occur with human or animal blood would not be expected to have any effect, no buffering was used. Urine samples, however, vary significantly in pH and need pH adjustment to 6.8.

The character of the fluorescent spectrum changed as the plate dried. The peak became larger and more defined at the given excitation and emission wavelengths (362 and 422 nm, respectively). There was also a significant shift of both the excitation and emission maximum wavelengths to 330 and 480 nm, respectively, when tribromsalan was in 7.5% acetic acid in methanol. Spectra obtained with wet plates resembled those in 7.5% acetic acid in methanol. The change of fluorescence in the presence of acid was also observed by other investigators with aspirin (9). Drying the plate for more than 24 hr in the air provided peaks with consistent intensities without a shift of maximum wavelengths on the fluorescent spectrophotometer.

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pH-Dependent Secretion of Procainamide into Saliva

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Abstract □ The relationship between serum and stimulated, mixed saliva concentrations of procainamide was determined in 12 chronically medicated patients. Samples were obtained at times chosen to approximate the maximum and minimum serum concentrations of the drug during a dosing interval. Marked intersubject variability was found in the ratio of saliva to serum concentration of the drug (0.27–8.93). There was no correlation between the dose (milligrams per kilogram per day) and the minimum serum or saliva concentration of procainamide. Saliva pH ranged from 6.3 to 8.0 in eight subjects. The ratio of saliva to serum concentration of procainamide increased with decreasing pH. This result can be largely explained by the pH-dependent ionization and distribution of procainamide, a weak base.

Keyphrases □ Procainamide—pH-dependent secretion into saliva, ratio of saliva to serum concentration, humans □ Saliva—concentration of secreted procainamide, pH dependent, compared to serum concentration, humans

Procainamide, an antiarrhythmic compound, demonstrates marked intersubject variability in absorption and elimination (1). The range of therapeutic serum concentrations, however, has been defined (1). Mean plasma concentrations ranging from 4 to 8 μg/ml are generally effective in controlling arrhythmias, while toxicity is likely to occur at concentrations above 12 μg/ml. Thus, serum concentration measurements are useful in monitoring therapy in individual patients. However, procainamide has a short half-life, approximately 3–6 hr, which necessitates multiple venipunctures to characterize the peak and minimum serum concentrations during a dosing interval and during continued therapy. These requirements may be excessively traumatic for routine clinical application. The purpose of this study was to assess whether saliva concentrations of procainamide can be used to monitor drug concentrations in the plasma of patients receiving the drug.

The salivary secretion of other drugs has been the subject of several investigations. Good correlation between either total or unbound serum and mixed saliva concentrations was demonstrated in humans for theophylline (2), lithium (3), digoxin (4), acetaminophen (5), salicylate (at low concentrations) (6), phenytoin (7), and several sulfonamides (8).

EXPERIMENTAL

Twelve patients in an acute coronary care unit[1], who had been receiving a fixed dose of procainamide hydrochloride[2] for at least 2 days, were studied. Daily doses, dosage intervals, and times of sample collection in relation to dose are listed in Table I for the individual patients. The clinical records indicated that renal and liver functions were essentially normal in all subjects.

Blood and mixed, stimulated saliva were collected simultaneously before and after a regularly scheduled dose of procainamide. Saliva flow was stimulated by having the patients chew on waxed film[3]. For Patients K and L, only one sample set was obtained. A third sample set was obtained at approximately 1 week after the initial samples for Patients A and E. All samples were obtained between 10 and 11 am and between 1 and 3 pm to avoid mealtimes and to minimize the possible effects of diurnal variation in saliva composition (9).

Saliva pH was determined immediately after sample collection for Patients E–L. The serum and saliva samples were refrigerated at 4° and analyzed within 2 days. Serum and saliva concentrations of procainamide were assayed by a double-extraction, spectrophotofluorometric method (1, 10).

[1] At the Millard Fillmore Hospital.

[2] Pronestyl.

[3] Parafilm.

Table I—Procainamide Dosage Regimens and Serum and Saliva Concentrations

Patient	Dosing Regimen: Dose, mg/kg/day	Dosing Regimen: Dosing Interval, hr	Sample 1: Hours before Dose	Sample 1: Saliva Concentration, μg/ml	Sample 1: Saliva pH	Sample 1: Serum Concentration, μg/ml	Sample 2: Hours after Dose	Sample 2: Saliva Concentration, μg/ml	Sample 2: Saliva pH	Sample 2: Serum Concentration, μg/ml
A[a]	41.3	4	0.50	16.6	—[b]	16.3	1.5	23.7	—[b]	23.6
B	39.8	3	0.50	10.8	—[b]	3.4	1.0	9.7	—[b]	3.5
C	27.0	6	0.25	19.3	—[b]	4.9	2.0	22.3	—[b]	7.1
D	23.1	6	0.50	15.0	—[b]	2.4	2.0	32.5	—[b]	4.4
E[a]	25.6	4	0.12	4.4	7.2	1.6	2.0	13.6	6.8	1.9
F	36.7	4	0.25	4.4	8.0	3.1	1.5	7.4	7.5	4.7
G	28.3	4	0.25	4.1	7.1	2.1	1.5	0.7	7.1	2.6
H	41.3	4	0.50	8.7	7.1	4.4	1.0	10.9	7.0	4.1
I	36.7	3	0.12	18.8	6.7	2.5	1.0	22.8	6.6	2.6
J	47.6	4	1.0	12.7	6.2	9.4	1.0	47.5	6.3	8.2
K	47.5	3	0.25	18.9	6.4	4.8	—	—	—	—
L	30.0	3	0.25	13.1	6.3	3.4	—	—	—	—

[a] Third samples were also collected for these patients. [b] Saliva pH was not measured.

RESULTS

The serum and saliva procainamide concentrations measured in the 12 subjects are listed in Table I. There was no correlation between the dose (milligrams per kilogram per day) and either the minimum serum or saliva concentration of procainamide. No attempt was made to correlate the drug dose with the maximum concentrations since a single sample taken 1–2 hr after the dose would not be expected to identify accurately the actual peak concentration. These observations are in agreement with the findings of Koch-Weser and Klein (1), who noted a substantial degree of intersubject variability in serum concentrations of procainamide among patients receiving similar doses.

The saliva–serum concentration ratio of procainamide is plotted *versus* the serum concentration of the drug in Fig. 1. Pronounced intersubject variation was observed in this ratio, with values ranging from 0.27 to 8.93. Substantial intrasubject variation was also observed. A notable example is Patient E, in whom the ratio varied from 2.75 to 7.16 to 3.28 on three different occasions.

One possible reason for this variation was thought to be the differences among patients with respect to saliva pH. Procainamide is a weak base with a pKa of 9.4. The unionized form of the drug is more lipophilic than the cation and would be expected to diffuse more freely across mucosal membranes and the epithelial cells lining the salivary ducts. The relationship between the saliva–serum concentration ratio of procainamide and saliva pH is shown in Fig. 2.

The theoretical distribution ratio expected for the total concentration of an ionizable species found on two sides of a lipoid membrane where differences in pH exist is also shown in Fig. 2. This ratio is described by the following relationship derived from the Henderson–Hasselbach equation:

$$\frac{C_{sal}}{C_{ser}} = \frac{1 + 10^{(pKa - pH_{sal})}}{1 + 10^{(pKa - pH_{ser})}} \qquad \text{(Eq. 1)}$$

where the subscripts sal and ser refer to the respective concentration (C) or pH values for saliva and serum. Serum pH values of 7.2–7.6 were used, reflecting the lower and upper limits of the theoretical range. There was a significant correlation ($r = 0.57$, $p < 0.05$) between the actual and theoretical ratios (at a serum pH of 7.4).

Besides accounting for much of the intersubject variability in salivary distribution of procainamide, the differences in saliva pH also accounted for intrasubject changes in the distribution ratio. For each patient who exhibited differences in saliva pH between the two collection periods, the change in ratio was in the direction predicted by Eq. 1.

DISCUSSION

The measurement of procainamide concentrations in saliva is not a useful method for monitoring the plasma concentration of procainamide in patients. The variability in the saliva–serum concentration ratio is large and unpredictable. Even if saliva pH measurements were made for the purpose of applying a correction factor based on Eq. 1, the range of saliva concentrations found at any given pH is appreciable. For example, three patients had saliva pH values near 7.0 and the saliva–serum concentration ratio varied more than fourfold (Fig. 2).

The variability in salivary distribution of procainamide may be partly due to several other factors. Since the drug is only 15% bound to serum proteins (10), protein binding is not a likely factor. The contact time of procainamide with the buccal mucosa may be of slight importance. Meyers *et al.* (11) instilled buffered (pH 5–11) solutions of procainamide into the mouth and measured the loss of drug after 5 min. The absorption rate of procainamide was slow; only 4.5 and 14.3% absorption occurred at pH 5 and 11, respectively. The magnitude of these differences in absorption rate is much less than anticipated based on the degree of ionization of procainamide. However, these investigators did not measure the pH of the solution after equilibrium in the mouth. Saliva contains sufficient bicarbonate (40–50 mEq/liter) (12) to alter the pH of weakly buffered acidic solutions and possibly mute the pH differences.

A major factor which may affect the distribution ratio is the intrinsic and effective pH values at the site of saliva secretion. Serum pH may not adequately reflect the pH of the interstitial fluid on one side of the salivary glands while saliva pH, after mixing with the contents of the mouth, may differ from that which was formed by the gland and which contacts the epithelial cells within the gland. Thus, the theoretical curves shown in Fig. 2 may only approximate the effects of real pH differences across the salivary gland on a microscopic level. Finally, the rate of secretion of saliva affects the saliva concentration of some small molecules. For example, the concentration of urea in saliva decreases with the rate of salivary flow (13).

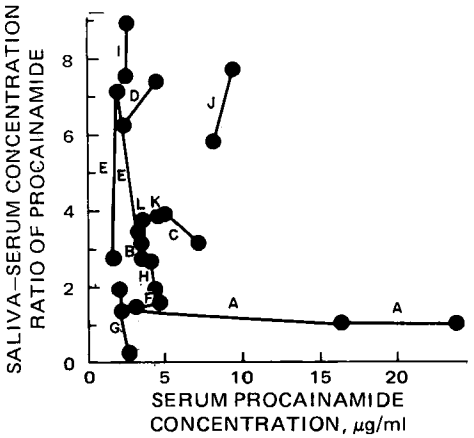

Figure 1—*Range of saliva–serum concentration ratios of procainamide as a function of serum concentration of procainamide in 12 patients during therapy with the drug. Lines connect data obtained repeatedly in individual patients. Letters identify data points for each patient in Table I.*

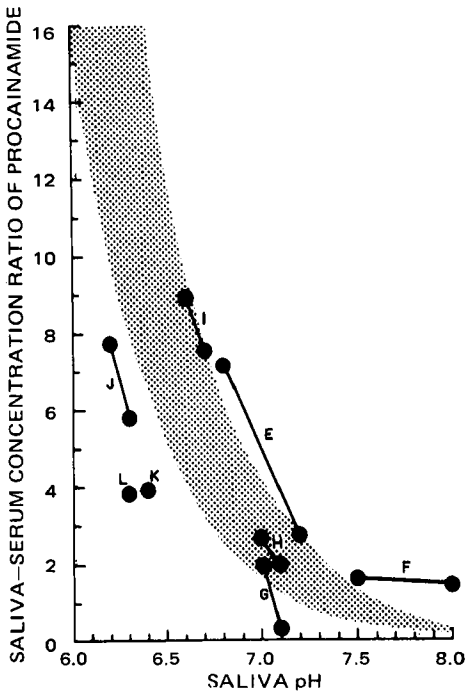

Figure 2—*Relationship between saliva–serum concentration ratios of procainamide and salivary pH in individual patients. The shaded area shows the theoretical concentration ratios calculated from Eq. 1, using a serum pH range of from 7.2 (lower edge) to 7.6 (upper edge).*

Examination of the effects of modifying pH on procainamide secretion into saliva in individual volunteer subjects does not appear feasible. Levy and Lampman (14) found that administration of large doses (5 g) of sodium bicarbonate did not significantly increase the pH of saliva of healthy subjects while urine pH changed appreciably. Doses of ascorbic acid or ammonium chloride were similarly without effect on saliva pH in single preliminary tests in this laboratory. Thus, the only reasonable approach to study the effect of pH on drug secretion into saliva is the present one, namely, a panel of subjects or patients with naturally different salivary pH values.

Other drugs have exhibited salivary secretion rates that were dependent on pH and protein binding. Gruneison and Witzgall (8) found a linear relationship between the saliva concentrations of several sulfonamides and the unbound drug in plasma of humans. Differences in ratios among the sulfonamides were consistent with the degree of ionization of these drugs. Sulfanilamide (pKa 10.4) produced the highest saliva–serum water concentration ratio (1.08), while sulfadiazine (pKa 6.28) yielded a ratio of 0.338. These studies were performed at only one salivary pH value (slightly lower than 7.4). However, the data of Rasmussen (15) on the same two drugs in cows and goats are of interest. These animals had a saliva pH of 8.2. Sulfanilamide, which would be essentially 100% unionized at pH 7.4 and 8.2, had a saliva–serum ultrafiltrate ratio of 1.0. Sulfadiazine, which would be 2% unionized at pH 8.2 and 11% unionized at pH 7.4, had a saliva–serum ultrafiltrate ratio of 1.3 in the animals.

Another weak base, trimethoprim (pKa 7.3), appears to behave like procainamide with respect to pH-dependent salivary secretion. Hansen *et al.* (16) reported a range in saliva-to-plasma concentration ratios of 1–8.3 in 14 patients. With the use of Eq. 1, the theoretical concentration ratios over a saliva pH range of 6.3–8.0 is 7.6 to 0.7, in close agreement with observed data.

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