Pergamon

Journal of Pharmaceutical & Biomedical Analysis, Vol. 12, No. 6, pp. 747–752, 1994

0731–7085/94 $7.00 + 0.00

0731–7085(93)E0025-I

Determination of guaiphenesin in anti-tussive pharmaceutical preparations containing dextromethorphan by first- and second-derivative ultraviolet spectrophotometry

A.R. LEE* and T.M. HU

School of Pharmacy, National Defense Medical Center, Taipei, Taiwan, Republic of China

Abstract: Rapid, simple and direct assay procedures based on selective first ($D1$)- and second ($D2$)-derivative spectrophotometry, using a zero-crossing technique of measurement at 279.2 and 280.0 nm, respectively, have been developed for the specific determination of guaiphenesin in the presence of dextromethorphan, drugs with closely overlapping absorption spectra, in synthetic admixtures and in pharmaceutical dosage forms (tablets and syrups). The methods do not require extraction with organic solvents and are easier to perform than their conventional counterparts. Calibration graphs were linear (r = 0.99999 for $D1$ and 0.99969 for $D2$, respectively). Good selectivity, accuracy and precision were found. However, the performance of the analysis of guaiphenesin by the second-derivative mode deteriorated when the ratio of dextromethorphan to guaiphenesin was greater than one. Thus, the first-derivative spectrophotometry is the method of choice for the assay of tablets and syrups containing the two drugs.

Keywords: *Guaiphenesin; dextromethorphan; first-derivative ultraviolet spectrophotometry; second-derivative ultraviolet spectrophotometry; zero-crossing technique; anti-tussive pharmaceutical preparations.*

Introduction

Guaiphenesin (guaifenisin) [3-(2-methoxyphenoxy)-1,2-propanediol] is a sedative expectorant that acts by stimulating gastric reflexes through stomach irritation; this action helps to reduce the viscosity of tenacious sputum and to thin bronchial secretions. Guaiphenesin is widely used alone or with dextromethorphan, for the symptomatic treatment of coughs due to acute or chronic bronchitis and bronchial allergic conditions, in commercially available anti-tussive products [1].

Official analytical methods in the British Pharmacopoeia (1988) [2] and the USP (1980, 1990) [3, 4] for the quantitation of guaiphenesin in the bulk drug and in dosage forms include titrimetric, spectrophotometric, gas chromatographic (GC), and high-performance liquid chromatographic (HPLC) procedures. In compound formulations, guaiphenesin has been determined by difference spectrophotometry [5], second- [6] and third- [7] derivative spectrophotometry, colorimetry [8], densitometry [9], GC [10] and HPLC [11–13]. Some of these methods are not specific for guaiphenesin and some require extensive sample manipulation. Guaiphenesin in compound preparations cannot be directly determined by conventional spectrophotometric methods because of the significant overlapping in spectra. By conventional ultraviolet–visible (UV–vis) absorption methods, a laborious extraction procedure is generally required to separate guaiphenesin from other components prior to quantitative measurement (USP XX, 1980) [3].

The derivative technique in UV–vis spectrophotometry offers a powerful approach for the enhancement of sensitivity and specificity. It has frequently been employed to overcome the problem of interference due to irrelevant spectral overlapping, which may be caused either by substances other than analytes or by excipient matrices commonly present in pharmaceutical formulations. In derivative spectroscopy, the conventional spectrum is rapidly and simply transformed to a function of its first derivative ($dA/d\lambda$) or more usefully to the second ($d^2A/d\lambda^2$) or higher derivatives, by an analogue or digital device as the spectrum is scanned. Consequently, fine structural features are sharpened to give improved resolution of overlapping peaks and potentially greater

* Author to whom correspondence should be addressed.

sensitivity. More detailed information that deals with various methods for the generation of derivative spectra has been presented [14] and applications of the technique to hypothetical situations and possible systematic and random errors were elegantly established by O'Haver and Green [15]. In the last 15 years, this technique has rapidly gained applications in the fields of pharmaceutical analysis, biochemistry, analytical chemistry and clinical chemistry [16].

The described first- and second-derivative UV spectrophotometric methods are simple, direct and accurate, and require no extraction procedures or organic solvents for the determination of guaiphenesin in the presence of dextromethorphan in synthetic admixtures and in anti-tussive formulations.

Experimental

Materials

Authentic standards of guaiphenesin and dextromethorphan were kindly provided by Veterans Pharmaceutical Plant (Taiwan). Commercial tablets and syrup were either provided by Veterans Pharmaceutical Plant (Decofen tablets and syrup) or purchased from A.H. Robins Co. (Australia; Robitussin-DM syrup).

Apparatus

Ordinary and derivative spectrophotometric analyses were performed on a Shimadzu UV-160 UV–vis recording spectrophotometer with standard 10-mm quartz semimicro cuvettes. Absorption spectra of samples were recorded at a scan speed of about 480 nm min^{-1} between 230 and 300 nm. The first-derivative spectra were recorded with a $\Delta\lambda = 7.2$ nm. The second-derivative spectra were recorded with a $\Delta\lambda = 3.5$ nm.

Calibration procedure

Aliquots of the freshly prepared stock solution of guaiphenesin (1 mg ml^{-1}) were diluted in distilled water. The first- and second-derivative spectra against water as a blank were then recorded over the concentration range of 10–100 μg ml^{-1} of guaiphenesin. The calibration graph was constructed by plotting the amplitude heights measured on the chart paper against the corresponding concentrations. The absolute values of the derivative were obtained by a zero-crossing technique with measurements at 272.9 and 280.0 nm for the first- and the second-derivative, respectively.

Sample analysis

Derivative spectrophotometry. The formulations containing a nominal amount of 100 mg of guaiphenesin and 15 mg of dextromethorphan per tablet or per 5 ml of syrup were analysed. At least 20 tablets were weighed and thoroughly powdered, and an accurately weighed portion of the powder (or an accurately measured volume of syrup) equivalent to about 100 mg of guaiphenesin was transferred into a 100-ml calibrated flask and dispersed in 50 ml of water by ultrasonic vibration for 5 min. The resulting suspension was diluted to volume with distilled water. The contents of the flasks were gently shaken by hand and allowed to settle and then filtered through a filter-paper. A 5-ml aliquot of the clear supernatant was diluted to 100 ml with distilled water. Then the assay was carried out according to the methods described above. The resulting first- and second-derivative spectra were recorded against water as a blank. By reference to the calibration graph the concentration of guaiphenesin in each sample was calculated. In order to establish the reliability of the proposed method, synthetic admixtures containing known amounts of pure drugs and excipients corresponding to the commercial products were similarly analysed.

Conventional spectrophotometry. The procedures for analysis of the tablets by conventional spectrophotometry were followed as directed in the assay of Guaifenesin Tablets, USP XX (1980) but with a slight modification; 5 ml of diluted H_2SO_4 was used instead of 10 ml of saturated sodium bicarbonate solution and washing was carried out with water instead of 1 M hydrochloric acid, after extraction with chloroform. For analysis of the syrups, the procedures were more complicated because of the necessity for pretreatment by extraction with four 25-ml portions of *n*-hexane and then by extracting the aqueous layer by six portions of chloroform. The remaining procedures were similar to those described for the analysis of tablets.

Results and Discussion

The UV spectra (zero-order) of the aqueous solutions of guaiphenesin and dextromethor-

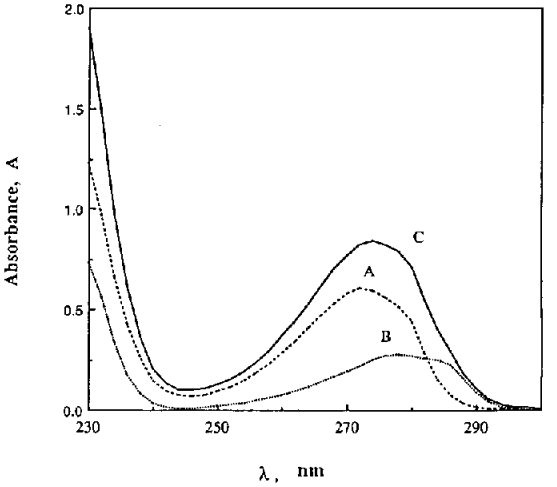

Figure 1
Zero-order spectra of 50 μg ml^{-1} guaiphenesin (A) and 50 μg ml^{-1} dextromethorphan (B) and their 1:1 admixture (C) in distilled water.

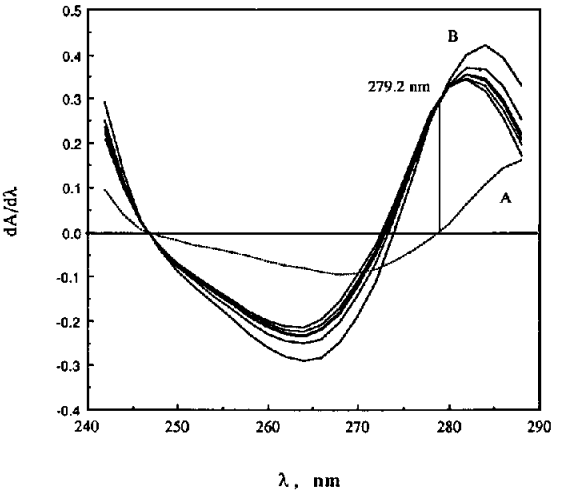

Figure 2
First-derivative spectra of 50 μg ml^{-1} dextromethorphan (A) and 50 μg ml^{-1} guaiphenesin in the presence of 0–50 μg ml^{-1} dextromethorphan (B) in distilled water.

phan and their admixture (1:1) in the region of 230–300 nm (Fig. 1) show absorption maxima at 272.4 and 277.4 nm, respectively. The similar behaviour of these two compounds makes the determination of guaiphenesin in the presence of dextromethorphan by conventional UV spectrophotometry extremely difficult. This problem has been solved satisfactorily by derivative spectrophotometry. When derivative UV spectra were recorded, sharp bands of large amplitudes of guaiphenesin were produced, which may offer more selective identification and specific determination of this drug. Figures 2 and 3 show the first- and the second-derivative spectra of guiaphenesin and dextromethorphan, respectively. Due to the zero-crossing characteristics of dextromethorphan at 279.2 nm for the first-derivative and at 272.4 and 280.0 nm for the second-derivative mode, the spectra of guaiphenesin show the same amplitudes in the presence of several different concentrations of dextromethorphan at these points. Accordingly, the analysis of guaiphenesin at these particular wavelengths is independent of the presence of dextromethorphan. In this study, the amplitudes at 279.2 and 280.0 nm for the first- and the second-derivative spectra were chosen for the assay of guaiphenesin.

Linearity

The linear regression equation and the statistical evaluation of the calibration plots for the analysis of authentic samples are listed in

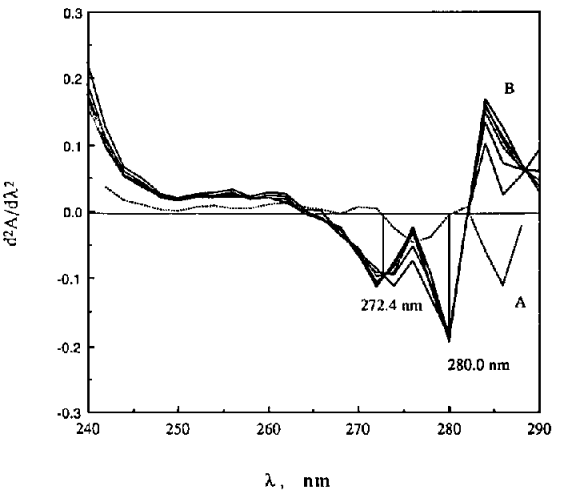

Figure 3
Second-derivative spectra of 50 μg ml^{-1} dextromethorphan (A) and 50 μg ml^{-1} guaiphenesin in the presence of 0–50 μg ml^{-1} dextromethorphan (B) in distilled water.

Table 1. Under the described experimental conditions, linear correlations were obtained between the zero-crossing amplitudes ($D_{279.2nm}$ and $D_{280.0nm}$ for the first- and the second-derivative spectra, respectively) at the chosen wavelengths over the concentration range of 10–100 μg ml^{-1} of guaiphenesin. The calculated correlation coefficients (r) of least-squares linear regression were 0.99999 for the first- and 0.99969 for the second-derivative method, respectively. The lower limits of detection (LOD) [17], calculated by the expression LOD = 3 Sb/b (where b is the slope of the calibration graph and Sb is the standard

Table 1
Statistical analysis of calibration plots for the determination of guaiphenesin by first- and second-derivative UV spectrophotometry*

Derivative mode	Signal measured (nm)	Regression parameters†	Correlation coefficient (r)	SD‡ ($\mu g\ ml^{-1}$)	LOD§ ($\mu g\ ml^{-1}$)
First-derivative ($D1$)	279.2	$a = 2.63 \times 10^{-4}$ $b = 5.74 \times 10^{-3}$	0.99999	$Sa = 6.70 \times 10^{-4}$ $Sb = 9.42 \times 10^{-6}$	0.35
Second-derivative ($D2$)	280.0	$a = 5.46 \times 10^{-3}$ $b = 3.43 \times 10^{-3}$	0.99969	$Sa = 2.16 \times 10^{-3}$ $Sb = 3.03 \times 10^{-5}$	1.89

*Concentration of standard samples ranged from 10 to 100 $\mu g\ ml^{-1}$; $n = 6$.
†$D = a + b\ C$, where D is the derivative amplitude and C is the concentration of the drug ($\mu g\ ml^{-1}$).
‡Standard deviation of the intercept (Sa) and slope (Sb).
§Lower limit of detection.

Table 2
Determination of guaiphenesin (G) at six concentration levels of admixtures containing 10 $\mu g\ ml^{-1}$ of dextromethorphan (D) by first and second-derivative UV spectrophotometry ($n = 6$)

		First-derivative λ = 279.2 nm			Second-derivative λ = 280.0 nm		
G/D ratio	Added ($\mu g\ ml^{-1}$)	Found ($\mu g\ ml^{-1}$)	Recovery (%)	RSD (%)	Found ($\mu g\ ml^{-1}$)	Recovery (%)	RSD (%)
1	10.0	9.91	99.1	0.24	9.16	91.6	1.10
2	20.0	20.2	100.8	0.41	19.9	98.6	0.94
3	30.0	30.1	100.2	0.25	29.6	98.6	0.72
4	40.0	40.1	100.2	0.29	39.8	99.5	0.46
5	50.0	50.2	100.3	0.08	49.8	99.6	0.34
10	100.0	100.1	100.1	0.23	98.3	98.3	0.46

deviation of the slope) were 0.35 and 1.89 $\mu g\ ml^{-1}$ for each method, indicating the excellent sensitivity of the results.

Selectivity

The interference from dextromethorphan in the determination of guaiphenesin was investigated. Six sets of solutions containing 10 $\mu g\ ml^{-1}$ of dextromethorphan and spiked with various amounts of guaiphenesin in the range of 10–100 $\mu g\ ml^{-1}$ were prepared and determined against a standard solution of guaiphenesin. The mean recoveries of guaiphenesin at different guaiphenesin (G)/dextromethorphan (D) ratios are given in Table 2. The recoveries for guaiphenesin are almost quantitative for the first-derivative mode in most G/D ratios and for the second derivative mode where G/D ≥ 1. In addition, the relative standard deviations (RSD) for six determinations at each concentration level are satisfactorily low (0.08–0.41% for the first-derivative, 0.34–1.10% for second-derivative) and indicate the good reproducibility of the proposed methods. However, the analysis of guaiphenesin in the second-derivative mode was subject to interference in the presence of a larger amount of dextromethorphan, i.e. with a G/D ratio ≤ 1. The interference was obviously associated with the proportion of dextromethorphan in the mixtures.

The feasibility of the determination of dextromethorphan in the presence of guaiphenesin was also investigated by a similar experiment. Six sets of solutions containing 10–100 $\mu g\ ml^{-1}$ of dextromethorphan and 10 $\mu g\ ml^{-1}$ of guaiphenesin with G/D ratios of 0.1–1 were assayed for dextromethorphan by the first-derivative mode at the guaiphenesin

Table 3
Determination of dextromethorphan (D) at six concentration levels in mixtures containing 10 $\mu g\ ml^{-1}$ of guaiphenesin (G) by first-derivative UV spectrophotometry ($n = 6$)

	First-derivative λ = 272.7 nm			
G/D ratio	Added ($\mu g\ ml^{-1}$)	Found ($\mu g\ ml^{-1}$)	Recovery (%)	RSD (%)
1	10.0	11.2	112.4	1.58
0.5	20.0	20.5	102.3	1.99
0.33	30.0	30.5	101.7	0.42
0.25	40.0	40.2	100.4	1.00
0.2	50.0	49.7	99.5	0.43
0.1	100.0	97.0	97.0	0.33

Table 4
Assay of guaiphenesin in commercail products by derivative and conventional absorption UV spectrophotometry. Results are the mean (RSD) of three determinations

Products*	First-derivative (%)	Second-derivative (%)	Conventional (%)
Decofen tablets	98.2 (0.9)	99.4 (0.5)	96.8 (0.8)
Decofen syrup	99.2 (0.5)	97.6 (0.8)	96.3 (1.0)
Robitussin-DM syrup	102.1 (0.8)	101.8 (0.4)	97.5 (0.8)

* Labelled to contain 100 mg of guaiphenesin and 15 mg of dextromethorphan per tablet or 5 ml of syrup.

zero-crossing point, 272.7 nm. The results are listed in Table 3. The data clearly indicate that the quantitative determination of dextromethorphan in the presence of guaiphenesin is best for a G/D ratio of about 0.1–0.5. The performance of the analysis deteriorates as the G/D ratio extends beyond this range. Since guaiphenesin is the major component in commercial products quantitatively and the G/D ratio is far greater than 1 (100:15), the proposed methods can only be applied successfully to the selective analysis of guaiphenesin.

Accuracy and precision

The accuracy and precision of these methods were verified by means of a recovery assay for the synthetic admixtures containing known amounts of drug and blank excipients based on the composition of commercial products. Recoveries were determined and compared with those of the corresponding standard solutions; values of 101.5 ± 1.3% for the first-derivative method and 97.8 ± 0.8% for the second-derivative method were obtained (n = 6).

The comparative results for the determination of guaiphenesin in commercial tablet and syrup preparations by derivative and conventional absorption spectrophotometry are given in Table 4. Since an extraction procedure was employed in the conventional methods to overcome interference from dextromethorphan and excipients, they were considered to be reliable reference methods. The results from both methods are comparable. Therefore, this work provides further evidence that the proposed derivative spectrophotometric methods are highly accurate and precise and can be used as alternatives in the analysis of guaiphenesin.

Conclusions

The use of UV derivative spectrophotometry permits powerful separation of guaiphenesin from sources of interference by mathematical means instead of by extraction or chromatographic procedures. The proposed methods are simple, rapid and show good selectivity, accuracy and precision. Prior sample preparation, such as solvent extraction or column chromatography, is not necessary for the analysis of these formulations. Elimination of these steps saves considerable analysis time and produces a mathematical separation within 5 min. It is worthy of mention that no expensive or toxic solvents or reagents are used; thus these methods have a significant advantage over other techniques and are extremely suitable in routine pharmaceutical analyses. As the performance of the analysis of guaiphenesin by the second-derivative mode deteriorates if the ratio of dextromethorphan to guaiphenesin is greater than one, it is suggested that first-derivative spectrophotometry is the method of choice for the assay of the tablets and syrup.

Acknowledgement — The authors thank Veterans Pharmaceutical Plant (Taiwan) for kindly supplying standards of guaiphenesin and dextromethorphan in addition to free samples of Decofen tablets and syrup.

References

[1] J.E.F. Reynolds (Ed.), *Martindale The Extra Pharmacopoeia*, 29th edn, p. 910. The Pharmaceutical Press, London (1989).
[2] *British Pharmacopoeia*, HMSO, London (1988).
[3] *The United States Pharmacopeia, XX Rev.*, The National Formulary XV edn, The US Pharmacopeial Convention, Inc. (1990).
[4] *United States Pharmacopeia, XXII Rev.*, The National Formulary XVVII edn, The US Pharmacopeial Convention, Inc. (1980).
[5] H.S.I. Tan and G.C. Salvador, *Anal. Chim. Acta* **176**, 71–76 (1985).
[6] Q. Yang, Y. Meng and G. Zhang, *Yaowu Fenxi Zazhi* **4**, 148–150 (1984).
[7] M.H. Abdel-Hay, M.S. El-Din and M.A. Abuirjeie, *Analyst* **117**, 157–160 (1992).
[8] G.R. Rao, A.B. Avadhanulu, R. Giridhar and C.K. Kokta, *East. Pharm.* **31**, 141–142 (1988).

[9] H. Tomankova and M. Vasatova, *Pharmazie* **44**, 197–198 (1989).
[10] M. Bambagiotti-Alberti, S. Pinzauti and F.F. Vincieri, *Pharm. Acta Helv.* **62**, 175–176 (1987).
[11] W.O. McSharroy and I.V.E. Savage, *J. Pharm. Sci.* **69**, 212–214 (1980).
[12] N. Muhammad and J.A. Bodnar, *J. Liq. Chromatogr.* **3**, 113–122 (1980).
[13] L. Carnevale, *J. Pharm. Sci.* **72**, 196–198 (1983).
[14] G. Talsky, H. Mayring and H. Kreuzer, *Angew. Chem. Int. Ed. Engl.* **17**, 785–799 (1978).
[15] T.C. O'Haver and G.L. Green, *Anal. Chem.* **48**, 312–318 (1976).
[16] A.R. Lee, *Chin. Pharm. J.* **44**, 87–96 (1992).
[17] G.L. Long and J.P. Winefordner, *Anal. Chem.* **55**, 712–724 (1983).

[*Received for review 3 August 1993; revised manuscript received 12 October 1993*]