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Application of the ΔA method to the determination of morphine

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Tablets, suppositories and ear-drops have been assayed using the ΔA method to eliminate interferences. The absorbance at 298 nm is measured for solutions in acid and alkaline media,the difference (Δ A) being taken as a measure of morphine content. The same method has been applied to suppositories after extraction of the drug from the base. Ear-drops containing phenol in addition to morphine were assayed by measuring ΔA at 288 and 298 nm and evaluating the morphine concentration from a pair of simultaneous equations.

If a compound contains a pH-sensitive auxochrome, its spectrum usually shifts when the pH changes over a suitable interval. In the ΔA method, such a "chemical shift" may be used to cancel the effect of irrelevant absorption (Aulin-Erdtman, 1955).

Morphine exhibits a maximum absorption at 285 nm in 0·1N sulphuric acid and at 298 nm in 0·1N sodium hydroxide (B.P. 1968). In these circumstances, pH-sensitive irrelevant absorption, z, may be cancelled by means of

$$\Delta A_i = (A_b + z)_i - (A_a + z)_i \quad .. \quad .. \quad .. \quad (1)$$

where A_a and A_b are the absorbances of 1 cm layers of acid and alkaline solutions containing the same concentration of morphine in each and the subscript "i" refers to the wavelength, 298 nm at which ΔA is maximum. The concentration of morphine, c_M, is given by:

$$c_M = \Delta A_i/\Delta\alpha_i \text{ where } \Delta\alpha_i = \alpha_{bi} - \alpha_{ai} \quad .. \quad .. \quad (2)$$

and α denotes the absorptivity of morphine.

By combining Vierordt's method and the ΔA method, a mixture of two-known pH-sensitive absorbing substances, A and B, may be determined in the presence of a pH-insensitive irrelevant absorption. Thus, the concentration of A and B can be evaluated from a pair of simultaneous equations of the following form:

$$\Delta A_1 = c_A\Delta\alpha_1 + c_B\Delta\beta_1 \quad .. \quad .. \quad .. \quad (3)$$

$$\Delta A_2 = c_A\Delta\alpha_2 + c_B\Delta\beta_2 \quad .. \quad .. \quad .. \quad (4)$$

The subscripts 1 and 2 refer to wavelengths; ΔA denotes the absorbance difference of the mixture in alkaline and acid media; c_A and c_B are the concentrations of A and B, whilst α and β are their respective absorptivities.

A good choice of wavelengths for a two-substance ΔA method is rather less obvious than for the one-substance ΔA method. Evidently, the error in an assay result based upon the former method which involves four absorbances measured at two wavelengths for solutions of A, B, and their mixture would be greater than that in a result based upon two absorbances measured at a single wavelength in a one-substance ΔA method. Thus, unless the principles of choosing wavelengths are well understood,

that is (i) avoidance of absorption curve slopes whenever possible; (ii) preference for λ_1 and $\lambda_2 = \lambda_{max}$ of the ΔA curves of substances A and B, the method may give unsatisfactory results.

EXPERIMENTAL

Instrument. A Unicam SP 500 photoelectric spectrophotometer.

Assay for tablets. Tablets were prepared, containing 10 mg of morphine sulphate per 0·30 g tablet (B.P. 1968). An accurately weighed quantity of the powder, representing 10–20 tablets, was quantitatively extracted with 0·1N sulphuric acid, filtered into a 100 ml volumetric flask and subsequently made to volume. Two 10 ml portions of the filtrate were diluted to 100 ml using 0·1N sulphuric acid and 0·1N sodium hydroxide respectively. The absorbances of 1 cm pathlengths of both solutions were measured at 298 nm.

Assay for suppositories. Suppositories were prepared, containing 15 mg of morphine hydrochloride per 1·5 g theobroma oil in each suppository. An accurately weighed quantity (10–20 g) of the melted suppositories was dissolved in 60 ml of light petroleum saturated with water. The solution was extracted with 3 × 20 ml of 0·1N sulphuric acid and subsequently washed with 3 × 5 ml portions of acidulated water. The combined aqueous extracts and washings (75 ml) were warmed on a water bath, cooled, and transferred to a 100 ml volumetric flask and made to volume. Final dilutions were prepared and absorbance measurements made by the method described for tablets.

Assay for ear-drops. This preparation contained morphine hydrochloride, 0·75 g and phenol, 1·5 g, made to 20 g (about 15 ml) with glycerol. An accurately weighed quantity (about 1·0 g) of the ear-drops was dissolved in water and final solutions prepared by the method described for tablets. Absorbances were measured at 288 and 298 nm.

To minimize the relative error in ΔA, the concentrations of the final solutions were controlled so that $(A_a + z)_1 + (A_b + z)_1$ was less than 1·0 (Junejo & Glenn, 1956; Glenn, 1965).

RESULTS AND DISCUSSION

When compared with the graph of log A versus λ for morphine sulphate, similar graphs for tablets and suppositories were distorted due to the presence of absorbing impurities. On the other hand, the graphs of log $|\Delta A|$ versus $\lambda(\Delta A \neq 0)$ for tablets and suppositories were completely superimposable with that for morphine sulphate, thus showing the irrelevant absorption to be unchanged in the two solvents used.

Table 1. *The determination of morphine in tablets and suppositories*

	Tablets		Suppositories	
Sample	mg added	% recovery	mg added	% recovery
1	8·0	100·25	40·0	96·7
2	10·0	101·20	25·0	98·4
3	16·0	100·25	20·0	99·5
4	20·0	100·00	15·0	98·7
5			10·0	102·0

It was not, therefore surprising that the above ΔA procedures gave mean percentage recoveries of 100·2 ± 0·8% for the morphine sulphate in four separate weighings of the powdered tablets and 99·1 ± 2·4% ($P = 0·05$) for the morphine hydrochloride in five separate weighings of the melted suppositories (Table 1).

By applying Vierordt's method (i) using absorbances and (ii) using absorbance differences, morphine in different concentrations, was determined in ear-drops containing phenol (Table 2). The mean percentage recovery was found to be 105·0 ±

Table 2. *The determination of morphine hydrochloride in ear-drops using Vierordt's method* ($\lambda_1 = 288$ nm and $\lambda_2 = 298$ nm)

Sample	Method mg added	Absorbance % recovery	Absorbance differences % recovery
1*	2·0	106·0	102·5
2*	2·4	107·5	102·9
3*	2·8	107·1	103·6
4	4·0	105·0	101·7
5	6·4	103·6	102·0
6	8·0	103·6	100·1

All samples contained 4·0 mg phenol.

* Low contribution of morphine hydrochloride.

2·7% using the former method (Glenn, 1960) and 102·1 ± 1·3% using the latter method (P 0·05). The poor results from the method using absorbances were due to irrelevant absorption in the glycerol which was cancelled by the method using absorbance differences.

Errors in both methods can also be attributed to (i) overall shifts in the wavelength scale which affect absorbance measurements made on steep slopes in the phenol and morphine hydrochloride absorption curves (Ismail & Glenn, 1964) and (ii) the low contribution of morphine to the mixture's total absorption curve especially, in samples marked with an asterisk (Glenn, 1960).

In the light of a satisfactory literature survey, the authors believe that the extension of the ΔA method to mixtures of two substances is original. The results obtained for the determination of morphine in presence of phenol are encouraging and suggest that the method warrants a careful study over a wide field of applications.

REFERENCES

AULIN-ERDTMAN, G. (1955). *Chemy Ind.*, **74**, 581–582.
British Pharmacopoeia (1968), pp. 630, 632.
Extra Pharmacopoeia: Martindale 25th Edition, **19**, p. 785.
GLENN, A. L. (1965). *Photoelectric Spectrometry Group Bulletin*, **16**, 464.
GLENN, A. L. (1960). *J. Pharm. Pharmac.*, **12**, 595–608.
ISMAIL, M. & GLENN, A. L. (1964). *Ibid.*, **16**, *Suppl.* 150*T*–155*T*.
JUNEJO, G. M. & GLENN, A. L.. (1956). *Chemy Ind.*, **75**, 813–814.