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Difference spectrophotometric assay of 5-hydroxymethylfurfuraldehyde in hydrolysed pharmaceutical syrups — I. Sodium borohydride reagent

A. G. DAVIDSON* and T. O. DAWODU

Department of Pharmacy, University of Strathclyde, George Street, Glasgow G1 1XW, UK

Abstract: A rapid difference spectrophotometric procedure is described for the assay of 5-hydroxymethylfurfuraldehyde (5-HMF) in hydrolysed pharmaceutical syrups. The assay involves measurement of the difference absorbance at 283 nm (ΔA_{283}) of a solution of 5-HMF at pH 8 relative to that of an equimolar solution in which the absorption of the 5-HMF has been destroyed by reduction of the carbonyl group by sodium borohydride. The ΔA_{283} is proportional to the concentration of 5-HMF and is unaffected by the presence of sucrose (the sugar component of syrup) or of dextrose or laevulose (the principal sugars of invert syrup). The accuracy, precision and selectivity of the method are discussed. The limits of detection and determination are 0.78 μg ml^{-1} and 9.6 μg ml^{-1}, respectively. The assay has been applied successfully to samples of syrup containing hydroxybenzoate (paraben) preservatives, invert syrup, simple linctus, ephedrine elixir and raspberry syrup.

Keywords: *5-hydroxymethylfurfuraldehyde; difference spectrophotometry; pharmaceutical syrups; invert syrup; simple linctus; ephedrine elixir; raspberry syrup.*

Introduction

The principal decomposition product of the acid-catalysed hydrolysis of dextrose and of laevulose is 5-hydroxymethyl-2-furaldehyde (5-HMF; 5-hydroxymethyl-2-furancarboxaldehyde). Quality control specifications for injections and dialysis fluids containing dextrose or laevulose normally include a quantitative or limit test for 5-HMF to control the extent of decomposition.

The techniques that are most frequently employed for the assay of 5-HMF are spectrophotometry and chromatography. The intense absorption of 5-HMF at its wavelength of maximum absorption (λ_{max}), 283 nm ($\epsilon_{283} = 1.63 \times 10^4$), provides the basis for a sensitive and precise method of analysis [1] but selectivity is reduced when

*To whom correspondence should be addressed.

other compounds that absorb at 283 nm are also present in the sample. Thus, the limit test in the British Pharmacopoeia monographs for Dextrose Injection and Laevulose Injection, which involves dilution of the injection to give a sugar concentration of 0.4% (m/v) or 0.2% (m/v), respectively, and then the measurement of absorbance at the λ_{max} near 284 nm, is designated as a test for "5-hydroxymethylfurfural and related substances" [2]. The maximum absorbance that is permitted is 0.25, which corresponds to a concentration of 5-HMF of about 2 $\mu g\ ml^{-1}$ in the diluted injection and about 0.05% (m/m) or 0.1% (m/m) of the sugar concentration in Dextrose Injection or Laevulose Injection, respectively. Methods involving formation of a coloured derivative and measurement of absorbance in the visible region give better selectivity for colourless samples but generally these procedures are time-consuming or involve toxic reagents [3, 4]. By using a high-performance liquid chromatographic method to separate 5-HMF and individual related products in autoclaved dextrose solutions, Hung *et al.* [5] showed that direct spectrophotometric procedures also provide inadequate control of those products that do not absorb at 284 nm.

A number of analytical techniques for assaying 5-HMF in honey, wine, jam and other foods containing sugars have been described [6–14]. One is a difference spectrophotometric procedure [8] which involves the reaction of 5-HMF with sodium hydrogen sulphite to give a sulphite addition product that does not absorb at 284 nm. However, the reagent itself absorbs weakly at 284 nm and, at the concentration of reagent used (0.1% m/v), the reaction is incomplete, 5.7% of the 5-HMF remaining underivatized.

Despite the evidence of high concentrations of 5-HMF in honey and other foods containing sugar, no investigations have been undertaken to determine the concentrations of 5-HMF in pharmaceutical syrups. In the present paper and in Part II [15] new difference spectrophotometric procedures are described for the assay of 5-HMF in certain syrups and syrup formulations based upon the reduction of the aldehyde group with sodium borohydride. The use of sodium borohydride in difference spectrophotometry was first described by Görög [16] for the reduction of 4-en-3-one steroids.

Experimental

Reagents

5-Hydroxymethylfurfuraldehyde (BDH Chemicals, Poole, UK), D-glucose, D-fructose and sucrose (BDH Chemicals) were of analytical reagent quality.

Sodium borohydride (BDH Chemicals) was of general purpose reagent quality. Sodium borohydride reagent (0.5% m/v) was prepared by dissolving 500 mg of sodium borohydride in ethanol and diluting to 100 ml. The solution is stable for 1 week.

Buffer solutions were prepared using substances of analytical reagent grade according to published formulae [17]: buffers pH 1 and pH 2 (potassium chloride and hydrochloric acid); pH 3, 4, 5, 6 and 7 (citric acid and disodium hydrogen phosphate); pH 8, 9 and 10 (potassium chloride, boric acid and sodium hydroxide).

Spectrophotometer

Absorption and difference absorption spectra of solutions in 1-cm silica quartz cells were recorded using a Perkin–Elmer 552 UV–visible spectrophotometer. The spectral bandwidth was 2 nm, the scan rate 1 nm s^{-1} and the response (time constant) 0.5 s. The difference absorbance values of the standard, sample and blank solutions at 283 nm were read from the digital display under non-scanning conditions.

High-performance liquid chromatography (HPLC)

HPLC was carried out using an Altex pump (model 100A). The chromatographic conditions were slight modifications of those described by Durham *et al.* [18]: stainless steel column (100 × 4.6 mm) containing Spherisorb 5 ODS; mobile phase, water; flow rate, 1.0 ml min^{-1}; spectrophotometric detection at 283 nm; volume injected, 20 μl (valve). The concentration of the standard solution of 5-HMF was 20 μg ml^{-1} and the syrups were diluted with at least four volumes of water to give a concentration of 5-HMF not exceeding that of the standard solution.

Procedure

Standard solutions. A standard solution of 5-HMF was prepared by dissolving *ca* 80 mg, accurately weighed, in 5 ml of ethanol and diluting to 1 l with water. A 5-ml aliquot was transferred to two 50-ml volumetric flasks. Sodium borohydride reagent (2 ml) was added to one flask and when the evolution of hydrogen had ceased (after about 5 min), the contents of both flasks were diluted to 50 ml with water. The difference absorbance at 283 nm (ΔA_{283}) of the untreated solution in the sample cell was measured relative to that of the solution treated with sodium borohydride in the reference cell.

Sample solutions. The *weight per ml* of the syrup was determined by weighing 10 ml of the syrup contained in a 10-ml volumetric flask. A quantity of the syrup equivalent to about 5 ml was weighed accurately into a 50-ml beaker and 15 ml of water was added. Alternatively, for low viscosity samples, a pipette (calibrated to contain 5 ml) was used to transfer 5 ml of the syrup to the beaker; the pipette was then rinsed with 3 × 5 ml of water. The pH of the solution was measured with a pH-meter; if necessary, the pH was adjusted to 8 by adding drops of 0.1 M or 1 M sodium hydroxide. The solution was then transferred with rinsing to a 25-ml volumetric flask and diluted to volume with water. Equal volumes of the solution up to 10 ml containing up to 400 μg, depending on the concentration of 5-HMF in the sample, were transferred to two 50-ml volumetric flasks and the procedure was continued as described above for *standard solutions* from the words "sodium borohydride reagent (2 ml) was added. ...". Solutions of syrups preserved with hydroxybenzoate esters (parabens) were acidified with 2 ml of 1 M hydrochloric acid and diluted to 50 ml with water.

The concentration of 5-HMF in the sample solutions, and hence in the syrup sample, was calculated from the proportional relationship that exists between the difference absorbance at 283 nm and the concentration of 5-HMF in the unreduced solution.

Results and Discussion

Difference spectrophotometry involves the measurement of the difference absorbance (ΔA) between the absorbance of two equimolar solutions of a substance in different chemical forms that exhibit different spectral characteristics. Substances whose spectra are dependent on the state of ionization, and consequently on the pH of the solution, can be assayed by measuring the ΔA between two equimolar solutions at different pH values. The pH values and wavelength of measurement normally are selected to provide the maximum difference in absorbance. Non-ionizable substances can be assayed by measuring the ΔA generated between equimolar solutions of the substance and of a derivative of the substance obtained by reaction with a suitable reagent. Ideally the chemical conversion should be rapid and complete. Assays involving oxidation [19],

reduction [16, 20], competitive condensation [21] and ester formation [22, 23] reactions that meet these requirements have been described.

Difference spectrophotometric techniques are selective for substances whose spectral properties can be altered provided that the absorbance of other substances in the sample is not affected by the reagents involved.

Development of the method

Figure 1a shows the UV absorption spectra of 5-HMF in the presence and absence of sodium borohydride. Reduction of 5-HMF by sodium borohydride eliminates the strong absorption band of 5-HMF around 283 nm and results in an increase in absorption at 222 nm. The difference absorption spectrum (Fig. 1b) obtained by recording the spectrum of the solution of unreduced 5-HMF against the equimolar solution of reduced 5-HMF in the reference cell shows a λ_{max} at 283 nm and an isosbestic point (wavelength of zero ΔA because of the equal absorptivity of unreduced and reduced 5-HMF) at 239 nm. The change in spectral properties of 5-HMF by reduction is due to the destruction of the dienone chromophore of 5-HMF (I) to yield the diene chromophore of 5-hydroxymethyl-2-furfuryl alcohol (II) which has a λ_{max} at 222 nm.

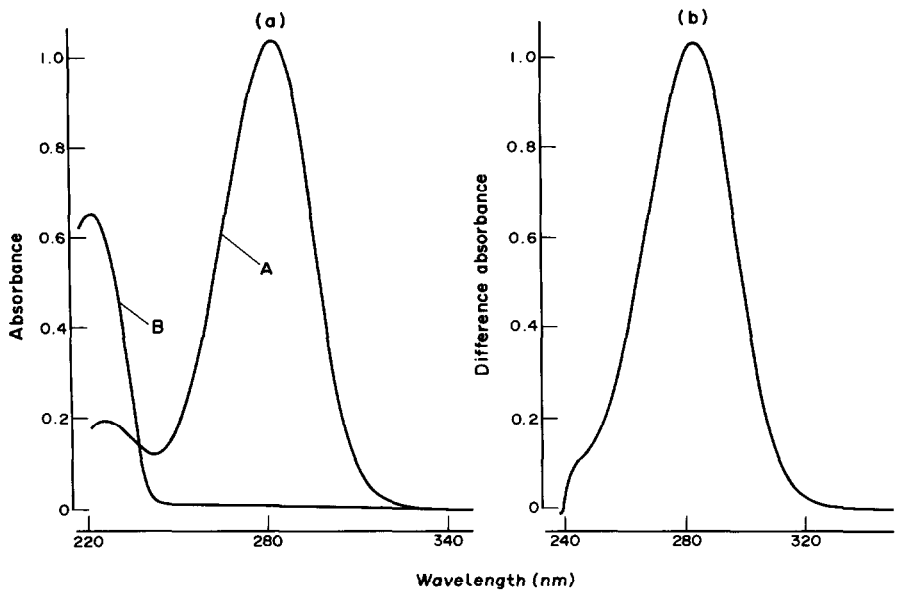
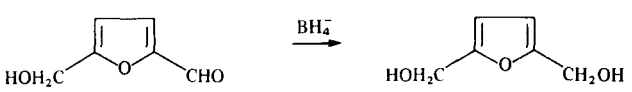


Figure 1
(a) UV absorption spectra of 5-HMF (8 μg ml^{-1}): (A) in water and (B) in sodium borohydride solution (0.2 mg ml^{-1}); (b) the difference absorption spectrum of solution A relative to solution B.



I II

The effect of pH on the reduction of 5-HMF was investigated by measuring the absorbance at 283 nm (A_{283}) of solutions of 5-HMF that had been buffered to various pH values in the range 1–10 before addition of the reducing agent. The A_{283} of solutions of 5-HMF buffered to the same pH values but without reducing agent was also measured. The results in Fig. 2 show that the A_{283} of 5-HMF is not dependent on pH and that a pH of 8 or greater is necessary for the complete reduction of 5-HMF.

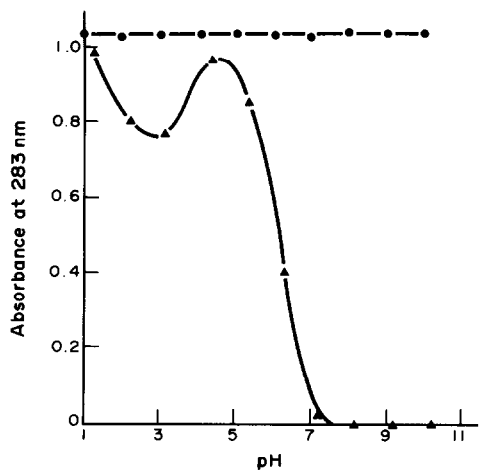


Figure 2
The effect of pH on the absorbance of 5-HMT (8 μg ml^{-1}) in the presence (-▲—▲-) and absence (-●—●-) of sodium borohydride (0.2 mg ml^{-1}).

Aqueous solutions of sodium borohydride are alkaline; the pH of a solution containing 0.2 mg ml^{-1}, the concentration in solutions of reduced 5-HMF, is *ca* 9.5. The addition of the reagent to simple aqueous solutions of 5-HMF raises the pH to a value which permits efficient reduction. It is therefore unnecessary to adjust the pH of the standard solution before the addition of the reagent. In contrast, some of the syrups, e.g. Invert Syrup which is prepared by the acid hydrolysis of sucrose and simple linctus which contains 2.5% (m/v) of citric acid, are so acidic that their solutions have a pH as low as 4 even after the addition of the reducing agent. Complete reduction of 5-HMF in these syrups is achieved only if the pH is increased to about 8 before addition of the reagent.

Figure 3 shows the effect of varying the concentration of freshly prepared sodium borohydride reagent on the reduction of 5-HMF in solutions buffered at pH 8. Complete reduction was obtained with all concentrations above 0.05 mg ml^{-1}. In parallel experiments, the stability of sodium borohydride in different solvents was investigated by measuring the efficiency of reduction of 5-HMF in freshly prepared solutions given by 0.5% (m/v) solutions of sodium borohydride in water, methanol, ethanol and propan-2-ol at various times after their preparation. The reagents in ethanol (Fig. 4) and in propan-2-ol were the most stable, giving complete reduction of 5-HMF at least 10 days after preparation. Ethanol rather than propan-2-ol was selected as the solvent for the reducing agent owing to the greater solubility of sodium borohydride in ethanol. The composition of the reducing agent was therefore selected to be 0.5% (m/v) in ethanol and a shelf-life of 7 days was assigned to the reagent. Slight turbidity occurred in some solutions of sodium borohydride in ethanol but this disappeared on addition of the reagent to aqueous solutions of 5-HMF and did not present any problems.

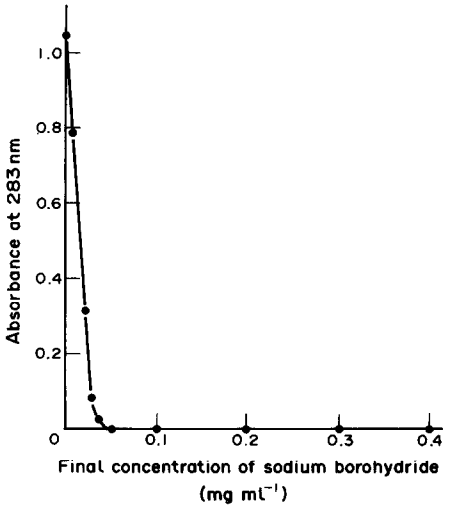


Figure 3
The effect of concentration of sodium borohydride on the absorbance of 5-HMF (8 μg ml^{-1}) at pH 8.

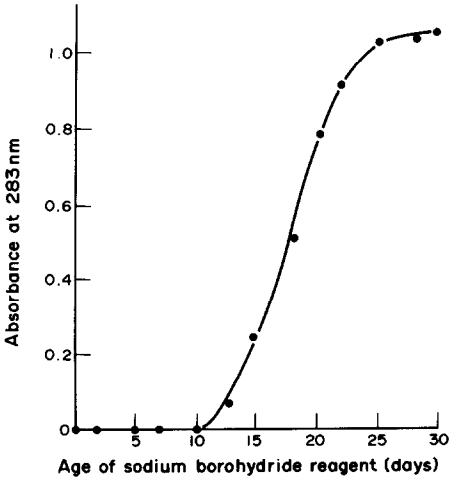


Figure 4
The effect of age of the sodium borohydride reagent (0.2 mg ml^{-1}) on the reduction of 5-HMF (8 μg ml^{-1}) at pH 8.

Validation

The proportionality of the ΔA_{283} values and the concentration of 5-HMF were checked by using a calibration series of solutions (0, 2, 4, 6, 8 and 10 μg ml^{-1}) of 5-HMF in the absence and presence of sucrose (5% m/v), glucose (5% m/v) and laevulose (5% m/v). The results in Table 1 show that the ΔA_{283} is proportional to the concentration of 5-HMF in the range 0–10 μg/ml and that the sugars at a level of 5% (m/v) do not interfere with the measured value. In complementary experiments, the concentration of 5-HMF was constant at 8 μg ml^{-1} and the concentration of each of the sugars was varied systematically from 0 to 6% (m/v). The ΔA_{283} of all the solutions containing sugar were within 99.1–100.7% of that of the solutions of 5-HMF only, confirming that the sugars do not affect the measured value.

The slope of the regression line for the series of solutions containing 5-HMF only (Table 1) corresponds with an A(1% m/v; 1 cm) value of 1296 and is in good agreement

Table 1
Calibration data

Composition of solutions	Regression equation*	r†
5-HMF	$y = 0.1296x - 0.001$	0.9999
5-HMF + glucose (5%)	$y = 0.1290x - 0.003$	0.9996
5-HMF + laevulose (5%)	$y = 0.1298x + 0.001$	0.9996
5-HMF + sucrose (5%)	$y = 0.1297x + 0.002$	0.9998

*$y = \Delta A_{283}$; x = concentration of 5-HMF in μg ml^{-1}.
†r = Correlation coefficient ($n = 6$).

Table 2
Recovery data

Sample*	Concentration of 5-HMF (μg ml^{-1}) Without added 5-HMF	With added 5-HMF (80 μg ml^{-1})	Recovery (as % of added 5-HMF)
Ephedrine elixir (1)	17.6	99.0	101.7
Invert syrup (6)	119.5	199.7	100.3
Simple linctus (9)	26.4	105.4	98.8
Syrup (preserved) (13)	ND†	79.2	99.0
Raspberry syrup (15)	148.4	229.0	100.7

*The numbers in brackets refer to the sample numbers in Table 3.
†ND = None detected; less than the limit of determination.

with the previously reported value of 1300 for the absorptivity of 5-HMF [24]. This confirms both that the purity of 5-HMF used as a reference standard is satisfactory and that complete reduction of 5-HMF occurs under the conditions of the assay.

The precision of the procedure was determined by assaying a sample of ephedrine elixir (Sample 3, Table 3) ten times. The mean concentration of 5-HMF in the syrup was 76.7 μg ml^{-1} and the standard deviation was 0.69 μg ml^{-1} (relative standard deviation, 0.90%) showing that the procedure has good precision.

The limit of detection of the assay, calculated as the concentration of 5-HMF in the syrup giving a ΔA_{283} in dilutions (1 + 24) of the syrup equal to twice the standard deviation ($n = 10$) of the ΔA_{283} of the blank solutions (water and 0.2 mg ml^{-1} sodium borohydride solution) was 0.78 μg ml^{-1}. The limit of determination, calculated as the concentration of 5-HMF in the syrup giving a ΔA_{283} of 0.05 in dilutions (1 + 24) of the syrup was 9.6 μg ml^{-1}.

The specificity of the assay of 5-HMF in those syrups containing known constituents that are UV-absorbing was investigated by measuring the ΔA_{283} of these substances under the conditions of the assay. Tartrazine (final concentration 3 μg ml^{-1}) and ephedrine hydrochloride (final concentration 120 μg ml^{-1}) gave a $\Delta A_{283} = 0$ and so do not interfere in the assay of 5-HMF in ephedrine elixir. Amaranth (final concentration 6 μg ml^{-1}) the colouring agent in simple linctus did, however, show a small ΔA_{283} that increased slowly with time. This was due to a slow decolorizing action of sodium borohydride on the dye. When the procedure was carried out quickly the ΔA_{283} of the amaranth was only 0.01, equivalent to a concentration of 5-HMF of 2.0 μg ml^{-1}, a value that was considered to be small in comparison to the concentrations of 5-HMF found to be present in simple linctus.

Table 3
Assay results

Sample	Manufacturer*	Age (years)	Initial pH†	Concentration of 5-HMF (μg ml^{-1})	
				This method	HPLC [18]
1. Ephedrine elixir	A	2 7/12	4.2	17.6	17.0
2. Ephedrine elixir	A	3 1/12	3.9	25.8	24.7
3. Ephedrine elixir	B	11	3.6	76.7	78.7
4. Invert syrup	C	5	3.5	261.3	262.0
5. Invert syrup	C	3 1/12	4.2	203.9	201.8
6. Invert syrup	C	2 9/12	4.4	119.5	118.4
7. Invert syrup	C	2 9/12	4.1	216.3	222.9
8. Invert syrup	D	12	3.6	344.1	348.3
9. Simple linctus	A	8/12	2.7	26.4	26.2
10. Simple linctus	A	1	2.7	62.9	62.0
11. Simple linctus	D	13 4/12	2.5	906	899
12. Simple linctus (paediatric)	D	14 8/12	2.7	463	469
13. Syrup (preserved)§	A	8/12	5.5	ND‡	ND‡
14. Syrup (preserved)§	E	1	8.3	ND‡	ND‡
15. Raspberry syrup	A	5 4/12	2.9	148.4	147.5

*A = Evans Medical; B = Bush Boake Allen; C = Thornton and Ross; D = Macarthys; E = William Ransom.
†pH of syrup diluted 1 + 3 with water.
‡ND = None detected; less than the limit of determination.
§The preservatives were shown by gas–liquid chromatography to be a mixture of methyl and propyl hydroxybenzoates.

Hydroxybenzoate preservatives were found to give a large negative ΔA_{283} under the conditions of the general procedure described for syrups, owing to the alkalinity of the reducing agent. The pH of the solution treated with sodium borohydride reagent is slightly higher than that of the untreated solution and this gives rise to a pH-induced ΔA_{283} of the preservatives because their spectra are markedly affected by even small changes of pH in the region of the pK_a of the phenolic group (8.4 [25]). The problem is avoided by re-acidifying the solutions to a low pH value at which the phenolic group is completed unionized.

Although the effect of other aldehydic substances on the ΔA_{283} of 5-HMF has not been investigated, it is likely that other aldehydes that absorb at 283 nm will be reduced by sodium borohydride and interfere in the assay of 5-HMF. However, confirmation of adequate specificity for all the syrups assayed for 5-HMF in this work (Table 3) was provided by the excellent agreement of the isosbestic points in the difference spectra of the standard and sample solutions (cf. Figs 1b and 5b).

The recovery of 5-HMF was measured by assaying one batch of each syrup formulation to which the method has been applied (Table 3), before and after the addition of 5-HMF (80 μg ml^{-1}). The results in Table 2 show that the added 5-HMF was assayed with satisfactory accuracy and indicated that the syrup ingredients do not affect the measured ΔA_{283}.

Applications and assay results

Many syrup formulations contain drugs and/or excipients that absorb intensely at 283 nm and this precludes the application of the method to these syrups. Only those syrups

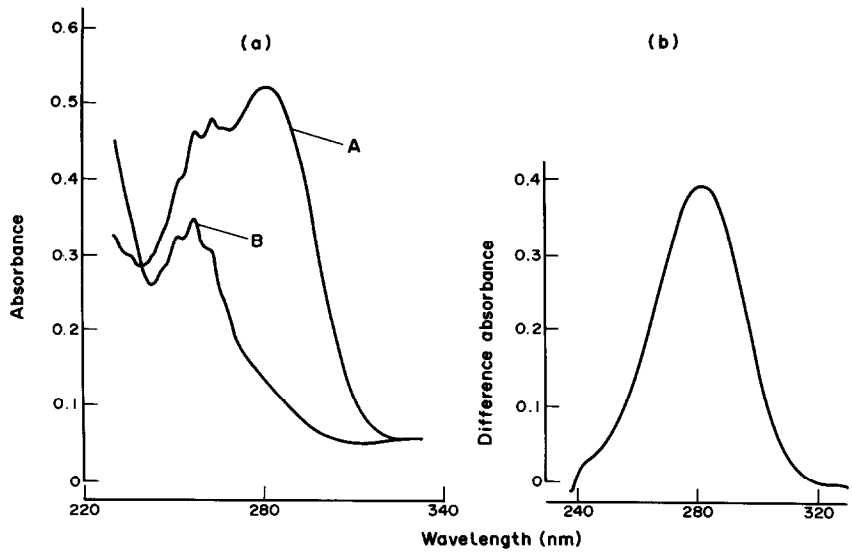


Figure 5
(a) UV absorption spectra of ephedrine elixir diluted 1 + 24: (A) in water and (B) in sodium borohydride solution (0.2 mg ml^{-1}); (b) the difference absorption spectrum of solution A relative to solution B.

that, after dilution (1 + 24) and reduction with sodium borohydride, give an A_{283} of less than 1 were considered to be suitable for assay by the present method. The assay results for a number of such syrups of varying ages are given in Table 3. For comparison, the concentrations of 5-HMF were also determined by an HPLC procedure based on that of Durham *et al.* [18]. The results in Table 3 show excellent agreement between the methods and confirm that the difference spectrophotometric procedure is both accurate and selective. Also, the coincidence of the retention times (5.1 min) of the principal peak in the chromatogram of the sample solutions and that of the standard solution of 5-HMF confirms that the substance that is assayed by the difference spectrophotometric procedure is indeed 5-HMF. The absorption and difference absorption spectra of solutions of ephedrine elixir (Sample 3; Table 3) are shown in Figs 5a and 5b, respectively; these spectra illustrate that, whereas a direct spectrophotometric assay of 5-HMF is subject to interference from other absorbing components of the syrup, the difference spectrophotometric assay is selective for 5-HMF.

Also recorded in Table 3 are the ages of the syrups and the pH values of the syrups, diluted 1 + 3 with water, before adjustment to pH 8 with sodium hydroxide solution. It is evident that the highest levels of 5-HMF are found in old syrups that have a low pH value. The high concentrations of 5-HMF in old samples of simple linctus, whose sugar basis is Syrup B.P., indicate that the sucrose first inverts to dextrose and laevulose before further hydrolysis to 5-HMF. This suggestion is supported by the observation that the samples of simple linctus were laevorotatory at the sodium D-line indicating that they had undergone extensive inversion.

A modified procedure that may be applied to certain other syrup formulations for which the present method is not applicable is described in Part II [15].

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References

[1] J. H. Turner, P. A. Rebers, P. L. Barrick and R. H. Cotton, *Anal. Chem.* **26**, 898–901 (1954).
[2] *British Pharmacopoeia*, Her Majesty's Stationary Office, London (1980).
[3] C. Viel, *Analusis* **4**, 471–488 (1976); *Anal. Abs.* **33**, 1D70 (1977).
[4] Y. Yoshihiro and M. Nakamura, *J. Chem. Soc. Japan, Ind. Chem. Sect.* **61**, 972–975 (1958); *Anal. Abs.* **6**, 1915 (1959).
[5] C. T. Hung, A. B. Selkirk and R. B. Taylor, *J. Clin. Hosp. Pharm.* **7**, 17–23 (1982).
[6] O. Winkler, *Z. Lebensmitt-Untersuch* **102**, 161–167 (1955); *Anal. Abs.* **3**, 836 (1956).
[7] R. Thalacker and I. Kaltwasser, *Monatschr. Brau.* **31**, 20–24 (1978); *Anal. Abs.* **35**, 1F26 (1978).
[8] J. W. White, *J. Assoc. Off. Anal. Chem.* **62**, 509–514 (1979).
[9] A. K. Dhar and B. R. Roy, *Analyst* **97**, 981–985 (1972).
[10] W. Postel, *Dt. Lebensmitt Rdsch.* **64**, 318–322 (1968); *Anal. Abs.* **18**, 1264 (1970).
[11] F. De Francesco and G. Margheri, *Boll. Lab. Chim. Provinciali* **13**, 466–477 (1962); *Anal. Abs.* **10**, 2896 (1963).
[12] M. A. Williams, R. C. Humphreys and H. P. Reader, *Am. J. Enol. Vitic.* **34**, 57–60 (1983); *Anal. Abs.* **46**, 9F51 (1984).
[13] K. Wackerbauer, P. Kraemer, F.-J. Methner and U. Marx, *Monatsschr. Brauwiss.* **36**, 439–442 (1983); *Anal. Abs.* **46**, 5F49 (1984).
[14] F. C. Alfonso, G. E. Martin and R. H. Dyer, *J. Assoc. Off. Anal. Chem.* **63**, 1310–1313 (1980).
[15] A. G. Davidson and T. O. Dawodu, *J. Pharm. Biomed. Anal.* (in press).
[16] S. Görög, *J. Pharm. Sci.* **57**, 1737–1741 (1968).
[17] H. A. M. McKenzie, in *Data for Biochemical Research* (R. M. C. Dawson, D. C. Elliot, W. H. Elliot and K. M. Jones, Eds), pp. 475–504. Oxford University Press, London (1968).
[18] D. G. Durham, C. T. Hung and R. B. Taylor, *Int. J. Pharm.* **12**, 31–40 (1982).
[19] A. G. Davidson, *J. Pharm. Pharmacol.* **28**, 795–800 (1976).
[20] A. G. Davidson, *J. Pharm. Pharmacol.* **30**, 410–414 (1978).
[21] A. G. Davidson, *Analyst* **107**, 422–427 (1982).
[22] A. G. Davidson, *J. Pharm. Biomed. Anal.* **2**, 45–52 (1984).
[23] A. G. Davidson, *J. Pharm. Sci.* **73**, 1582–1584 (1984).
[24] C. Franzke and H. Iwainsky, *Fette, Seifen, Anstrichm.* **58**, 859–862 (1956); *Anal. Abs.* **4**, 3115 (1957).
[25] *Martindale The Extra Pharmacopoeia*, 28th edn, pp. xxvi and xxvii. The Pharmaceutical Press, London (1982).

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