in acidic media, their absorption maxima are quite well separated from each other. In this attempt, the Vierodt's method, one of the standard methods for simultaneous estimation of binary mixture was employed [5].

***For correspondence**
E-mail: vaishali614@yahoo.com.

Nalidixic acid was procured from M/s Ranbaxy Laboratories Ltd., Devas and metronidazole from M/s Oscar Laboratories Pvt. Ltd, New Delhi as gift samples. The tablets (referred as T_1 and T_2) and suspension (referred as S) of the said combination were purchased from a local pharmacy. (The label claim for both T_1 and T_2 contained 300 mg nalidixic acid and 200 mg of metronidazole and each 5 ml of S contained 150 mg of nalidixic acid and 100 mg of metronidazole). All the chemicals and solvents used were of AR/GR grade. The instrument, LKB Ultrospec 4050 Spectrophotometer and a Shimadzu 150 UV/Vis Spectrophotometer were used for spectrophotometric readings.

To establish the suitability of the proposed method for determination of nalidixic acid and metronidazole in the pharmaceutical formulation, the method was first tried for the estimation of components in standard laboratory mixture of two drugs.

For estimation of drugs in standard laboratory mixture, accurately weighed quantities of nalidixic acid (equivalent to 50 mg) and metronidazole (equivalent to 25 mg) were taken in 50 ml volumetric flask and the mixture was dissolved in 40 ml of methanol by warming. The solution was cooled to room temperature and was adjusted to the mark by adding sufficient methanol. An aliquot of 10 ml of this solution was diluted to 100 ml with 0.1 N hydrochloric acid. A portion (5 ml) of the resultant solution was further diluted to 100 ml with 0.1 N hydrochloric acid and the absorbance of the solution was measured at 257 nm and 277 nm, respectively. The contents of nalidixic acid and metronidazole were calculated by substituting the absorbance values at 257 nm and 277 nm in the simultaneous equation given as follows, $c_1 = \lambda_2 a_2 \times A\lambda_1 - \lambda_1 a_2 \times A\lambda_2 / \lambda_1 a_1 \times \lambda_2 a_2 - \lambda_1 a_2 \times \lambda_2 a_1$ and $c_2 = \lambda_1 a_1 \times A\lambda_2 - \lambda_2 a_1 \times A\lambda_1 / \lambda_1 a_1 \times \lambda_2 a_2 - \lambda_1 a_2 \times \lambda_2 a_1$, where, c_1 and c_2 are the concentrations and λ_1 and λ_2 are the wavelengths (257 and 277 nm) of nalidixic acid and metronidazole, respectively. $A\lambda_1$ and $A\lambda_2$ are the absorbances of the sample solution at λ_1 and λ_2. $\lambda_1 a_1$ and $\lambda_2 a_1$ are the absorptivity values of nalidixic acid at λ_1 and λ_2, respectively and $\lambda_1 a_2$ and $\lambda_2 a_2$ are the absorptivity values of metronidazole at λ_1 and λ_2, respectively.

For estimation of nalidixic acid and metronidazole in tablets, twenty tablets (Abdogyl-Biddle Sawyer Pharmaceutical Ltd.) were weighed and finely powdered. An accurately weighed quantity of tablet powder equivalent to the about 50 mg nalidixic acid was transferred to 50 ml volumetric flask and 40 ml methanol was added to it and mixture was warmed for about 15 min. The solution was cooled to room temperature and was adjusted to mark with methanol. The solution was then filtered and 10 ml portion of the filtrate was diluted to 100 ml with 0.1 N hydrochloric acid. A portion (5.0 ml) of the resultant solution was further diluted to 100 ml with 0.1 N hydrochloric acid. The absorbance of the solution was measured at 257 and 277 nm, respectively against a blank. The content of nalidixic acid and metronidazole were calculated using simultaneous equations. The results are shown in Table 1.

For estimation of nalidixic acid and metronidazole in suspension, weight per ml of the suspension was determined at room temperature and accurately weighed quantity of suspension equivalent to about 50 mg of nalidixic acid was transferred to 50 ml volumetric flask. About 40 ml methanol was added in the flask and the mixture was warmed for about 15 min. The solution was cooled to room temperature and was adjusted to mark with methanol. The solution was then filtered and 10 ml portion of the filtrate was diluted to 100 ml with 0.1 N hydrochloric acid. A portion (5.0 ml) of the resultant solution was further diluted to 100 ml with 0.1 N hydrochloric acid. The absorbance of the solution was measured at 257 and 277 nm, respectively against a blank. The con-

TABLE 1: RESULTS OF ASSAY OF NALIDIXIC ACID AND METRONIDAZOLE IN PHARMACEUTICAL FORMULATIONS*

Samples and formulations	Amount of drug (mg)		% of Labeled claim**	% Recovery
	Taken	Found		
Lab. Mixture				
Nalidixic Acid	49.4	48.7±0.06	98.5±0.13	
Metronidazole	22.4	22.9±0.04	101±2.2	
Tablet T_1				
Nalidixic Acid	~50.0	49.1±0.22	98.2±0.81	99.3±1.54
Metronidazole	~33.3	33.8±0.24	99.5±1.0	101±1.4
Tablet T_2				
Nalidixic Acid	~50.	48.9±0.35	96.8±0.69	100±0.7
Metronidazole	~33.3	32.7±0.08	98.1±1.83	99.7±1.0
Suspension (S)				
Nalidixic Acid	~50.0	48.7±0.08	97.3±0.16	99.9±0.74
Metronidazole	~33.3	32.6±0.06	97.7±0.24	98.5±1.01

*Brand name of the marketed formulations -Abdogyl (Biddle Sawyer). **All the observations are the mean of five readings.

tent of nalidixic acid and metronidazole were calculated using simultaneous equations. The results are shown in Table 1.

Recovery studies for tablet formulations were then done by accurately weighing the quantity of tablet powder equivalent to 100 mg of nalidixic acid and transferring to 50 ml volumetric flask and then it to 10 mg of pure nalidixic acid and 10 mg of metronidazole were added accurately. The mixture was then dissolved in methanol with slight warming and diluted to 50.0 ml. The solution was then filtered and 10 ml portion of the filtrate was diluted to 100 ml with 0.1N hydrochloric acid. A portion (5.0 ml) of the resultant solution was further diluted to 100 ml with 0.1 N hydrochloric acid. The absorbance of the solution was measured at 257 nm and 277 nm against a blank. The content of nalidixic acid and metronidazole were calculated using simultaneous equations. The percent recovery was then calculated with respect to the amount of pure nalidixic acid and metronidazole added.

For recovery studies for suspension formulation, the same procedure as detailed under recovery studies for tablet was repeated by weighing the quantity of suspension equivalent to about 50 mg of nalidixic acid and adding accurately weighed quantities of nalidixic acid and metronidazole. The results are shown in Table 1.

Accuracy of the analysis was determined by calculating recovery of nalidixic acid and metronidazole by standard addition method. The results indicated that the recovery of nalidixic acid ranged between 99.0-100 and that of metronidazole between 98.0-100, ensuring that the method is accurate and reproducible. It also appears that the excepients present in formulation do not interfere in the proposed method. In addition to this, the method is simple rapid and cost effective. Hence, the method may be employed for routine quality control of formulations containing nalidixic acid and metronidazole.

ACKNOWLEDGEMENTS

The authors are thankful to M/s Ranbaxy Laboratories Ltd., Devas and M/s Oscar Laboratories Pvt. Ltd, New Delhi for generously donating the pure drugs nalidixic acid and metronidazole used for present investigation as gift samples. They are also thankful to Prof. V. T. Kharkar, Principal, J. L. Chaturvedi College of Pharmacy, Nagpur for his support and kind cooperation in completion of this research work.

REFERENCES

1. Indian Pharmacopoeia, 4th Edn., Ministry of Health and Family Welfare, New Delhi, 1996, 497.
2. Reynolds, J.E.F., Eds., In; Martindale, The Extra Pharmacopoeia, 30th Edn., The Pharmaceutical Press, London, 1993, 184.
3. Indian Pharmacopoeia, 4th Edn., Ministry of Health and Family Welfare, New Delhi, 1996, 488.
4. Dollery, S.C., In; Therapeutic Drugs, Vol. II, Churchill Livingstone, London, 1992, M-170.
5. Heilmeyer, L., In; Spectrophotometry in Medicines, Adam Hilger Ltd., London, 1943, 65.

Validation of a RP-LC Method for Simultaneous Determination of Paracetamol, Methocarbamol and Diclofenac Potassium in Tablets

G. SUBRAMANIAN[1]*, M. VASUDEVAN, S. RAVISANKAR AND B. SURESH

J. S. S. College of Pharmacy, Rocklands, Ooty-643 001

Department of Pharmaceutical Quality Assurance, Manipal College of Pharmaceutical Sciences, Manipal-576 104

Accepted 2 April 2005

Revised 27 October 2004

Received 4 March 2004

A simple, fast, precise and accurate liquid chromatographic method was developed for the simultaneous estimation of paracetamol, methocarbamol and diclofenac potassium in tablets. Drugs were chromatographed on a reverse phase Hypersil C_{18} column using a mobile phase, 25 mM

*For correspondence

E-mail: ganrajesh@yahoo.com