Analysis of Plasma Ampicillin Extracts—GLC, TLC, HPLC, and adsorption chromatographic procedures have been developed for ampicillin. However, these methods cannot be adapted for the detection and quantitation of ampicillin and its metabolites in plasma since sensitivities are not adequate. The best sensitivity attained was 10 μg/ml by HPLC (9). Fluorescence methods with a sensitivity at 0.1 μg/ml have been used (10–12). However, these methods at best are specific for intact ampicillin but do not measure decomposition products or metabolites. Enhanced extraction with tetraheptylammonium chloride followed by chromatography offers the possibility of quantitating plasma ampicillin levels in the 1-μg/ml range.

Metabolites could also be extracted, followed by identification and quantitation. The procedure could be modified for the separation of ampicillin from its prodrugs. For example, pivampicillin could be easily separated with a prior chloroform extraction. Ampicillin would then be quantitated using the described procedure.

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COMMUNICATIONS

Spectrophotometric Analysis of Binary Mixtures of Antazoline and Naphazoline

Keyphrases □ Antazoline hydrochloride—spectrophotometric analysis simultaneously with naphazoline nitrate in pharmaceutical formulations □ Naphazoline nitrate—spectrophotometric analysis simultaneously with antazoline hydrochloride in pharmaceutical formulations □ Spectrophotometry—analyses, antazoline hydrochloride and naphazoline nitrate, simultaneously in pharmaceutical formulations □ Antihistaminics—antazoline hydrochloride, spectrophotometric analysis simultaneously with naphazoline nitrate in pharmaceutical formulations □ Adrenergic agents—naphazoline nitrate, spectrophotometric analysis simultaneously with antazoline hydrochloride in pharmaceutical formulations

To the Editor:

Two-component spectrophotometric analysis (1) is based on solving a set of two linear equations. Glenn (2) formulated the solution of the two linear equations in terms of absorbance ratios that are independent of concentration. Pernarowski *et al.* (3) used absorbance ratios for the analysis of binary mixtures and derived an equation similar to Glenn's equation. However, these authors (3) assumed that an isoabsorptive point must be chosen to apply their equation. The equation published by Cho and Pernarowski (4) to obtain absolute concentration was derived under the impression that an isoabsorptive point must be present to apply the equation of Pernarowski *et al.* (3). The three published equations (2–4) are consistent with each other and can be applied using any suitably chosen pair of wavelengths. None of the selected wavelengths needs to be an isoabsorptive point.

According to Glenn's limitations (2), if the contribution of one component to the absorption curve of the total mixture is low, erroneous results are obtained whenever the two-wavelength method of analysis is applied. In this connection, we suggest the use of least squares (5) to minimize the instrumental errors during the analysis of the minor component in binary mixtures.

These methods were applied to the determination of antazoline hydrochloride (I) (0.5% w/v) and naphazoline nitrate (II) (0.025% w/v) in nasal drops (6–8) also containing chlorobutanol (0.5% w/v) and sodium chloride (0.6% w/v). Thus, by diluting 1 ml of the nasal drop solution to 50 ml with 0.1 *N* H_2SO_4, measuring the absorbances of 1-cm pathlengths at 281 and 295 nm[1], and applying the Glenn and Cho and Pernarowski equations, the mean percentage recoveries for I were 99.5 ± 1.05 and 99.7 ± 1.24, respectively, for 10 samples; for II, they were 107.1 ± 2.35 and 107.1 ± 2.64, respectively, for 10 samples. The recoveries obtained using the two equations were consistent. The relatively high percentage recoveries obtained for II were due to its low contribution to the absorption curve of the total mixture at the concentration used.

To improve the accuracy and precision for the determination of II, the method of least squares (5) was applied. Fifteen absorbances measured at the wavelengths of 267–295 nm at 2-nm intervals gave the best results (mean ± *SD* = 100.7 ± 1.10) for the determination of II in the same 10 samples. The final wavelengths used covered the peak characteristics of II (8). Neither chlorobutanol nor sodium chloride interfered with the determinations.

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Simplified Method to Study Stability of Pharmaceutical Systems

Keyphrases □ Stability—pharmaceutical preparations, criticism of previously reported method relating plot of log $t_{0.9}$ and reciprocal of absolute temperature to shelflife □ Shelflife—pharmaceutical preparations, criticism of previously reported method of prediction by relating to plot of log $t_{0.9}$ and reciprocal of absolute temperature

To the Editor:

A recent study (1) reported a simplified method to study the stability of pharmaceutical preparations. According to this method, shelflife or $t_{0.9}$, the time required for 10% degradation, is determined at elevated temperatures, and, by plotting log $t_{0.9}$ against the reciprocal of the absolute temperature, the shelflife at room temperature can be calculated by extrapolation of the apparent straight line. This approach is suggested (1) to be applicable to all orders of reactions since the initial decay of up to 10% can be fitted by a first-order equation regardless of the actual order of reaction.

Although some limited academic applications of this method (1) can be demonstrated, it has little utility because of erroneous assumptions inherent in the approach and the unfeasible experimental methods suggested.

This study suggests the use of the Arrhenius approach for all orders of reactions, an erroneous assumption. In fitting a straight line through a log $t_{0.9}$ and reciprocal absolute temperature profile, the intercept on log $t_{0.9}$ can be obtained only if $1/T$ approaches zero or T approaches infinity. Thus, no significance should be attached to such intercepts since they represent only a mathematical treatment parameter.

Although Amirjahed (1) tried to prove that, for up to 10% degradation, the concentrations can be fitted by straight lines, no correlation was made between the calculated rate constants reported in the literature. For example, no correlation exists between the rate constants calculated for zero-order reactions ($r = 0.078$) as reported in Table VI of Ref. 1.

Even in those instances where a straight line can be fitted to the log $t_{0.9}$–temperature profile, the slope of the line will be highly dependent on the initial concentration except for a first-order reaction. For example, an allowable (±5%) content variation will result in a 10% variation in the calculated shelflife of a zero-order reaction. Thus, the statement made by the author that "the present method does not depend on x and k" is misleading. Briefly, therefore, a shelflife obtained at one concentration level cannot be extrapolated to other levels and is only valid for the sample studied, making it an evasive method with little practical utility.

The conclusions drawn (1) were based on either theoretically generated curves or published data obtained by more rigorous methods. It would have been more convincing if the author had used actual laboratory data for decompositon up to 10% to calculate the stability at room temperature, since this approach will require extremely sensitive analytical techniques to obtain concentration profiles. Generally, the techniques available for the analysis of dosage forms are not sensitive enough to detect small variations accurately. Thus, unless appropriate analytical techniques are available, the suggested method (1) has little utility, especially in "small laboratories and hospital pharmacy manufacturing units" as recommended by the author (1).

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Use of Area under the Curve to Estimate Absolute Bioavailability of Digoxin

Keyphrases □ Digoxin—bioavailability estimated using area under the curve from 0 to 24 and 0 to 72 hr and 0 to infinity □ Bioavailability—digoxin, estimated using area under the curve from 0 to 24 and 0 to 72 hr and 0 to infinity □ Cardiotonic agents—digoxin, bioavailability estimated using area under the curve from 0 to 24 and 0 to 72 hr and 0 to infinity

To the Editor:

The absolute bioavailability of digoxin from tablets has been discussed widely with respect to the actual numerical value as well as the most appropriate method of measurement. Reported values are based on the use of the area under the serum digoxin concentration–time curve from 0 to a finite time, such as 24 or 72 hr, and range from 55 to 65% (1–6). These areas for a finite time approximate the theoretically correct area from zero to infinity (7). This communication compares the approximate method of estimating the absolute bioavailability of a digoxin tablet[1] using the area under the curve from 0 to 24 hr and from 0 to 72 hr and the theoretically correct method using the area under the curve from 0 to infinity.

The absolute bioavailability of a digoxin tablet[1] was measured in 12 normal volunteer subjects. The tablet was given at doses of 0.5 and 1.0 mg (two or four 0.25-mg tab-

[1] Lanoxin Tablet, 0.25 mg, lot 022-1, Burroughs Wellcome and Co.