Int. J. Chem. Sci.: 7(3), 2009, 1766-1770

SIMULTANEOUS DETERMINATION OF OFLOXACIN AND METRONIDAZOLE FROM COMBINED DOSAGE FORMS

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ABSTRACT

A new spectrophotometric method for the simultaneous and individual estimation of ofloxacin and metronidazole in binary tablet formulations has been described. The method is based on the estimation of one drug in presence of another drug by absorbance difference method.

Key words : Spectrophotometry, Ofloxacin, Metronidazole.

INTRODUCTION

The combination formulations of ofloxacin and metronidazole have been in the market for their use in dysentery. Various methods for simultaneous analysis of ofloxacin and metronidazole in binary tablet formulations have been reported. These include spectrophotometry[1-9], colorimetry[10], HP-TLC method[11], HPLC method[16] and RP-HPLC method[17]. No method for the simultaneous analysis of ofloxacin and metronidazole in binary tablet formulations has been reported by absorbance difference method. The aim of the present work is to develop a simple, rapid, precise, reproducible and economical method for the simultaneous analysis of the binary drug formulations by using absorbance difference method without any interference from each other.

EXPERIEMNTAL

Materials and methods

A Spectronic 1001, spectrophotometer with 10 mm quartz cells was used for measuring absorbance values of the drug solution. All the chemicals used were of analytical grade. AR Grade methanol was used as solvent.

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Preparation of standard ofloxocin solution

Pure ofloxacin (50 mg) was dissolved in 50 mL methanol. Further 1 mL of the stock solution was further diluted to 200 mL with methanol to get working concentration of 20 μg/mL.

Preparation of standard metronidazole solution

Fifty mg of pure metronidazole was dissolved in 50 mL methanol to obtain the working concentration of 1 mg/mL. One mL of the above stock solution was further diluted to 50 mL with methanol to get working concentration of 20 μg/mL.

Preparation of mixed solution

Two solution, the first containing 20 μg/mL of ofloxacin and the second containing 20 μg/mL of metronidazole were used as mixed solutions. Four mixed standard solutions were made by taking 4, 3, 2 and 1 mL of ofloxacin solution into series of test tubes and the metronidazole stock solution was also added into series of test tubes to make the total volume at 5 mL.

Preparation of ofloxacin curve

Various aliquots (5, 6, 7 and 8 mL) of ofloxacin solution were transferred into a series of 10 mL standard flask and the volume in each flask was adjusted to 10 mL with distilled water. The absorbance of these solutions was scanned over the range of 220 to 385 nm. Again various aliquots (5, 6, 7 and 8 mL) of metronidazole solutions were transferred into a series of 10 mL volumetric flask and the volume in each flask was adjusted into a series of 10 mL volumetric flask and the volume in each flask was adjusted to 10 mL with distilled water. These solutions were scanned over the range of 220 to 370 nm. Two wavelengths, 260 and 305 nm are selected for ofloxacin. At these two wavelengths the absorbance values are almost zero and in case of metronidazole, at the same wavelength 260 and 305 nm, there was maximum absorbance difference. A calibration curve was drawn between the absorbance difference values of metronidazole and the amount of metronidazole in μg/mL. The amount of metronidazole present in the sample was estimated from calibration curve. Similarly two wavelengths 295 and 350 nm were selected for metronidazole. At these two wavelengths, the absorbance difference was almost zero and in case of ofloxacin, maximum absorbance difference values was there at the same wavelengths 295 and 350 nm. A calibration curve was drawn between absorbance difference values of ofloxacin and amount of ofloxacin in μg/mL. Amount of ofloxacin

present in the sample was estimated from calibration curve.

Various aliquots of mixture of ofloxacin and metronidazole solutions in different proportions were transferred into series of test tubes and the volume in each test tube was kept at 5 mL. The absorbence values were measured at two wavelength 295 and 350 nm for estimation of ofloxacin and two wavelengths 260 and 305 nm for estimation of metronidazole. A calibration curve was drawn between the absorbance difference values of ofloxacin and the amount of ofloxacin present in μg/mL. A calibration graph was drawn between the absorbance difference values of metroinidazole and the amount of metronidazole present in μg/mL. A linear curve in each case was obtained. The linearity of the curves obtained indicates that it obeys Beer's law and this method is suitable for the simultaneous determination of the two drugs in admixture.

Extimation of ofloxacin in formulations

Tablets were weighed and powdered. An average weight of the tablet containing the two drugs ofloxacin and metronidazole in the ration of 2 : 3 and the amount of 580 mg was dissolved in 30 mL methanol by vigorously shaking and the volume was made upto the mark. The solution was then filtered through Whatmann filter paper No. 41 and the solution was diluted to get a final concentration of 20 μg/mL of fofloxacin and 50 μg/mL of metronidazole. The sample solutions were measured at 295 and 350 nm for ofloxacin and 260 and 305 nm for metronidazole in the Spectronic 1001, spectrophotometer. The results are represented in the Table 1.

Table 1: Estemation of ofloxacillin and metronidazole in pharmaceutical formulations

Sample	Label claim (mg/tab)		Amount found by proposed method (mg)		% Recovery by proposed method	
	*OF	**MD	*OF	**MD	*OF	**MD
Tablet 1	50	100	79.96	99.98	99.92	99.98
Tablet 2	50	100	49.96	99.86	99.92	99.86
Tablet 3	50	100	49.86	99.92	99.72	99.92

Average of five determinations, *ofloxacin (OF) and **metronidazole (MD)

RESULTS AND DISCUSSION

The present study was carried out to develop a simple, rapid, sensitive, precise, reproducible and accurate spectrophotometric method for the estimation of simultaneous determinations of ofloxacin and metronidazole in pharmaceutical dosage forms. The proposed absorbance difference method was simple, less time consuming, low cost and found to be one of the best versatile analytical technique employed for routine analysis purpose like assay and pharmaceutical formulations. No method for the simultaneous analysis of ofloxacin and metronidazole in binary tablet formulations has been reported in the literature by absorbance difference method. The results obtained by proposed method are in good agreement with label claim of the tablets. The additive and excipients usually present in tablets do not interfere. As a check on accuracy of the method, recovery experiment was performed and percent recovery values are tabulated (Table 2). The statistical analysis was carried out by proposed method. The values of standard deviation and coefficient of variation values were satisfactorily low, indicating accuracy and the reproducibility of the method. Student's 't' test shows that the calculated 't' values are less than 't' theoretical value 2.78 with 4 degree of freedom at 5% level of significance indicating that there is no significant difference between proposed and official method.

In conclusion, the results indicate that the proposed absorbance difference method was found to be simple, rapid, precise, accurate and less time consuming. Hence, it can be used for the routine analysis of simultaneous determination of ofloxacin and metronidazole in pharmaceutical formulations.

Table 2 : Statistical analysis of estimation of ofloxacin and metronidazole

Sample	[a]Standard deviation		[a]Coefficient of variation		[a]t_{cal}	
	*OF	**MD	*OF	**MD	*OF	**MD
Tablet 1	0.2880	0.4324	0.5766	0.4324	0.3105	0.1034
Tablet 2	0.6387	0.4219	1.278	0.4224	0.1400	0.7423
Tablet 3	0.2408	0.2167	0.4830	0.2169	1.301	0.8255

*ofloxacin and **Metroniazole

[a]Average of five determinations based on label claim

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Accepted : 21.03.2009