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Notes

Simultaneous UV spectrophotometric determination of procaine hydrochloride and phenazone in an otic formulation

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Summary

A UV spectroscopic method for the simultaneous determination of procaine hydrochloride and phenazone in eardrops was developed. The individual concentrations were calculated by means of a previously described algorithm (using factors based on absorbance ratios) bundled to the computer-aided apparatus. The selected wavelengths were 290 and 242 nm, corresponding to the λ_{max} of procaine hydrochloride and phenazone, respectively. The method allows rapid and accurate determination of the binary mixture in the tested concentration range of 2–9 $\mu g\ ml^{-1}$ for procaine hydrochloride and 10–40 $\mu g\ ml^{-1}$ for phenazone, with a relative standard deviation of less than 1.8%. Second-order derivative spectroscopy and zero-crossing derivative methodology were tested but were found to be unsuitable in this case.

UV-visible spectrophotometry has been widely used in pharmaceutical analysis because of its rapidity, simplicity and applicability to a host of substances. Moreover, the simultaneous spectrophotometric determination of drugs in multicomponent systems is an interesting alternative to specific determination with other techniques such as GC or HPLC, insofar as the analytical procedure is simpler and quicker. However, the individual spectrophotometric analysis of a drug combination is often complicated by interference from the formulation matrix or from spectral overlapping.

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The problem of interference has been dealt with by means of several mathematical methods, including orthogonal polynomial assay (Wahbi and Unterhalt, 1976), differential spectrophotometry (Korany and Haller, 1982) and the least-squares method (Madsen et al., 1974; Wahbi et al., 1978). These methods, however, require special care in selecting experimental conditions and may prove to be of little use in practical application (Korany et al., 1984; El-Yazbi et al., 1986).

Only in recent years has the rapid progress in computer technology led to a revival of interest and considerable improvement in multicomponent UV spectrophotometric analysis (Sala et al., 1988). In fact, specific software provides the mathematical processing of spectral data for mixture deconvolution, allowing an accurate and more rapid application of the method. In any case, before a

multicomponent analysis can be performed, it must be verified that each drug obeys the Beer-Lambert law over the wavelength and concentration range of interest without mutual interference.

Because of its anaesthetic and anti-inflammatory action, a combination of procaine hydrochloride and phenazone is commercially available and widely used here in Italy, in spite of the controversy over its clinical effectiveness and safety in treating otic disorders (Miller, 1986). On the other hand, an antypirine and benzocaine otic solution in glycerol is described in the U.S.P. XXI (1985). Several methods have been reported for the determination of procaine hydrochloride (Clarke, 1986b) and phenazone (Clarke, 1986a) but none has been described for the quantitation of this binary mixture. In this work a multicomponent analysis method, based on a previously described algorithm (Moussa and Safwat, 1979; Parmentier et al., 1984), was applied to the simultaneous UV determination of procaine hydrochloride and phenazone in an otic solution, allowing resolution of the problems due to the overlapping spectral bands of the two drugs in the 295–200 nm region (Fig. 1), which makes their simultaneous determination by direct UV spectrophotometry difficult.

Chemicals: Procaine hydrochloride (PCN) and phenazone (PHZ) (Sigma) were used as received.

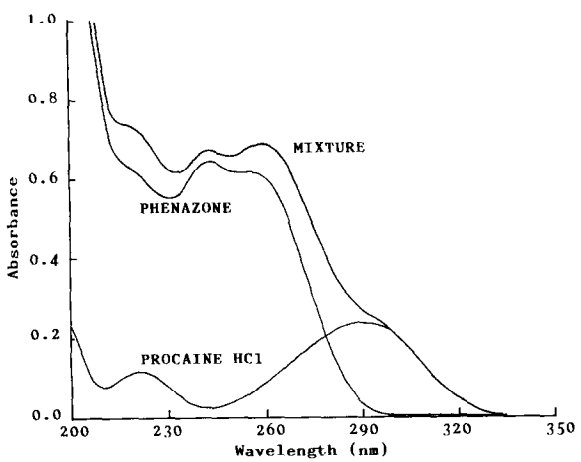

Fig. 1. UV spectra of aqueous solutions of procaine hydrochloride (3.4 $\mu g\ ml^{-1}$), phenazone (15.8 $\mu g\ ml^{-1}$) and their binary mixture.

Samples of five batches of an otic solution, manufactured for the Italian army and labelled to contain (w/w) 1.5% PCN, 7% PHZ and glycerin q.s., were obtained from the Stabilimento Chimico Farmaceutico Militare (Florence, Italy). Other reagents used were of analytical reagent grade and solutions were prepared in bidistilled water passed through a Milli-Q water purification system (Millipore).

Apparatus: A Varian DMS Model 200 UV-visible spectrophotometer equipped with the Moussa algorithm specific software (Moussa and Safwat, 1979) was used. The operating parameters were: 1 cm quartz cells; wavelength range 350–200 nm; slit width 2 nm; scan speed 200 nm min^{-1}. Second derivative spectra were recorded in triplicate using the same instrumental parameters and a response time of 6 s.

Solutions: Stock solutions of PCN (0.4 mg ml^{-1}) and PHZ (2 mg ml^{-1}) were prepared in water. Concentrations of working standard solutions, obtained by appropriate dilution with water, ranged from 2 to 9 $\mu g\ ml^{-1}$ for PCN and from 10 to 40 $\mu g\ ml^{-1}$ for PHZ. From these standard solutions, mixtures of the two components were prepared at various concentrations so that their ratio was always the same as that of the assayed dosage form (1.5 : 7).

Analysis of the otic product: 5 g of the eardrops were diluted to volume with water in a 100 ml volumetric flask; 1 ml of the resulting solution was transferred into a 100 ml volumetric flask and diluted to volume with water. The measured specific absorbances for PCN at 242 and 290 nm (59 and 679, respectively) and those for PHZ (486 and 19) were memorized by the specific software. The absorbance values at 290 and 242 nm were recorded and the concentration of each compound was directly obtained from the computer-aided instrument. In fact, the spectrophotometer bundled algorithm permitted the automatic solution of the following two-equation system (Moussa and Safwat, 1979; Parmentier et al., 1984):

$$C_x = A_1 - \beta A_2/(1 - \alpha\beta)\,a_1$$

$$C_y = A_2 - \alpha A_1/(1 - \alpha\beta)\,b_2$$

where C_x and C_y are the concentrations of PCN and PHZ, respectively, calculated as % w/v; A denotes the absorbance of the measured solution; a and b represent the values of specific absorbances for PCN and PHZ, respectively; the subscripts 1 and 2 denote the λ_{max} of components x and y (at 290 and 242 nm, respectively); α and β are constants derived by dividing the absorbance at λ_{max} of the other component by the absorbance at its λ_{max}; i.e. $\alpha = A_2^x/A_1^x$; $\beta = A_1^y/A_2^y$.

The accuracy of the results essentially depends on the sufficient difference of the absorption spectra of the components. The method is particularly reliable when, as in our case, the values of α and β are very small (Parmentier et al., 1984). The most suitable wavelengths for PCN and PHZ were observed to be 290 and 242 nm, respectively, which correspond to their λ_{max} (Fig. 1), where the absorptivity of the overlapping component is small. Standard curves were determined for each component. In both cases, plots of absorbance vs concentration were linear in the examined concentration ranges (2–9 μg ml^{-1} for PCN and 10–40 μg ml^{-1} for PHZ).

For both wavelengths considered, the sum of the absorbances of PCN and PHZ, determined separately, was compared with the absorbances of the two-component mixture at identical concentrations. At all examined concentrations, the data were in good agreement, confirming the actual additivity of the absorbances and the absence of interaction between the components which would affect their spectra.

There was good agreement between the concentration values of known mixtures and those calculated from the apparatus bundled software. The percent error, calculated from the measured concentration/theoretical concentration ratio, was always less than 1.5%.

Derivative spectrophotometry has been proved to be a useful means of resolving overlapping spectra (Fell, 1978) and it has been widely applied to the simultaneous determination of drugs in multicomponent dosage forms. This method was thus also tried in this study. The second-order derivative spectra of PCN and PHZ showed significant differences in some areas (Fig. 2); nevertheless, preliminary experiments showed that the

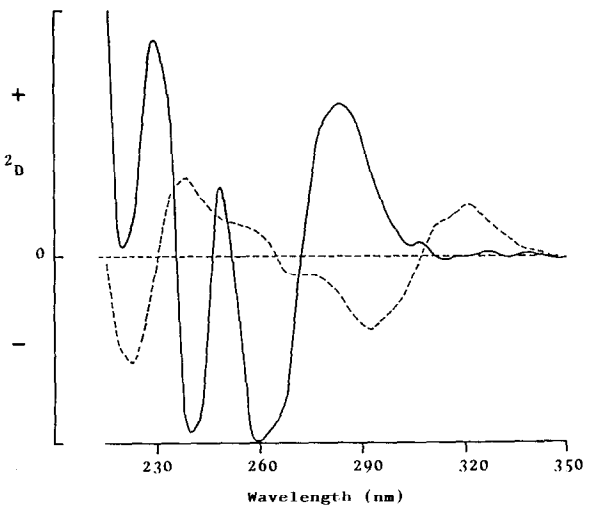

Fig. 2. Second-order derivative UV spectra of aqueous solutions of procaine hydrochloride (9.0 μg ml^{-1}, broken line) and phenazone (40.0 μg ml^{-1}, solid line).

enhancement of spectral detail obtained with the second-derivative mode was not sufficient to permit correct use of graphic measures of derivative amplitude, i.e., peak-to-peak or peak-to-derivative zero, owing to reciprocal interferences for all recorded peaks of both drugs. In similar cases, 'zero-crossing' second-derivative spectrophotometry may be useful (O'Haver and Green, 1976; O'Haver, 1979; Morelli, 1988). This involves the measurement of the absolute value of the total-derivative spectrum (sum curve) at a zero-crossing wavelength of the interfering component. Of these wavelengths, 231.5 and 322 nm were selected as optimal for the determination of PHZ and PCN, respectively. However, in triplicate scans, insufficient precision was obtained for the derivative signals at the selected working wavelengths. The significant error observed in repetitive determinations in the derivative mode has been discussed (O'Haver, 1979), and is probably due to a degradation of the signal-to-noise ratio. Moreover, the zero-crossing measures are particularly sensitive to small changes in the position of the interfering band (O'Haver and Green, 1976). Attempts simultaneously to analyze the PCN-PHZ mixture using the second derivative mode were thus unsuccessful and the method was discarded.

TABLE 1

Analyses of commercial batches of an otic glycerol solution labelled to contain 1.5% (w/w) procaine hydrochloride and 7% phenazone

Batch	Percent average recovery (CV, $n = 4$)	
	Procaine hydrochloride	Phenazone
1	99.2 (1.8)	98.4 (1.4)
2	101.9 (1.7)	100.9 (1.4)
3	102.9 (1.2)	98.1 (1.5)
4	100.0 (1.7)	100.4 (1.1)
5	102.5 (1.6)	98.2 (1.4)
6 [a]	102.1 (0.72)	99.5 (0.78)
7 [a]	101.8 (0.41)	99.1 (0.25)
8 [a]	101.4 (0.66)	98.3 (0.67)

[a] Pilot-size batch prepared as reference control.

In this study, the validity and applicability of a multicomponent analysis method were demonstrated by the assay of an otic preparation containing a PCN-PHZ combination. The good accuracy and precision of the proposed method is demonstrated in Table 1, which summarizes the results of the analyses of three standard and five commercial batches. The relative standard deviations for both drugs were less than 1.8%.

UV multicomponent spectrophotometry, though less specific than chromatographic techniques, may be a useful analytical tool in certain cases, particularly to meet the demands of routine usage in the production and stability controls of the pharmaceutical industry, on account of its simplicity, rapidity and fairly high precision and accuracy.

Acknowledgement

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References

Clarke, Isolation and Identification of Drugs, 2nd Edn, The Pharmaceutical Press, London, 1986a, pp. 872–873.

Clarke, Isolation and Identification of Drugs, 2nd Edn, The Pharmaceutical Press, London, 1986b, pp. 925–926.

El-Yazbi, F.A., Korany, M.A., Abdel-Razak, O. and Elsayed, M.A., Derivative spectrophotometric determination of some corticosteroids in combination with other drugs. J. Assoc. Off. Anal. Chem., 69 (1986) 614–619.

Fell, A.F., Analysis of pharmaceutical dosage forms by second derivative ultraviolet-visible spectrophotometry. Proc. Anal. Div. Chem. Soc., 15 (1978) 260–267.

Korany, M.A. and Haller, R., Spectrophotometric determination of oxazepam and dipyridamole in two-component mixtures. J. Assoc. Off. Anal. Chem., 65 (1982) 144–147.

Korany, M.A., Wahbi, A.M., Elsayed, M.A. and Mandour, S., Application of first derivative spectrophotometry to the determination of certain drugs in single component dosage forms. Farmaco, Ed. Prat., 39 (1984) 243–252.

Madsen, B.W., Herbison-Evans, D. and Robertson, J.S, Computer analysis of multicomponent ultraviolet spectra. J. Pharm. Pharmacol., 26 (1974) 629–636.

Miller, K.O., Otic products. In Handbook of nonprescription Drugs, 8th Edn, Am. Pharm. Ass., Washington, DC, 1986, pp. 419–435.

Morelli, B., Second-derivative spectrophotometric assay of mixtures of dicloxacillin sodium and ampicillin sodium in pharmaceuticals. J. Pharm. Sci., 77 (1988) 1042–1046.

Moussa, A.F.A. and Safwat, H.M., A new simplified formula for the calculation of individual absorbances in binary mixtures. Pharmazie, 34 (1979) 159–160.

O'Haver, T.C., Derivative and wavelength modulation spectrometry. Anal. Chem., 51 (1979) 91A–100A.

O'Haver, T.C. and Green, G.L., Numerical error analysis of derivative spectrometry for the quantitative analysis of mixtures. Anal. Chem., 48 (1976)312–318.

Parmentier, A., Barthélémy, J.F. and Andermann, G., Analyse de mélanges médicamenteux binaires par spectrophotométrie UV. I: cas favorables. Labo-Pharma, Probl. Tech., 32 (1984) 531–535.

Sala, G., Maspoch, S., Iturriaga, H., Blanco, M. and Cerda, V., Simultaneous multiwavelength spectrophotometric quantitation of active components in analgesic formulations. Comparative study of three calculation methods. J. Pharm. Biomed. Anal., 6 (1988) 765–772.

U.S.P. XXI, 1985, US Pharmacopeial Convention, Rockville, MD, pp. 71–72.

Wahbi, A.M., Abdine, H., Korany, M.A. and El-Yazbi F.A., Spectrophotometric analysis of binary mixtures of antazoline and naphazoline. J. Pharm. Sci., 67 (1978) 140–141.

Wahbi, A.M. and Unterhalt, B., The convolution of Gaussian absorption curves. Z. Anal. Chem., 282 (1976) 31–35.