RP-HPLC METHOD FOR SIMULTANEOUS DETERMINATION OF PARACETAMOL AND ASPIRIN IN TABLETS, SOFIA, BULGARIA

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ABSTRACT

A specific, rapid and simple RP-HPLC method with good sensitivity has been developed and validated for the simultaneous quantification of aspirin and paracetamol in standard solutions and tablets. The separation was carried out by using a mobile phase consisting of acetonitrile:phosphate buffer pH 7.0 in ratio of 60-40 (*v/v*). The column used was *Phenomenex-Luna, C8 (125 mm x 4.6 mm i.d., 5 μm)* with flow rate of 1.0 ml/min using UV detection at 230 nm. The described method was linear over a concentration range of 2-64 μg/ml ($R^2 > 0.9999$) for both drugs. The retention times of paracetamol and aspirin were found to be 3.05 and 4.85, respectively. Results of analysis were validated statistically and by recovery studies (mean recovery =99.20 % for paracetamol and 99.83 % for aspirin). The limit of quantification (LOQ) for paracetamol and aspirin were found to be 0.1 μg/ml and 0.2 μg/ml, respectively. It can be concluded from the results that present method for the simultaneous determination of aspirin and paracetamol in tablets is specific, rapid and simple with good sensitivity. The analytical method can be successfully adopted for quality control tests for these drugs in tablet dosage forms.

Keywords: RP-HPLC, Validation, Paracetamol, Aspirin, Quality Control

INTRODUCTION

Aspirin and paracetamol have been extensively used as antipyretic and analgesic drugs. They are frequently prescribed in admixture with each other or in combination with other drugs [1, 2].

Literature survey revealed that several methods have been reported for the determination of paracetamol and aspirin. Ramos-Martos *et al*., [3] applied liquid chromatography to the simultaneous

analysis of acetylsalicylic acid, caffeine, codeine, paracetamol, pyridoxine and thiamine in pharmaceutical preparations. Vidal *et al*., [4] determined simultaneously aspirin, acetaminophen and caffeine using UV-spectrophotometry. Martos *et al*., [5] conducted spectrofluorimetric determination of aspirin and codeine in pharmaceuticals. Szostak and Mazurec [6] presented quantitative determination of aspirin and paracetamol in tablets by FT-Raman spectroscopy. Cemal *et al*., [7] developed a rapid assay of aspirin, paracetamol and their degradation products in tablets by HPLC. Abu-Qare and Abou-Donia [8] described a HPLC method for the determination of pyridostigmine bromide, acetaminophen, aspirin and caffeine in rat plasma and urine. Gondalia *et al*., [9] developed and validated spectrophotometric method for simultaneous estimation of paracetamol and ibuprofen.

The aim of the present study was to develop and validate a HPLC method for the simultaneous determination of paracetamol and aspirin in tablets. The method described complied with validation requirements of ICH and could be used for routine quality control of pharmaceutical formulations in ordinary laboratories.

METHODOLOGY

Chemicals and Reagents

Working standards of aspirin RS and paracetamol RS were provided by (Sigma-Aldrich). LC-grade acetonitril was supplied from Merck (Germany). All other chemical reagents were of analytical grade.

Instrumentation and Chromatographic Conditions

Chromatographic separation was performed on modular HPLC system LC-10A Shimadzu (Japan) arranged with a LC-10A pump, solvent degasser DGU-3A, Rheodyne injector, column oven CTO-10A, SPD-M10A diode array detector and communication bus module CBM-10A. Separation was achieved isocratically with a *Phenomenex-Luna, C8 (125 mm x 4.6 mm i.d., 5 μm)* column eluted with a mixture consisting of acetonitrile:phosphate buffer pH 7.0 in ratio of 60-40 (*v/v*) as the mobile phase at flow rate 1.0 ml/min. Detection was carried out by absorbance at 230 nm. The analysis was carried out at an ambient temperature and injection volume was 20 μl.

Stock Standard Solutions

Stock standard solutions of aspirin (100 μg/ml) and paracetamol (100 μg/ml) were prepared at the same manner by dissolving 20 mg aspirin or paracetamol in methanol in 200 ml volumetric flask. The solutions were further diluted to get various working solutions.

Working Solutions of Aspirin Plus Paracetamol

The calculated volumes of aspirin and paracetamol solutions were taken from stock standard solutions, mixed and prepared serial dilutions with methanol to obtain a concentrations as follows: 2, 4, 8, 16, 32 and 64 µg/ml of each, aspirin and paracetamol.

Sample Solutions

The pharmaceutical preparation containing paracetamol and aspirin (100 mg of each drug) was analyzed using this method. Twenty tablets were accurately weighed and crushed into powder. A weighed quantity of tablet powder equivalent to 50 mg aspirin and 50 mg of paracetamol were transferred into a 100 ml volumetric flask, diluted with approximately 70 ml methanol, sonicated for 10 min and made up the volume to the mark with methanol. Then the resulting solution was filtered through a 0.22 µm filter and filtrate was suitably diluted to produce the desired concentrations (2, 4, 8, 16, 32 and 64 µg/ml) for both, aspirin and paracetamol.

RESULTS AND DISCUSSION

A reverse-phase HPLC method was developed for the simultaneous determination of paracetamol and aspirin in tablets. From the chromatogram shown in **Figure 1**, it is evident that under the proposed chromatographic conditions both analytes were completely separated from each other, which indicated that the method is selective and could be applied for their identification and quantification simultaneously. No peaks were observed in the chromatogram of a blank sample, which showed that no interferences from the excipients occurred.

Linearity

Working solutions of both drugs in concentration range of 2 to 64 µg/ml were prepared and analysed chromatographically to construct calibration curve between concentration and chromatographic areas. The values of regression coefficient (R^2) for aspirin and paracetamol were 0.9925 and 0.9905, respectively which indicate a good correlation between concentration and area within the concentration range tested. The linear regression data for calibration curves were presented in **Table 1**.

Precision

To check precision (percentage RSD) of analytical method, six replicate samples of the same concentrations of aspirin and paracetamol were analysed. The precision for aspirin and paracetamol were 0.50 and 0.41 %, respectively **(Table 2)**.

Accuracy

To study accuracy, recovery experiments were carried out. The average percentage recovery of paracetamol and aspirin was 99.20% and 99.83% respectively.
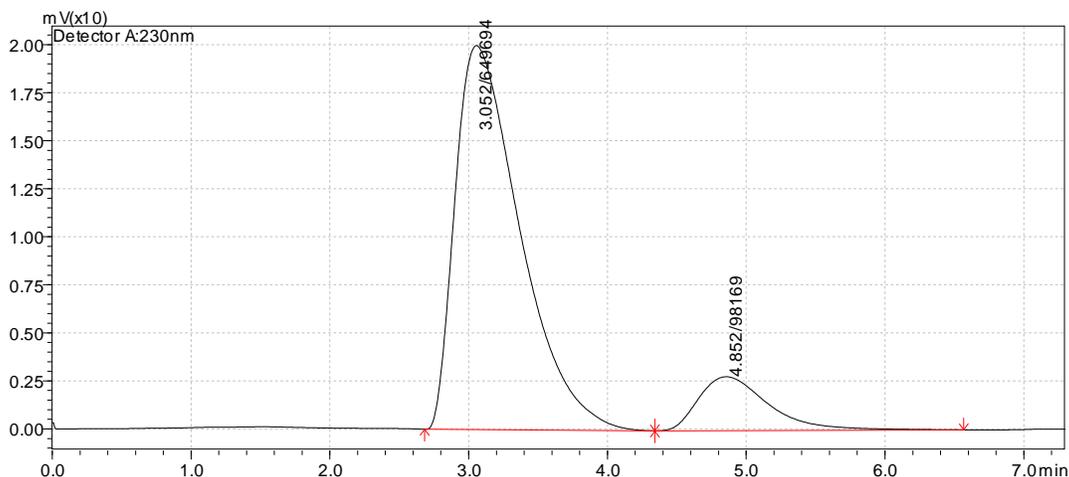
Figure 1: Chromatogram obtained from paracetamol RS and aspirin RS

Table 1: Linear Regression Data for Calibration Curves

Drugs	Paracetamol	Aspirin
Slope	105412.4	105261.2
Intercept	-1254.0	-1332.2
Correlation coefficient (R^2)	0.9905	0.9925
Limit of quantitation (µg/ml)	0.10	0.20
Limit of detection (µg/ml)	0.02	0.05

Table 2: Values of S_d and RSD as Confirmation of Precision

Paracetamol		Aspirin	
Amount claimed (mg/tablet)	Amount found (mg/tablet)	Amount claimed (mg/tablet)	Amount found (mg/tablet)
100.0	99.54	100.0	100.5
	99.85		99.24
	100.2		99.51
	100.1		100.1
	99.24		99.72
	99.30		100.4
Mean	99.71	Mean	99.91
S_d	0.407	S_d	0.504
%RSD	0.41	%RSD	0.50

CONCLUSION

To summarize, the method was evaluated in a mass of facets, such as best condition, linear relation including coefficient of correlation, accuracy, specificity and precision. The proposed method gives a good resolution between paracetamol and aspirin within a short analysis time and can

be conveniently adopted for routine quality control analysis.

REFERENCES

[1] Sweetman S, The Complete Drug Reference, 33rd Edition, Pharmaceutical Press, London, 2002, 356.

[2] Hardmann J and Limbird L, Goodman & Gilman's, The Pharmacological Basis of Therapeutics, 9th Edition, McGraw-Hill Companies, New York, USA, 1996, 124.

[3] Ramos-Martos N, Aguirre-Gomez F, Molinz-Diaz A and Capitan-Vallvey L, Application of liquid chromatography for the simultaneous determination of acetylsalicylic acid, caffeine, codeine, paracetamol, pyridoxine and thiamine in pharmaceutical preparations, J. AOAC Int., 84, 2001, 676-683.

[4] Vidal A, Reyers J, Barrales P and Diaz A, UV-spectrophotometric flowthrough multiparameter sensor for the simultaneous determination of acetaminophen, acetylsalicylic acid and caffeine, Anal. Letter, 35, 2002, 2433-2447.

[5] Martos N, Diaz A, Navalon A and Capitan-Vallvey L, Spectrofluorimetric determination of acetylsalicylic acid and codeine mixtures in pharmaceuticals, Anal. Letter, 34, 2001, 579-595.

[6] Szostak R and Mazurec S, Quantitative determination of acetylsalicylic acid and acetaminophen in tablets by FT-Raman spectroscopy, Analyst, 127, 2002, 144-148.

[7] Cemal A, Ismail T, Ahmet A, Yalcin O and Huamettin G, Rapid and simultaneous determination of acetylsalicylic acid, paracetamol, and their degradation and toxic impurity products by HPLC in pharmaceutical dosage forms, Turk. J. Med. Sci., 38, 2008, 131-173.

[8] Abu-Qare AW and Abou-Donia MB, A validated HPLC method for the determination of pyridostigmine bromide, acetaminophen, acetylsalicylic acid and caffeine in rat plasma and urine, J. Pharm. Biomed. Anal., 26, 2008, 939-947.

[9] Gondalia R, Mashru R and Savaliya P, Development and validation of spectrophotometric methods for simultaneous estimation of ibuprofen and paracetamol in soft gelatin capsule by simultaneous equation method, Int. J. Chem. Tech. Res., 2, 2010, 1881-1885.