SOLID-STATE DISTRIBUTION ANALYSIS USING THE ELECTRON MICROPROBE

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While conventional analytical techniques are not amenable to solid-state distribution analysis, the electron microprobe is easily adapted to this type of study. Sulfisoxazole tablets were subjected to microprobe analysis to demonstrate the applicability of using this instrumentation on solid, pharmaceutical-dosage forms. Chemical assay and microprobe analysis indicated a uniform distribution of sulfisoxazole in the tablets from several manufacturers.

Conventional analytical techniques used in the pharmaceutical industry are directed at qualitive identification or bulk quantitative determination of the therapeutic component of a dosage form. In particular, the USP content uniformity test (1) specifies that twenty whole tablets be assayed. Depending on the particular drug, several assays may be available to the analyst, yet few of these are specifically adaptable to distribution analysis.

Distribution analysis of a drug in a solid dosage form would provide valuable information concerning the uniformity of distribution of the therapeutic component within the dosage form. Additional applications, unique to particular formulations, could be proposed, e.g., following the migration of a volatile component as a function of time or observing solid-state diffusion.

The electron microprobe, which combines features of electron microscopy with those of x-ray diffraction spectroscopy (2), may be used to define the distribution of a drug in individual tablets. In microprobe analysis, when a focused beam of electrons is allowed to impinge on a solid target, excitation of electrons to a higher energy level results. As these energized electrons return to their original energy state, x-rays characteristic of the element whose electrons were energized are emitted. If the element is unique to the drug in question, that drug may be traced, in a distribution analysis of the solid sample, by moving the beam to scan the surface. A major limitation in microprobe distribution analysis is that the drug moiety must contain a unique element, one which can be distinguished from the remainder of the matrix.

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The present study was undertaken to investigate possible utilization of the electron microprobe for distribution analysis of solid dosage forms.

METHODS

Sulfisoxazole tablets (500 mg) were chosen as a model system for several reasons: (a) sulfur is unique to the sulfisoxazole entity in the tablet; (b) because a large amount of sulfur is present, signal intensity should not be a problem; (c) tablets are scored, an indication that the distribution is uniform to ensure proper therapeutic response to a portion of the tablet. For a review of assay procedures available for sulfisoxazole analysis, see Agarwal, et al. (3).

The tablets to be analyzed were placed edgewise on an aluminum disc, usually four tablets on one disc, with the bisecting groove of the score perpendicular to the disc surface. Tablets were affixed to the disc by means of a Duco cement and a charcoal adhesive mixture containing approximately 5% charcoal. (The charcoal is necessary to provide a good conducting medium between the disc and the tablets.) The sample was placed on a lathe and one-half of each of the tablets was removed to expose the halved edge as a flat, smooth surface.

The tablets were then placed in a Japan Electron Optics Vacuum Evaporator where a conductive carbon layer, approximately 200 Å thick, was deposited. The sample was then secured in the sample carrier and inserted into the electron probe analyzer, a Norelco Electron Probe, model AMR3. An operating voltage of 20,000 volts was

used to minimize surface destruction. The beam diameter was 0.5μ . Most reported analyses were performed using an oscillating beam, producing an effective beam width of 320μ . The scans were run lengthwise at a rate of 625μ per min; each scan required about 20 min. Three tablets of each of eight brands marketed by different manufacturers were scanned.

The U.S.P. XVIII assay procedure for sulfisoxazole tablets was modified as to sampling technique, i.e., three whole single tablets were assayed individually, rather than as a group of 20 tablets, to ascertain the comparative uniformity among tablets. Three halves of tablets were also assayed individually as a basis for comparing the distribution of sulfisoxazole within the tablets according to the U.S.P. XVIII assay procedure with the results of microprobe analysis. The U.S.P. assay consisted of titrating the weakly acid sulfisoxazole with standard sodium methoxide in dimethylformamide to a thymol blue endpoint.

Several tablets were analyzed to ascertain that extraneous sulfur, i.e., other than that contained in the sulfisoxazole, was not present. These analyses were made by Galbraith Laboratories, Inc., Knoxville, Tenn.

RESULTS AND DISCUSSION

A summary of the results of U.S.P. assays and of microprobe analysis for distribution of sulfisoxazole in the eight brands of tablets examined is given in Table 1. It will be noted that both the U.S.P. assay and microprobe analysis indicate a uniform distribution of sulfisoxazole in the tablets.

TABLE 1. *Sulfisoxazole distribution by USP assay and by microprobe analysis*

	USP assay range[a]		Microprobe analysis[b] Distribution (%) in each half tablet	
Brand	Whole Tablet (mg)	Half Tablet (mg)	1st Half	2nd Half
I	491-503	253-255	44.7	55.3
II	492-501	248-252	53.1	46.9
III	498-510	233-251	48.9	51.1
IV	497-505	245-250	49.3	50.7
V	465-470	236-241	50.6	49.4
VI	478-502	239-240	49.4	50.6
VII	495-514	250-285	51.4	48.6
VIII	487-506	245-257	50.3	49.7

[a] Standard deviation on assay ± 3 mg.

[b] Average of three determinations.

$$\% \text{ Relative distribution} = \frac{\text{Area under scan of half tablet}}{\text{Area under total scan}} \times 100$$

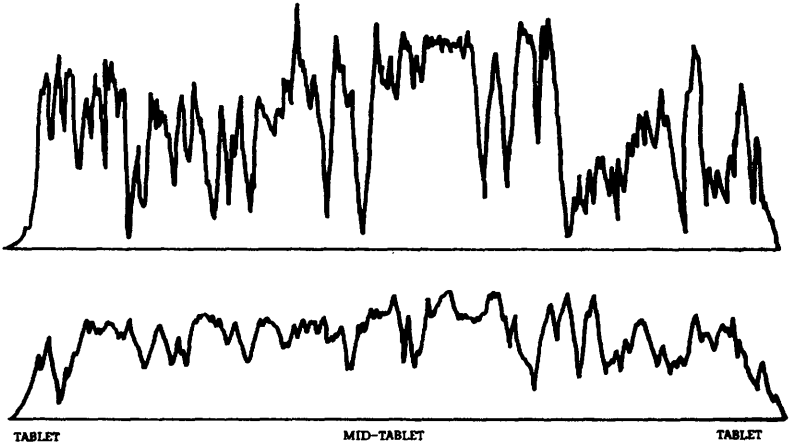

FIGURE 1. Comparison of results of microprobe analysis using electron beams of different widths. Upper scan = 0.5μ beam width; lower scan = 320μ beam width.

FIGURE 2. Demonstration of the reproducibility of the microprobe scan. Results of two consecutive scans of the same tablet over the same path are shown.

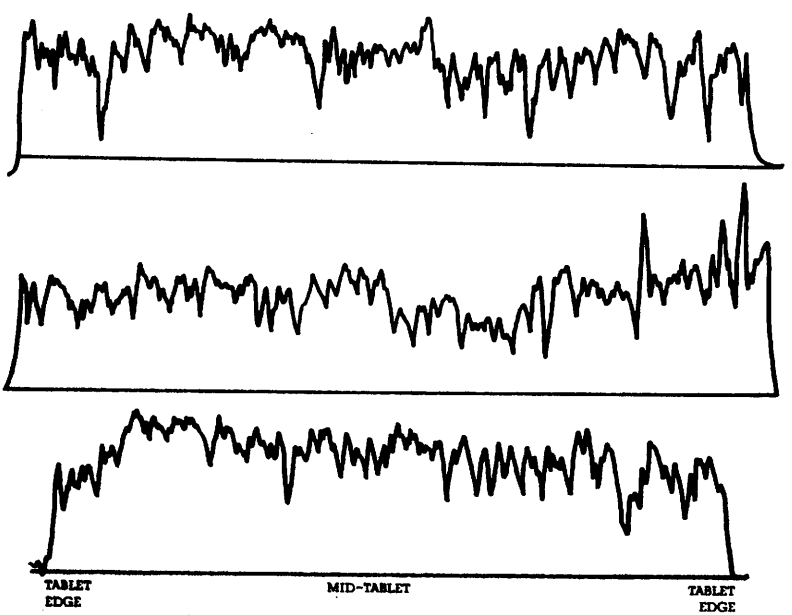

FIGURE 3. Representative scans of tablets from one manufacturer.

Results of the sulfur analyses, as presented in Table 2, verified that all the sulfur present was in the sulfisoxazole entity.

TABLE 2. *Percentage of sulfur in sulfisoxazole tablets*

Brand	Sulfur content	
	Theoretical[a] (%)	Experimental[b] (%)
VIII	9.13	9.39
VII	9.30	9.55
I	8.79	8.52
V	9.14	8.61

[a] Based on weight of tablet and the labeled amount of sulfisoxazole present (500 mg).

[b] Microanalyses by Galbraith Laboratories, Inc. Knoxville, Tenn.

Figure 1 shows a comparison of the two types of scans used in this study, i.e., those employing 0.5μ and 320μ beam widths. Most previously reported analyses have used an oscillating beam of 320μ width. The 320μ beam analyzes a larger surface and, in our experience, yielded a better representation of the distribution of the sulfisoxazole in the tablet. The intensity level appeared to be more uniform owing to the averaging of a larger area. There was a concomittant reduction in signal intensity when using the oscillating beam. This reduction resulted from the decreased energizing period the beam was on a surface point.

The reproducibility of the microprobe scan is demonstrated in Figure 2. Two consecutive scans of the same tablet over the same path are shown. A one-to-one intensity correlation between the peaks and valleys is observed; the peaks are high concentrations of sulfisoxazole while the valleys are sulfisoxazole-deficient areas in the tablet. The minor irregularities in the distribution most probably are a result of the granulation procedures used prior to tableting.

Figure 3 depicts representative scans of tablets from one manufacturer. The scans themselves are distribution curves of the sulfisoxazole in the tablet matrix.

Microprobe analysis provides unique instrumentation for obtaining distribution of a drug in a solid dosage form. The sample preparation is simple and requires little manipulative time. The information derived from the scans is easily interpreted and is representative of the distribution of the therapeutic ingredient.

REFERENCES

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