RESEARCH ARTICLE

# Spectrophotometric estimation of ambroxol hydrochloride and cetirizine hydrochloride in tablets

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A simple, rapid, accurate, precise and reproducible simultaneous equation method has been developed for simultaneous estimation of ambroxol hydrochloride and cetirizine hydrochloride in tablets. Ambroxol hydrochloride has absorbance maxima at 243 nm, while cetirizine hydrochloride has absorbance maxima at 229 nm in glass-distilled water. The method developed involves no separation or extraction process. The proposed methods were successfully applied to the determination of ambroxol hydrochloride and cetirizine hydrochloride in tablets, with high percentage of recovery, good accuracy, and acceptable precision. Different analytical performance parameters such as linearity, precision, accuracy, limit of detection, limit of quantitation, and robustness were determined according to International Conference on Harmonization ICH Q2B guidelines. Results of analysis of formulation given as percentage of label claim ± relative standard deviation were found to be 99.27 ± 0.8083 and 102.43 ± 1.5357 for ambroxol hydrochloride and cetirizine hydrochloride respectively. Results of recovery studies given as percentage of label claim ± relative standard deviation were found to be 99.88 ± 0.3811 and 100.36 ± 2.0480 for ambroxol hydrochloride and cetirizine hydrochloride respectively.

***Key words:*** *Ambroxol hydrochloride, cetirizine hydrochloride, simultaneous equation*

## INTRODUCTION

Ambroxol hydrochloride is semisynthetic derivative of vasicine obtained from Indian shrub *Adhatoda vasica*. It is the metabolic product of bromhexine. It is official in Martin Dale-The Extra Pharmacopoeia.[1] Chemically it is trans-4-(2-amino-3,5-dibromobenzylamino) cyclohexanol hydrochloride. It acts as a bronchosecretolytic and expectorant drug. It stimulates the transportation of the viscous secretions in the respiratory organs and reduces the accumulation of the secretions.

Several spectrophotometric methods[2,3] have been used for the qualitative and quantitative determination of ambroxol hydrochloride in pharmaceutical formulations. Different high-performance liquid chromatographic (HPLC)[4,5] methods have been reported for determination of ambroxol hydrochloride in pharmaceutical formulations and biological fluids. Cetirizine is the carboxylated metabolite of hydroxyzine, and it has high specific affinity for histamine $H_1$ receptor. Cetirizine is chemically known as 2-[4-(4-chlorobenzhydryl) piperazine-1-yl] ethoxy acetic acid. Several spectrophotometric (Prakash MS *et al.*, 2000; Ramesh KC *et al.*, 2002; Ming M *et al.*, 2007; Azhagvunel S *et al.*, 2007) methods have been reported for determination of cetirizine in pharmaceutical formulations and in human plasma. Different HPLC methods[6] (Walily *et al.*, 1998) have been reported for determination of cetirizine in pharmaceutical formulations and biological fluids. Literature survey reveals that high-performance thin-layer chromatography has been reported for the simultaneous determination of ambroxol hydrochloride and cetirizine hydrochloride in pharmaceutical formulations.[7]

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## MATERIALS AND METHODS

### Apparatus

A PC-based Jasco V-530 recording spectrophotometer with spectral bandwidth of 2 nm and wavelength accuracy ±0.5 nm (with automatic wavelength correction) was employed for all measurements using a matched pair of 10-mm quartz cells. Shimadzu AY 120 analytical balance was used for weighing.

### Reagents

Ambroxol hydrochloride was procured from Litaka Pharmaceuticals, Pune (India), and cetirizine hydrochloride was obtained as gift sample from

Mediorals, Satara (India). All solvents and other reagents used in the spectrophotometric analysis were of analytical grade.

## Experimental

Standard stock solution containing ambroxol hydrochloride (AM) and cetirizine hydrochloride (CE) was prepared by dissolving 10 mg of AM and CE separately in 20 mL of methanol; and then final volume of both the solutions was made up to 100 mL with glass-distilled water to get stock solution containing 100 µg $mL^{-1}$ of AM and 100 µg $mL^{-1}$ of CE in two different 100-mL volumetric flasks. Solutions containing 10 µg $mL^{-1}$ of AM and CE were prepared and scanned in the UV region separately. The wavelengths selected were 229 and 243 nm for simultaneous determination of AM and CE.

By appropriate dilution of standard drug solutions with glass-distilled water, six working standard solutions containing 10, 15, 20, 25, 30, and 35 µg $mL^{-1}$ of AM and CE were prepared separately and scanned in the range of 200 to 350 nm. The values of absorbance were recorded at the selected wavelengths, and the absorptivity and molar absorptivity values were determined for AM and CE. The absorptivity values for AM and CE are given in Table 1. Molar absorptivity values determined for AM at 229 and 243 nm were 7731.75 and 10,370.8 $cm^{-1}$ $mol^{-1}$ $lit^{-1}$, while respective values for CE at 229 and 243 nm were 14,359.33 and 2217.92 $cm^{-1}$ $mol^{-1}$ $lit^{-1}$. Molecular weight of AM and CE is 414.6 and 439.89 respectively.

$$A_1 = 14359.33\ C_1 + 7731.75\ C_2 \quad (1)$$

and

$$A_2 = 2217.92\ C_1 + 10370.8\ C_2 \quad (2)$$

where $A_1$ and $A_2$ are the values of absorbance of sample at 229 and 243 nm respectively, and $C_1$ and $C_2$ are concentrations of CE and AM in moles $lit^{-1}$ respectively. Overlay spectra of AM and CE is given as Figure 1.

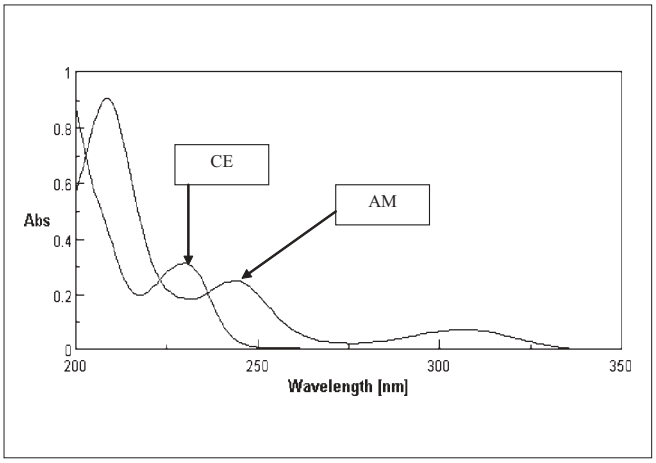


**Figure 1:** Overlay spectra of AM and CE

### Preparation of mixed standard solutions

Each marketed tablet formulation of the two drugs contains AM 60 mg and CE 5 mg. The standard stock solution of AM and CE was used to prepare seven mixed standards in the concentration range of 30 to 48 µg $mL^{-1}$ and 7.5 to 12 µg $mL^{-1}$ for ambroxol hydrochloride and cetirizine hydrochloride as given in Table 2; and results of analysis of mixed standards are shown in Table 3.

### Sample preparation

Marketed tablet formulations containing 60 mg of AM and 5 mg of CE were analyzed by this method. From the triturate of 20 tablets, an amount equivalent to 30 mg of AM and 2.5 mg of CE was weighed and transferred to 100-mL volumetric flask. Five milligrams of pure CE was added to the volumetric flask. The contents of the flask were dissolved in the 60 mL of the solvent with the aid of ultrasonication for 10 min. The solution was filtered through Whatmann filter paper no. 41, and then final volume of the solution was made up to 100 mL with glass-distilled water to get a stock solution containing 300 µg $mL^{-1}$ of AM and 75 µg $mL^{-1}$ of CE. After appropriate dilutions, the absorbances were measured, and the concentration of each analyte was determined using the equations generated. The statistical data obtained after replicate determinations ($n$ = 9) is shown in Table 4.

**Table 1: Absorptivity values.**

| Conc. (µg $ml^{-1}$) | Absorptivity for AM | | Absorptivity for CE | |
|---|---|---|---|---|
| | 229 nm | 243 nm | 229 nm | 243 nm |
| 10 | 187.29 | 250.3 | 330 | 51.58 |
| 15 | 187.44 | 249.63 | 325.06 | 51.29 |
| 20 | 186.13 | 250.5 | 326.86 | 49.92 |
| 25 | 185.62 | 249.67 | 324.64 | 50.85 |
| 30 | 186.73 | 250.35 | 325.13 | 49.14 |
| 35 | 185.43 | 250.39 | 326.89 | 49.74 |
| Mean | 186.49 | 250.14 | 326.43 | 50.42 |
| S.D. | 0.9241 | 0.3854 | 1.9956 | 0.9631 |
| R.S.D. | 0.4955 | 0.1540 | 0.6113 | 1.9102 |
| M.A.* | 7731.75 | 10370.8 | 14359.33 | 2217.92 |

*M.A. is molar absorptivity in $cm^{-1}$ $moles^{-1}$ $lit^{-1}$ determined from mean value of absorptivity

**Table 2: Concentration of mixed standard solutions.**

| Standard no. | 1 | 2 | 3 | 4 | 5 | 6 | 7 |
|---|---|---|---|---|---|---|---|
| Concentration of AM (µg $ml^{-1}$) | 30 | 33 | 36 | 39 | 42 | 45 | 48 |
| Concentration of CE (µg $ml^{-1}$) | 7.5 | 8.25 | 9.0 | 9.75 | 10.5 | 11.2 | 12.0 |

**Table 3: Results of analysis of mixed standards.**

| Analyte | % Concentration estimated* (Mean ± S.D.) | R.S.D. |
|---|---|---|
| AM | 100.23 ± 1.2283 | 1.2254 |
| CE | 101.23 ± 2.1855 | 2.1587 |

*Average of seven determinations; R.S.D.: relative standard deviation

### Recovery study

Accuracy and sensitivity of analysis were determined by performing recovery studies by spiking different concentrations of pure drug in the preanalyzed tablet sample. Results of recovery studies indicated that the method is rapid, accurate, and reproducible. Results of recovery studies are shown in Table 5.

### Method validation

The proposed method was validated according to ICH Q2B guidelines for validation of analytical procedures in order to determine accuracy, precision, repeatability, robustness, limit of detection, and quantitation. Results are shown in Tables 5 to 10.

## RESULTS AND DISCUSSION

Beer's law was obeyed in the concentration range of 5 to 50 µg $mL^{-1}$ and 7.5 to 75 µg $mL^{-1}$ for AM and CE respectively. For both the drugs, seven mixed standards were prepared. Interday and intraday studies showed high degree of repeatability of this analytical method under normal operating conditions. Results of tablet analysis showed relative standard deviation in the range of 0.3156 to 1.4244 and of 0.6729 to 2.5371 for AM and CE respectively, which indicates repeatability of the method.

The accuracy of the method was determined by investigating the recovery of the two drugs using spiked concentrations of the standard drug. The results indicated excellent recoveries - ranging from 99.13% to 101.23% and 98.22% to 102.33%, with a mean of 99.88% and 100.36%, for the two drugs AM and CE respectively. Recoveries obtained for the drugs do not differ significantly from 100%, which showed that there was no interference from the common excipients used in the tablet formulation, indicating accuracy and reliability of the method. Precision was determined by analysis of tablets containing AM and CE.

## CONCLUSION

The method developed is simple, accurate, rapid, and reproducible. Simultaneous equation method may be used for routine quality-control analysis of investigated drugs in two-component pharmaceutical preparations.

**Table 4: Results of tablet analysis.**

| Analyte | Label claim (mg) | % Label claim estimated* (Mean ± S. D.) | R.S.D. |
|---|---|---|---|
| AM | 60 | 99.27 ± 0.8024 | 0.8083 |
| CE | 5 | 102.43 ± 1.5731 | 1.5357 |

*Average of nine determinations; S.D.: standard deviation; R.S.D.: relative standard deviation

**Table 5: Results of recovery study.**

| Analyte | Label claim (mg) | % Label claim estimated* (Mean ± S. D.) | R.S.D. |
|---|---|---|---|
| AM | 60 | 99.88 ± 0.3807 | 0.3811 |
| CE | 5 | 100.36 ± 2.0555 | 2.0480 |

*Average of nine determinations; S.D.: standard deviation; R.S.D.: relative standard deviation

**Table 6: Results of repeatability.**

| Analyte | Label claim (mg) | Tablet analysis % label claim estimated* (Mean ± R.S.D.) | Recovery study % label claim estimated* (Mean ± R.S.D.) |
|---|---|---|---|
| AM | 60 | 99.15 ± 0.7150 | 0.7211 |
| CE | 5 | 101.45 ± 1.3135 | 1.2946 |

*Average of nine determinations; R.S.D.: relative standard deviation

**Table 7: Limit of detection and limit of quantitation.**

| LOD (µg $ml^{-1}$)* | | LOQ (µg $ml^{-1}$)* | |
|---|---|---|---|
| AM | CE | AM | CE |
| 0.0234 | 0.0241 | 0.0783 | 0.0806 |

*Average of nine determinations; R.S.D.: relative standard deviation

**Table 8: Results of robustness using saline solution.**

| Analyte | Label claim (mg) | Tablet Analysis % label claim estimated* (Mean ± R.S.D.) | Recovery study % label claim estimated* (Mean ± R.S.D.) |
|---|---|---|---|
| AM | 60 | 101.21 ± 0.4164 | 99.48 ± 0.8573 |
| CE | 5 | 99.84 ± 1.3157 | 101.75 ± 1.5798 |

*Average of nine determinations; R.S.D.: relative standard deviation

**Table 9: Results of robustness using syrup solution.**

| Analyte | Label claim (mg) | Tablet Analysis % label claim estimated* (Mean ± R.S.D.) | Recovery study % label claim estimated* (Mean ± R.S.D.) |
|---|---|---|---|
| AM | 60 | 101.21 ± 0.3250 | 101.06 ± 0.3696 |
| CE | 5 | 99.74 ± 1.9382 | 98.75 ± 2.5371 |

*Average of nine determinations; R.S.D.: relative standard deviation

**Table 10: Results of interday and intraday precision.**

| Day | Label claim estimated (%)* (Mean ± %R.S.D.) | | Recovery estimated (%)* (Mean ± %R.S.D.) | |
|---|---|---|---|---|
| | AM | CE | AM | CE |
| Day 1 | 99.98 ± 1.1334 | 101.46 ± 0.9594 | 99.93 ± 1.355 | 101.46 ± 1.4619 |
| Day 2 | 101.68 ± 0.4312 | 98.94 ± 0.6877 | 99.60 ± 1.4244 | 102.06 ± 1.2292 |
| | 102.55 ± 0.8896 | 99.66 ± 1.9747 | 101.23 ± 0.3156 | 102.22 ± 0.6729 |

*Average of nine determinations; R.S.D.: relative standard deviation

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