was analyzed to estimate its variability. The following findings are briefly summarized.

1. By performing a size analysis on the granulation and then determining the corresponding drug content of each sized fraction, it was possible to ascertain whether the drug is uniformly distributed throughout the different granules.
2. If the drug is not uniformly distributed, the technique of sampling is extremely important in obtaining a true representation of the drug distribution in the mix. Misleading estimates of variance were produced by the samples taken by *Method A*.
3. To determine the uniformity of drug content at different locations in the mixer, location samples were compared with random samples. Through the computation of standard deviations, it is possible to estimate the homogeneity of drug content in the mixer and to compare the same stage of manufacture for different batches.
4. In determining the standard deviation of the drug content at the different stages of granulation manufacture, it is possible to pinpoint the stage which is interfering with good mixing.

To evaluate weight and drug content variability of the tablets, random and systematic sampling plans were employed. The systematic sampling plan gave more information about the tablet compression operation than the random sampling plan. The following information was gained from this study.

1. The sampling technique is important for obtaining random samples.
2. The influence of compression time and compression sides of a double rotary press on tablet weight and drug content can be determined by analysis of variance.
3. The relationship between tablet weight and drug content was determined by computing the correlation coefficient and was not significant. This was because for each of the three batches of tablets studied, the coefficient of variation for drug content was about fivefold that of the tablet weight.

Although this report described certain methodology and sampling plans, it should be realized that similar and possibly more extensive information could be obtained through the use of other sampling plans. However, before any sampling plan is decided upon, careful consideration must be given to the factors that are to be evaluated. Only then is it possible to design the correct methodology and sampling plans which would result in the accumulation of sufficient random samples to evaluate adequately each factor and eliminate the possibility of taking samples from materials which may have undergone unmixing.

Additional studies presently are underway on different dosage forms using modified sampling plans to obtain maximum information regarding the relationship that may exist between drug heterogeneity in the several stages of granulation manufacture and the final tablets.

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Notes

Titrimetric Assay of Sulfonamides by Diazotization Using Ferrocyphen as Indicator

By W. M. BANICK, Jr., and J. R. VALENTINE

A study has been made concerning the use of ferrocyphen [dicyano-bis-(1,10-phenanthroline)-iron(II) complex] as an internal, reversible indicator for the diazotization titration of sulfonamides. Many of the sulfonamides of pharmaceutical interest can be determined using the ferrocyphen visual end point. The titration procedure is simple and rapid.

Most of the sulfonamides of pharmaceutical interest have the general formula $H_2NC_6H_4SO_2NHR$, in which the amino group and the sulfonamide group are in a *para* position to each other. Many different methods have been described (9) for the analysis of these sulfonamides. Diazotization of the primary amino group appears to be the preferred method, probably because it is applicable to nearly all sulfonamides and employs a titrant which is stable, readily available, and easily standardized.

The end point in the diazotization titration may

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TABLE I.—TITRIMETRIC ASSAY OF SULFONAMIDES USING FERROCYPHEN AS INDICATOR

Common Name	Structure H_2N—SO_2NHR, R=	% Purity[a,b]
Sulfanilamide	H	100.2
Sulfaguanidine	—C(=NH)NH_2	99.7
Sulfadiazine	2-Pyrimidinyl	99.9 (99.5)
Sulfamethazine	4,6-Dimethyl-2-pyrimidinyl	100.2 (100.4)
Sulfacetamide	Acetyl	100.0 (99.6)
Sulfachloropyridazine	6-Chloro-3-pyridazinyl	99.4 (99.9
Sulfachloropyrazine	6-Chloro-3-pyrazinyl	100.4 (100.2)
Sulfamethoxypyrimidine	6-Methoxy-4-pyrimidinyl	[c]
Sulfamethoxypyridazine	6-Methoxy-3-pyridazinyl	[d]
Sulfamerazine	4-Methyl-2-pyrimidinyl	[d]
Sulfapyridine	2-Pyridinyl	[d]
Sulfathiazole	2-Thiazolyl	[d]

[a] Average of at least four determinations. [b] Values in parentheses were obtained by a diazotization procedure employing a starch-iodide end point (2). [c] End point not stable. [d] Visual end point detection was impossible since titration gives colored products.

be determined potentiometrically (2, 7), amperometrically (6), with starch-iodide external indicator (9), with diphenylbenzidine disulfonic acid (1), or with orange IV (8).

Schilt (5) recently described the use of a metal complex as an internal, reversible indicator for the diazotization titrations of some primary aromatic amines (aniline, *p*-bromoaniline, *o*-chloroaniline, and 2,4-dichloroaniline). He proposed the trivial name "ferrocyphen" for the dicyano-bis-(1,10-phenanthroline)-iron (II) complex.

We have studied the use of this indicator in the diazotization titration of 12 sulfonamides of pharmaceutical interest. Seven of the sulfonamides can be titrated using ferrocyphen as indicator. The titration procedure using ferrocyphen as indicator is characterized by speed and simplicity.

EXPERIMENTAL

Reagents

Ferrocyphen [Dicyano-bis-(1,10-phenanthroline)-Iron (II)].—The reagent was prepared as the dihydrate using the procedure described by Schilt (4).

Ferrocyphen Solution, 1.0%.—Dissolve 0.50 Gm. of the reagent in 50 ml. of concentrated sulfuric acid.

Sodium Nitrite, 0.1 M.—Dissolve 14.2 Gm. of reagent grade sodium nitrite in distilled water to give 2000 ml. of solution. Standardize against purified sulfanilic acid as follows. Dissolve an accurately weighed sample of about 0.7 Gm. in 25 ml. of distilled water and 5 ml. of 1.0 *M* sodium hydroxide. Add 200 ml. of 6 *M* hydrochloric acid, 1.0 ml. of the ferrocyphen solution, and titrate with the 0.1 *M* sodium nitrite. At the end point, the color will change from yellow to violet. The end point is stable at least 3 minutes. Run a blank titration on the reagents.

Sulfonamides.—All the sulfonamides used in the investigation were equivalent to U.S.P. grade.

Procedure

Accurately weigh about 0.5 Gm. of the sulfonamide and dissolve in 100 ml. of 6 *M* hydrochloric acid. Add 1.0 ml. of the 1.0% ferrocyphen solution and titrate with 0.1 *M* sodium nitrite. Introduce the titrant under the surface of the solution with a buret having an elongated tip. In the vicinity of the end point, withdraw the tip and complete the titration dropwise to a pale violet or rose end point which is stable at least 3 minutes. Run a blank titration.

$$\% \text{ Purity} = \frac{A \times M \times F.W.}{G \times 10}$$

where A is the net ml. of M molar sodium nitrite, $F.W.$ is the formula weight of the sulfonamide, and G is the Gm. of sample.

RESULTS AND DISCUSSION

The results obtained for 12 sulfonamides analyzed by the diazotization procedure are summarized in Table I. Since previous investigators (2, 6) have shown that titration at 5–10° is not a necessary requirement for the diazotization titration, all the titrations using ferrocyphen as indicator were performed at room temperature (25°). The purity values obtained using ferrocyphen as indicator are in good agreement with the values obtained by a procedure employing a starch-iodide end point (3).

In all the titrations for which quantitative data were obtained, the ferrocyphen responded reversibly in the same way as described by Schilt (5). The diazotization of all the sulfonamides and the indicator response were extremely rapid in the 6 *M* hydrochloric acid.

Visual end point detection was impossible for the last four compounds in Table I because titration gives colored products (yellow or yellow-orange) which obscure the end point.

Precision.—The standard deviation calculated using 29 degrees of freedom is ±0.24%.

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