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Research Paper

The USP Performance Verification Test, Part I: USP Lot P Prednisone Tablets—Quality Attributes and Experimental Variables Contributing to Dissolution Variance

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Purpose. Beyond instrumental qualification, proficiency testing is not usually a prerequisite for many analytical procedures, given reliance on a manufacturer's assay validation coupled with regulatory review and inspection. Given the special features of the dissolution procedure, proficiency testing was put in place initially by pharmaceutical manufacturers and carried on by USP. Proficiency testing is designed to help ensure that execution of a dissolution procedure for solid oral dosage forms adequately supports administrative and legal decisions so that measurements made at different times, by different analysts, or with different methods can be confidently compared. USP has applied metrological principles to aid practitioners in carrying out the dissolution procedure alone and in collaborative studies to facilitate understanding potential sources of variability.

Materials and Methods. The present study aimed to identify key dissolution variables associated with USP Lot P Prednisone Tablets in conjunction with the USP Performance Verification Test (PVT). Using five dissolution test assemblies from different manufacturers, at least four of six analysts determined percents prednisone dissolved on dissolution Apparatus 1 (basket) and Apparatus 2 (paddle) on each assembly. Six replicate experiments were performed on each analyst–assembly combination with a set of six to eight tablets in each experiment.

Results and Conclusions. Statistical analysis demonstrated that dissolution test assemblies were the largest factor contributing to dissolution variability. Inherent tablet variability was low, and USP Lot P Prednisone Tablets did not contribute importantly to dissolution variability. Contributions from analyst and analytical procedure also were estimated to be low.

KEY WORDS: dissolution; performance verification; performance verification test; quality assurance; United States Pharmacopeia.

INTRODUCTION

For a specified solid oral dosage form, the dissolution procedure is primarily a quality control tool in the absence of an *in vitro–in vivo* correlation (IVIVC). It assesses the "performance" component of quality. Procedures and general acceptance criteria for dissolution are described in the *United States Pharmacopeia* (*USP*) General Chapter *Dissolution* <711> (1). When adapted to a specific solid oral dosage form, the procedure with acceptance criteria becomes one of several tests in either a private or public compendial dosage form specification, as described, e.g., in a *USP* dosage form monograph. Dissolution indicates acceptable bioavailability (BA) and bioequivalence (BE) if the appropriate scientific links are established and maintained with these developmental characterization studies (2). When a dissolution method is correlated to *in vivo* performance by an IVIVC or can be relied upon in the application of the biopharmaceutics classification system (BCS) to allow waiver of an *in vivo* study, dissolution is used increasingly in regulatory and World Health Organization (WHO) guidelines as a means of documenting BA and BE (3,4).

The dissolution procedure relies on a test assembly by which an analyst prepares samples to measure percent released from a dosage form as it dissolves over time. Test assemblies (*assemblies* hereafter) with different types of vessels, stirrers, temperature controls, and media have advanced technologically in recent years, with substantial improvement in capability. Metrologic science itself has advanced in this time, as a result of harmonizing efforts in national metrology organizations such as the National Insti-

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tutes of Standards and Technology and nongovernmental bodies such as the International Organization for Standardization (ISO) and the International Bureau of Weights and Measures (Le Bureau international des poids et mesures, BIPM). Both types of advances merit careful consideration regarding the dissolution procedure, given the latter's wide and increasing application. This is particularly important because the dissolution procedure is not an easy one and requires not only modern apparatus but also carefully trained personnel, a well-validated analytical procedure for collected samples, and close attention to detail.

Many elements are involved in ensuring the integrity of the dissolution procedure and focus on the assembly itself, including instrumental qualification (IQ) and operational qualification (OQ). OQ is performed by mechanical calibration, usually at 6-month intervals. Performance qualification (PQ) is performed by conduct of an Apparatus Suitability Test as described in <711>, again usually at 6-month intervals. USP provides official USP Reference Standard Tablets for PQ, containing, for *USP* dissolution Apparatus 1 and 2, either prednisone or salicylic acid, together with acceptance criteria drawn from a collaborative study (5). When supplied with a technical data sheet and troubleshooting guide, USP's Reference Standard (RS) Tablets can be used by first parties (manufacturers), second parties (purchasers), and third parties (independent or governmental laboratories) to determine whether results within their laboratories are similar to the results from the USP collaborative study (6,7). Thus, USP's RS Tablets are not *calibrator* tablets—they are used in performance verification—and USP will no longer use the term *calibrator* to describe them. USP RS Tablets are used in proficiency testing in which a single laboratory assesses its capability relative to laboratories in the USP collaborative study. Similarly, the USP Biopharmaceutics Expert Committee has abandoned the term "Apparatus Suitability Test" in favor of the technically more accurate Performance Verification Test (PVT). The present paper, the first of two, uses USP Lot P Prednisone RS Tablets to investigate the sources of intralaboratory variation, and the second paper invokes metrological science (8–10) to examine interlaboratory variation.

Dissolution testing has long been associated with issues of repeatability and reproducibility (2,11,12). USP Prednisone RS Tablets were first introduced to industry in 1978 as a collaborative effort of pharmaceutical manufacturers and USP to address these issues, focusing especially on interlaboratory variability. Over the years, a persistent concern regarding the USP Prednisone RS Tablets has been the broad range of their acceptance criteria. For example, the current USP Lot P Prednisone RS Tablets, when used in a PVT, carry a specification of 47–82% dissolved at 30 min for Apparatus 1 and 37–70% dissolved at 30 min for Apparatus 2. These acceptance criteria are based on the variable results obtained during collaborative testing in different laboratories and have led some to believe that the Prednisone RS Tablet itself is the major source of variability.

As noted, dissolution testing is a sequence of complex processes, including sample preparation in a test assembly followed by a conventional analytical procedure to determine percent of label claim dissolved over time. Therefore many experimental variables such as instrument, analyst, and analytical method can affect the final test results. In order to assess these variables, this paper reports on dissolution variance studies using USP Lot P Prednisone RS Tablets in dissolution Apparatus 1 (basket) and Apparatus 2 (paddle). Because our focus is on use of Prednisone RS tablets in the dissolution PVT, we restrict attention to dissolution as measured in the PVT, namely only at 30 min. As Part I of an investigation of Performance Verification Testing (PVT), this paper outlines metrological aspects of the *USP* PVT and the experimental approach to address repeatability and intermediate precision. Part II shows the results of a collaborative study that addresses the interlaboratory variability or reproducibility (5).

MATERIALS AND METHODS

Lot P Prednisone Tablets

USP Lot P Prednisone Tablets were prepared under cGMP at Aptuit (Kansas City, MO) under stringent quality control/assurance.

Chemicals

USP provided all RS materials, including Lot M Prednisone RS, Lots O and P Prednisone RS Tablets, Salicylic Acid Lot Q RS Tablets, Salicylic Acid Lot J RS, and Acetanilide Lot M RS. Milli-Q water was used for dissolution media preparations. Ethyl alcohol USP, methanol, and tetrahydrofuran were obtained from Fisher (Pittsburgh, PA). All chemicals were reagent grade or better.

Equipment

Equipment used in the study included a Distek HC 97 Hardness Tester; a Distek DF-3 Friabilator; a VanKel VK100 Disintegration Tester; a Perkin Elmer Lambada 40 UV–Vis spectroscopy apparatus; and an HP 1100 HPLC system.

Dissolution Assemblies

Five assemblies from five different manufacturers were used in this study, as follows: Sotax AT7Smart, Hanson SR8Plus, Distek Evolution 6100, VanKel 7010, and Erweka DT700. An *assembly* is defined as an integrated system consisting of tester (bath), vessels, shafts, and baskets or paddles as the stirring elements. These assemblies have been identified only by Greek letters to avoid disclosure of individual assembly performance.

Section I—Quality Attributes of Lot P Prednisone RS Tablets

The USP Lot P Prednisone RS Tablets were tested for appearance, hardness, friability, disintegration, assay, content uniformity, and weight variation using *USP* compendial methods.

Section II—Dissolution Experiments for Lot P Prednisone RS Tablets

General Study Design

Dissolution experiments of USP Lot P Prednisone RS Tablets on Apparatus 1 and Apparatus 2 were conducted on

five dissolution assemblies referred to as Alpha, Beta, Gamma, Delta, and Epsilon by at least four of six analysts termed A, B, C, D, E, and F on each assembly. Six, seven, or eight tablets were examined in each experiment depending on the capacity of the dissolution assembly. Six experiments were performed for each analyst–dissolution assembly combination, leading to the evaluation of almost 1,000 tablets on each apparatus.

Study Characteristics

To minimize the effects of random experimental errors, all vessels, shafts, baskets, and paddles were individually identified and kept in the same positions for all experiments. Efforts were also made to keep the vessel orientation the same for all experiments by aligning a mark on the vessel and a corresponding mark on the baseplate of the assembly.

Operational and Performance Qualification

For operational qualification (OQ) prior to conducting the dissolution test with the USP Lot P Prednisone Tablets, preventive maintenance was performed on all dissolution assemblies by their own manufacturers. As a further part of OQ, analysts verified that all mechanical check results met the mechanical calibration requirements in <711>. To ensure performance qualification (PQ), the USP PVT with USP Lot Q Salicylic Acid Tablets and Lot O Prednisone Tablets was carried out on all dissolution assemblies for both Apparatus 1 and 2. All results conformed to the then current *USP* acceptance range for the specified lots. All the participating analysts were fully trained and experienced in the conduct of the dissolution experiments.

Dissolution Procedure

Dissolution was carried out at 37 ± 0.5°C in 500 mL of Milli-Q water deaerated according to <711> at a rotation speed of 50 rpm, and six, seven, or eight prednisone tablets were tested in each dissolution assembly, depending on its configuration. Experiments were started, staggered at 1 min intervals, immediately after the temperature of the medium was equilibrated. At 30 min, about 35 mL of the dissolution solution was withdrawn manually from each vessel, and sample solutions were filtered immediately through a 0.45-μm disk filter (Millipore Millex-HV). The first 5-mL portion of filtrate was discarded prior to collecting and cooling to ambient temperature for UV analysis at 242 nm.

Statistical Analysis

Statistical analyses were used to identify the sources of variability in dissolution results. For the initial analyses, log transformed data [see Part II of the series (5)] from each of the five dissolution assemblies were analyzed separately. This was the primary analysis, chosen to examine sources of variability separately for each assembly. The first analysis for each assembly was a variance components analysis of variance of the logarithm of percent dissolved. Standard deviations (S) in the natural log scale were transformed to coefficients of variation (CV) in the original scale by $CV = \sqrt{\exp(S^2) - 1}$. Random effects were analyst, experiment within analyst, and position of vessel within the dissolution assembly. Because of the manner in which the apparatus was used, position represents the combination of actual position in the apparatus as well as vessel, shaft, and paddle. A residual variance incorporates all other sources of variance, including the inherent variability of the tablets, assay variability, and any variability associated with placing the tablet into the vessels. CV results reported are for the variance components. The total CV was found by first summing all the variances in the log scale and then converting to CV as above. Residual variability as a percent of total was determined from the variances in the log scale. The second analysis for each apparatus was like the first but treated vessel position as a fixed effect. This was done to examine whether some assemblies had "hot spots." For an additional set of analyses suggested by the initial results, the data from the five assemblies were analyzed together. The specific analysis was for variance components, as above, with assembly as a fixed effect and position nested within assembly as a random effect. Models were fit assuming that the variance components were constant across assemblies and then allowing the residual and position variances to vary by assembly. The Aicke Information Criterion (AIC) was used to compare statistical models with different variance assumptions.

All analyses were done with SAS for Windows version 9.1 (SAS, Inc., Cary, NC) using Proc Mixed and the defaults of REML and variance components.

Table I. Physical and Chemical Characteristics of USP Lot P Prednisone Tablets

Test	Mean Value[a]	SD[b]	%RSD
Appearance	Round in shape and white in color	NA	NA
Weight variation (mg)	222.0 ($n = 108$)	2.4	1.1%
Hardness (N)	64.4 ($n = 10$)	4.2	6.6%
Friability (% of weight loss)	<0.1%	NA	NA
Disintegration (s)	18 ($n = 18$)	<0.5	<3.0%
Prednisone assay (% of label claim)	96.2% ($n = 20$)	0.8%	0.8%
Prednisone content uniformity (mg/tablet)	9.7 ($n = 30$)	0.2	2.1%
Disintegrant content uniformity (mg/tablet)	4.5 ($n = 10$)	0.1	2.2%

[a] n = the number of tablets tested. See MATERIALS AND METHODS for details.
[b] SD = the standard deviation.

Table II. Summary of Percent of Prednisone Dissolved for Different Assembly–analyst Combinations on Apparatus 1 (Baskets)

		Percent Prednisone Dissolved[a]			
Assembly	Analyst	Minimum–Maximum	Mean[b]	SD	%RSD
Alpha	A	50.6–74.7	63.5	5.7	8.9
	B	51.5–79.9	62.6	6.5	10.3
	C	46.2–73.9	59.3	5.6	9.5
	D	55.9–75.4	64.5	5.0	7.7
Beta	B	52.5–78.2	61.3	6.3	10.2
	C	52.7–74.1	61.7	5.2	8.4
	D	48.9–71.7	59.3	5.5	9.3
	E	53.2–73.7	62.8	5.4	8.7
	F	50.4–79.4	63.1	6.4	10.2
Gamma	A	51.2–62.8	58.6	3.2	5.5
	B	44.8–63.5	56.2	3.8	6.8
	C	52.3–67.8	57.8	3.6	6.1
	D	48.5–74.4	57.8	4.8	8.4
Delta	A	50.7–78.6	59.3	6.0	10.1
	B	51.1–72.4	58.1	4.3	7.3
	C	43.2–66.3	52.5	5.6	10.6
	D	50.9–70.0	60.6	5.3	8.7
Epsilon	B	50.7–71.3	62.0	5.2	8.5
	C	45.1–71.7	60.1	5.4	8.9
	D	42.8–70.8	58.9	5.8	9.8
	E	48.7–75.3	61.4	5.8	9.5

[a] Conditions: 50 rpm in 500 mL deaerated water at 30 min on Apparatus 1 at 37 ± 0.5°C.
[b] Mean values were the average of six dissolution experiments for each assembly–analyst combination with six to eight tablets in each experiment.

RESULTS

Section I Results—Quality Attributes of USP Lot P Prednisone Tablets

None of the nearly two thousand tablets tested in this study displayed any observable defects. All were found to be white and round with smooth edges. Table I summarizes the analytical testing results of the quality attributes of the Lot P Prednisone Tablets. The physical and chemical properties, particularly the weight variation, assay, and content uniformity, directly describe the intrinsic tablet-to-tablet variability. All results met the corresponding acceptance criteria of the *USP* monograph for Prednisone Tablets as articles of commerce.

Section II Results—Apparatus 1: Dissolution Experiments on Lot P Prednisone Tablets

Prednisone tablets disintegrated rapidly in the rotating baskets. At the end of testing, the majority of disintegrated solid residuals remained within the baskets. Small amounts of solid particles were observed on the bottom of vessels in irregular patterns. Table II summarizes the dissolution values of different assembly–analyst combinations. The observed mean percent prednisone dissolved is consistent across all analysts and assemblies. Although not significantly different, Assembly Gamma showed somewhat lower variability.

Statistical analysis results for Apparatus 1 are summarized in Table III. Compared to analyst, position, and experiment, the major contribution to variability was residual,

Table III. Statistical Analysis Summary for Apparatus 1 (Baskets)

			CV%					
Assembly	Geometric Mean	95% Confidence Limits	Between Analyst	Between Position	Between Experiment	Residual	Total	Residual as % of Total Variance
Alpha	62.3	60.2–64.4	0.0%	2.0%	0.9%	9.0%	9.2%	94.6%
Beta	61.2	59.8–62.6	0.0%	1.4%	0.6%	9.1%	9.3%	97.2%
Gamma	57.3	55.2–59.4	2.1%	1.9%	0.0%	7.9%	8.4%	89.0%
Delta	57.3	51.7–63.5	6.3%	1.2%	0.0%	9.0%	11.1%	66.4%
Epsilon	60.6	58.6–62.6	0.0%	3.2%	0.6%	8.8%	9.4%	87.9%

which represented about 90% of total variance. This variance could be attributed to a number of factors not considered in the experimental design, e.g., tablet placement and environmental factors. The data exhibit excellent precision between experiments (Table III, column six). There are statistically significant difference across the assemblies' geometric means ($p < 0.001$), although the differences are not large. The residual and position variance components did not differ statistically significantly by assembly.

Section III Results—Apparatus 2: Dissolution Experiments on Lot P Prednisone Tablets

General Observations

USP Lot P Prednisone Tablets showed rapid disintegration upon introduction to the medium in the vessel. With Apparatus 2, the overall disintegration process took less than 20 s. A characteristic cone was formed at the bottom of the vessel within minutes after the start of the experiment. Table IV shows a summary of percent prednisone dissolved for different assembly–analyst combinations.

Assembly

Table IV summarizes results on all dissolution assemblies by different analysts. The results demonstrate the different performance characteristics among assemblies with respect to both the average percent dissolved and %RSD values, particularly on assemblies Alpha and Gamma. Results for assembly Gamma show the best precision (lowest %RSD) among the five assemblies investigated across all analysts. The observed mean percent prednisone dissolved is consistent across all assemblies and analysts—with the exception of assembly Alpha, which was also the assembly exhibiting the most variability.

In an initial experiment to understand the effects of different assembly and vessel combinations, dissolution vessels were exchanged between Alpha and Gamma, and the experiment was repeated. The results are shown in parenthesis in Table IV. Interestingly, the mean percent prednisone dissolved and variability (%RSD) with Alpha were reduced by the exchange. For assembly Gamma, variability increased. These results clearly indicate different dissolution results were obtained by vessel switching. Recently completed studies in

Table IV. Summary of Percent Prednisone Dissolved for Different Assembly–analyst Combinations for Apparatus 2 (Paddles)

		Percent Prednisone Dissolved[a]			
Assembly	Analyst	Minimum–Maximum	Mean[b]	SD	%RSD
Alpha	A	45.4–74.4	61.2	8.5	13.8
	B	44.0–74.9	57.2	9.0	15.8
	C	41.8–70.8	53.7	9.4	17.5
	D	41.3–71.3	55.5	9.5	17.1
	B	(38.2–45.9)[c]	(41.4)	(1.7)	(4.2)
	C	(39.6–54.2)	(44.7)	(2.9)	(6.6)
Beta	B	42.8–56.1	47.9	2.7	5.7
	C	41.8–66.5	46.9	4.6	9.9
	D	42.2–72.6	48.8	7.0	14.5
	E	38.8–70.0	52.7	8.9	16.8
	F	40.0–67.2	49.1	7.2	14.7
Gamma	A	38.5–50.8	46.0	2.3	4.9
	B	42.0–51.5	46.5	2.0	4.3
	C	40.2–47.1	43.7	1.7	3.8
	D	43.7–54.3	47.7	2.6	5.4
	F	37.8–45.8	41.4	1.8	4.3
	B	(40.4–71.3)[c]	(49.7)	(7.6)	(15.3)
	C	(40.6–53.4)	(44.0)	(2.6)	(5.9)
	E	(37.9–49.3)	(42.9)	(2.6)	(6.2)
Delta	A	41.5–51.2	44.9	1.9	4.2
	B	38.1–68.9	44.5	7.2	16.1
	C	43.5–69.4	47.3	5.3	11.3
	D	41.6–57.5	45.6	3.4	7.5
Epsilon	B	42.6–65.2	47.1	4.3	9.2
	C	43.5–76.9	48.9	6.8	13.9
	D	41.8–68.0	49.7	6.0	12.1
	E	39.7–48.2	43.4	1.8	4.1

[a] Dissolution conditions: 50 rpm in 500 mL deaerated water at 30 min on Apparatus 2 at 37 ± 0.5°C.
[b] Mean values were the average of six dissolution experiments for each assembly–analyst combination with six to eight tablets in each experiment.
[c] Values in parenthesis were obtained after dissolution vessels were switched between Alpha and Gamma.

Table V. Statistical Analysis Summary for Apparatus 2 (Paddles)

Assembly	Geometric Mean	95% Confidence Limits	CV% Between Analyst	CV% Between Position	CV% Between Experiment	CV% Residual	CV% Total	Residual as % of Total Variance
Alpha	58.7	(51.1–67.4)	6.6%	11.1%	1.6%	11.8%	17.7%	45.0%
Beta	48.4	(46.5–50.3)	0.0%	3.7%	1.0%	11.6%	12.3%	90.3%
Gamma	44.9	(41.4–48.6)	6.3%	1.0%	0.7%	4.3%	7.8%	30.8%
Delta	48.0	(43.2–53.3)	6.2%	2.3%	2.3%	8.1%	10.7%	57.4%
Epsilon	46.9	(43.0–51.0)	5.4%	3.9%	0.3%	8.7%	11.0%	63.2%

USP laboratories further confirmed that dissolution vessels are important sources of dissolution variability (13,14).

Results of statistical analyses are summarized in Table V. The largest effect is the difference between assemblies in geometric mean percent dissolved ($p < 0.001$). Assembly Alpha results exhibit a geometric mean of 58.7% compared to 47.0% for the other four assemblies combined. The degree of variability also differed considerably and statistically significantly among assemblies both for vessel position and residual variability. Results for assembly Alpha were considerably more variable with reference to position compared to the other four assemblies, and results for assembly Gamma exhibited less variability in terms of residual variability. The data shown in Table V again indicate excellent between-experiment precision. Fig. 1 illustrates the repeatability of dissolution experiments on assemblies Alpha and Gamma by different analysts. Even though greater vessel-to-vessel variation was observed on assembly Alpha within the experiment, experiment-to-experiment performances on both Alpha and Gamma assemblies were consistent across all analysts.

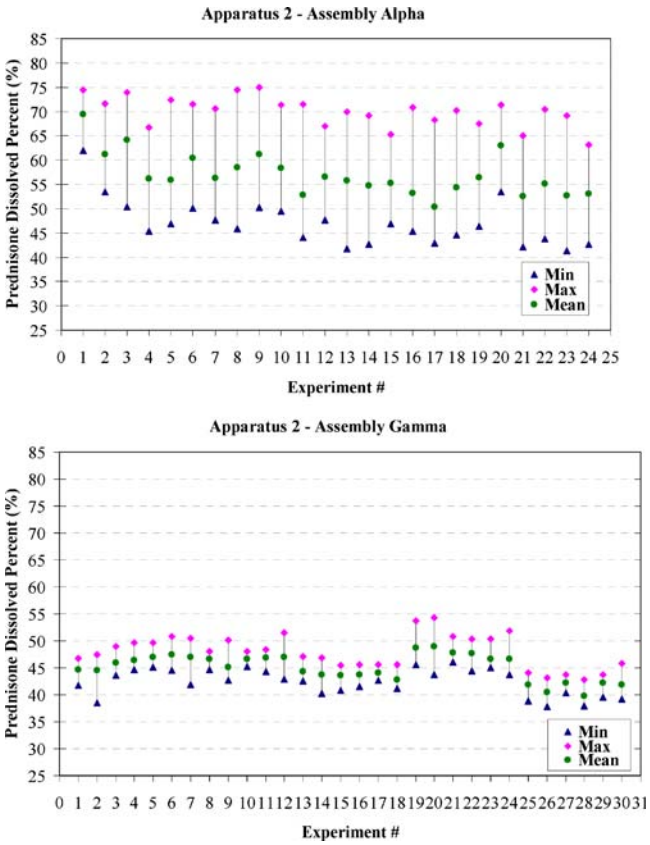

Fig. 1. Minimum, maximum, and arithmetic mean of percent prednisone dissolved for assemblies Alpha and Gamma (Apparatus 2) by different analysts: Experiments 1–6 are Analyst A; 7–12 are Analyst B; 13–18 are Analyst C; 19–24 are Analyst D; and 25–30 are Analyst F.

Figure 2 provides a direct comparison of data distribution patterns of dissolved prednisone on all five assemblies. The *y*-axis represents a relative frequency distribution in terms of percent of tablets exhibiting a particular percentage of dissolved prednisone. Distinct performance characteristics of different assemblies are evident. The dissolved prednisone data for assembly Alpha were relatively evenly distributed over the entire dissolution range of 41–75%. In contrast, the distributions of the other four assemblies indicate definite maxima. The maximum peak for assembly Gamma appeared at approximately 47% within a much tighter dissolution range of 37–54%. Assemblies Beta, Delta, and Epsilon showed significant tailing effects in the higher dissolution range of 55–75%. The total numbers of tablets tested on each assembly were in the range of 144–240 tablets. The spread of the distributions graphically represents the total variability given in Table V.

Analyst

As shown in Table V, between-analyst variability was consistent across assemblies—about 6%—but lower for assembly Beta. Experiment-to-experiment variability within analyst was low for all assemblies.

Vessel Position

For all five assemblies, the vessel positions differed statistically significantly on average ($p \leq 0.012$). As indicated in Table VI, four assemblies had one or two vessel positions that were high on average relative to the other positions; one assembly had two positions that were low on average relative to the other positions. For instance, positions 5 and 6 on assembly Alpha, position 3 on assembly Gamma, and position 1 on assembly Epsilon were high. Positions 2 and 4 on assembly Delta, and position 6 on assembly Epsilon were low. For some assemblies, high and low positions varied by analyst (data not shown).

Residual Variability

One key objective of this study was to determine the inherent tablet-to-tablet variability in dissolution. The upper limit for this variability was established by the analysis of residual variability in Table V. The residual variability includes the tablet assay and tablet placement variability in addition to the inherent

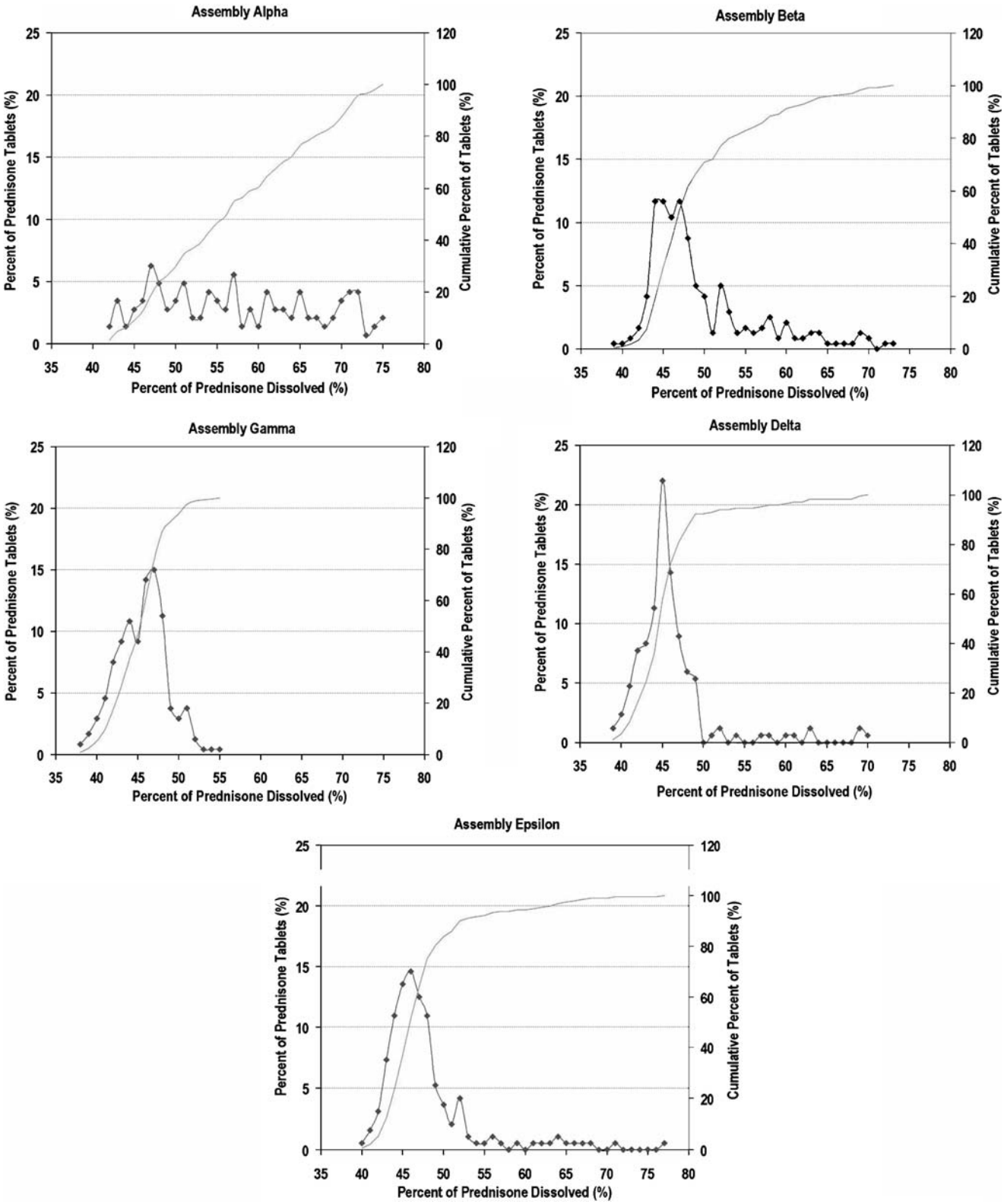

Fig. 2. Distribution of percent of prednisone dissolved for different assemblies for Apparatus 2 (Paddles).

tablet variability. The low residual value of 4.3% for assembly Gamma suggests that the inherent tablet variability must be less than 5%. This conclusion is also supported by the low %RSD values in Table IV on assembly Gamma across all analysts. It also accords with the primary quality data shown in Table I.

Analytical Variance

The variance of the final UV analysis was determined by the distribution of absorptivity of prednisone at 242 nm in the prednisone standard preparations. During this study, a total

Table VI. Percent of Prednisone Dissolved by Different Vessel Positions for Apparatus 2 (Paddles)

		Analyst[a]					
Assembly	Vessel Position	A	B	C	D	E	F
Alpha	1	57.1	*50.4*	54.4	50.2	NA[d]	NA
	2	61.4	52.2	48.6	56.1	NA	NA
	3	*53.2*[b]	57.0	55.1	50.9	NA	NA
	4	60.3	51.2	*44.1*	*45.9*	NA	NA
	5	67.7	61.0	52.6	**67.4**	NA	NA
	6	**67.8**[c]	**71.6**	**68.9**	62.3	NA	NA
Beta	1	NA	*46.5*	**50.6**	49.9	53.3	43.2
	2	NA	**49.6**	49.0	46.1	**62.7**	*42.6*
	3	NA	47.0	45.4	**55.5**	*43.3*	44.6
	4	NA	47.2	45.9	*43.9*	46.4	**52.4**
	5	NA	48.3	*43.6*	44.8	58.1	49.0
	6	NA	46.8	48.7	46.2	54.8	51.1
Gamma	1	45.7	47.4	44.0	*46.8*	NA	*39.9*
	2	45.9	46.9	44.9	46.9	NA	41.5
	3	**47.1**	**48.2**	**44.9**	**49.1**	NA	**42.8**
	4	46.3	*44.6*	42.3	48.7	NA	41.7
	5	46.4	46.1	*41.8*	47.7	NA	41.7
	6	*45.4*	46.0	44.2	48.1	NA	41.7
Delta	1	45.0	42.6	49.1	**46.4**	NA	NA
	2	44.2	*42.1*	*45.0*	44.7	NA	NA
	3	45.1	44.7	48.1	44.5	NA	NA
	4	*43.5*	42.2	46.2	*43.0*	NA	NA
	5	**45.6**	43.8	46.1	45.0	NA	NA
	6	45.0	**50.2**	**51.2**	45.5	NA	NA
Epsilon	1	NA	**53.8**	**59.3**	48.6	43.1	NA
	2	NA	45.4	46.6	47.2	42.3	NA
	3	NA	46.8	48.9	**51.8**	*42.1*	NA
	4	NA	46.3	48.7	50.5	44.0	NA
	5	NA	46.9	46.2	46.5	43.7	NA
	6	NA	*44.0*	*45.8*	*45.2*	**45.0**	NA

[a] All values are the average of six dissolution experiments for each assembly–analyst combination.
[b] Values in *italic* were the minimum values among the vessel positions.
[c] Values in **bold** were the maximum values among the vessel positions.
[d] NA = not investigated.

of 134 prednisone *Working Standard* solutions were prepared independently by all participating analysts. The absorptivity was calculated as 43.57 ± 0.65 (g^{-1} L cm^{-1}), resulting in a 1.49%RSD. The variance contribution of the final UV analysis to the overall variability was thus low.

DISCUSSION

With reference to the objectives of the study, we make the following observations:

- Prednisone RS Tablets were selected and developed to detect the impact of dissolution variables on results.
- Variable dissolution results obtained in this study are in agreement with historical values (5).
- The intrinsic dissolution variability of Lot P Prednisone RS Tablets was determined to be between 4 and 5%. The claim that Prednisone Tablets are the major

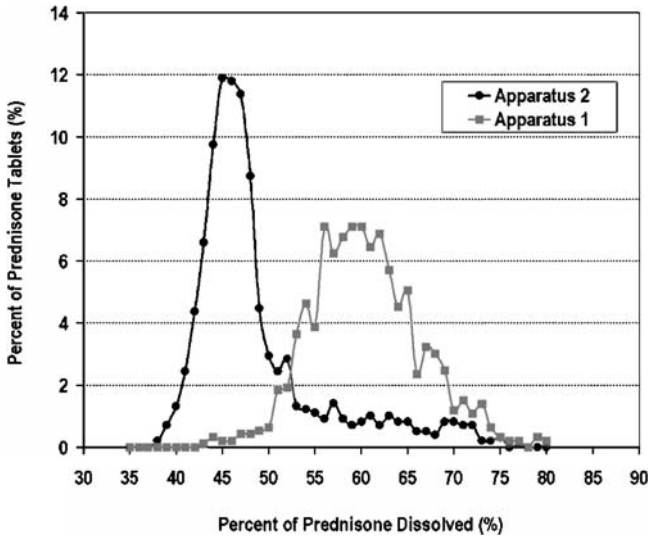

Fig. 3. Overall distribution patterns of percent of prednisone dissolved for Apparatus 1 (Baskets) and 2 (Paddles).

sources of variability in a PVT is a misinterpretation of dissolution data.

- Numerous studies in past decades tackled the issues of repeatability and reproducibility (intra- and interlaboratory variability) associated with dissolution testing (15–17). However, the sources of variability were still not well understood. In this study, the identities of dissolution assemblies were blinded throughout the study, care was taken to ensure consistent positioning of assemblies and all assemblies met current requirements of operational qualification and mechanical calibration. Still, this large study of a single physical standard, with varying experimental conditions, clearly demonstrates the very different performance characteristics of assemblies (see Assembly, above, and Table IV for data about assemblies Alpha and Gamma). The results from switching vessels show that the vessels are one of the important contributors to differences among assemblies.
- The Apparatus 2 distribution patterns may provide some insights into the fluid dynamics of dissolution testing. Deviations from an optimally functioning assembly system, such as paddle wobble or misalignment of shaft or vessel, may introduce additional energy to the system (18,19). As a consequence, increased dissolution values should be anticipated assuming other parameters (e.g., temperature, rotation speed, and paddle height) are properly controlled.
- Figure 3 shows overall distributions patterns for percent prednisone dissolved for both paddle and basket systems. Although the overall dissolution ranges were similar, the shapes of the distributions were different. The paddle system results were less symmetric, as seen from the tailing results in the higher dissolution range. The basket system results were more symmetric but showed a larger half-peak width. These results may provide insights into the inherent variability of basket and paddle systems and help improve future designs.

CONCLUSIONS

Dissolution experiments and statistical analyses demonstrated that the dissolution assembly and its associated variables contribute importantly to variability in dissolution results. This variability is particularly apparent for Apparatus 2 (compare data for Apparatus 1 and 2 in, e.g., Tables II and IV and III and V and the different manufacturers' assemblies therein). Vessel-switching experiments demonstrate that the contribution of vessels to variability should neither be overlooked nor underestimated. The present study was not designed to identify all the factors among different assemblies that contribute to variability, but irregularity of vessel dimensions (13) and vibration (data not shown), among others, clearly contribute. A challenge to a PVT compendial requirement is that the quality of the physical tablet is an important contributor to variability and thus to wide acceptance criteria for the PVT. The present article refutes the first part of the challenge; the second article in this two-part series confirms the finding of wide acceptance criteria. Data in this article indicate that dissolution assemblies and vessels are important contributors to the intra- and interlaboratory variability, which in turn leads to wide acceptance criteria. Studies in this report suggest that better understanding and control of assembly performance can reduce this variability. To provide guidance on this topic, USP has developed a "toolkit" that was posted on the USP Web site after consideration by the Council of Expert's Biopharmaceutics Expert Committee (www.usp.org/pdf/EN/dissolutionProcedureToolkit2007-10-04.pdf, accessed October 19, 2007). At times, the suggestion arises that a manufacturer may develop its own physical dosage form standard for a PVT. For procedure- rather than method-dependent tests, a "private" dosage form reference standard may be appropriate, subject to independent testing (8–10). Additional studies may be needed to ensure that a private physical dosage form standard is sensitive to variables that contribute to variability arising in the procedure. USP provides its RS tablets to conserve manufacturers' resources in conducting these studies and to allow a common standard for interlaboratory comparisons. USP supports mechanical calibration as a means of enhancing experimental results by means of OQ. However, mechanical calibration alone is not adequate to assess the performance of this procedure-based test, and the likelihood that an apparatus (or operator–apparatus system) may contribute to variability reinforces the importance of rigorous PVT in conjunction with mechanical calibration (20). In terms of ISO 5725-3, mechanical calibration, as it is commonly practiced, by itself cannot detect trueness and precision, which are the two components of accuracy (8). USP's reference standard tablets are not a substitute for mechanical calibration—instead, both USP RS Tablets and mechanical calibration are components of well-designed overall program of IQ, OQ, and PQ, with the latter now termed the PVT. The observations in this paper are important to practitioners: An assembly that runs "hot" can allow a poorly performing nonsolution orally administered dosage to pass dissolution acceptance criteria and thus appear to be of good quality when it is not.

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REFERENCES

1. USP, *USP 30–NF 25*, *Dissolution* <711>, United States Pharmacopeial Convention, Inc., Rockville, MD, 2001, pp. 277–282.
2. W. W. Hauck, T. Foster, E. Sheinin, T. L. Cecil, W. Brown, M. Marques, and R. L. Williams. Oral dosage form performance tests: new dissolution approaches. *Pharm. Res.* **22**:182–187 (2005).
3. FDA, Center for Drug Evaluation and Research. Guidance for industry: waiver of *in vivo* bioavailability and bioequivalence studies for immediate-release solid oral dosage forms based on a biopharmaceutics classification system. (2000). www.fda.gov/cder/guidance/3618fnl.pdf. Accessed October 19, 2007.
4. WHO. 2005. Multisource (generic) pharmaceutical products: guidelines on registration requirements to establish interchangeability.

Geneva, Switzerland: WHO. (2005). www.who.int/medicines/services/expertcommittees/pharmprep/QAS04_093Rev4_final.pdf. Accessed October 19, 2007.
5. M. Glasgow, S. Dressman, W. Brown, T. Foster, S. Schuber S, R. G. Manning, S. Z. Wahab, R. L. Williams, and W. W. Hauck W. W. The USP performance verification test, part II: collaborative study of USP's Lot P Prednisone Tablets. DOI 10.1007/s11095-007-9482-2 (2008).
6. NIST. Guidance on federal conformity assessment activities. *Fed. Regist.* **65**:48894–48902 (2000).
7. FDA. Guidance for industry: text on validation of analytical procedures. (1995). www.fda.gov/cder/guidance/ichq2a.pdf. Accessed October 19, 2007.
8. ISO. *ISO 5725 1-6: Accuracy (Trueness and Precision) of Measurement Methods and Results*. ISO, Geneva, Switzerland, 1994.
9. ISO. *ISO/IEC Guide 43-1: Proficiency Testing by Interlaboratory Comparisons*. ISO, Geneva, Switzerland, 1997.
10. J. Dressman, and J. Kramer. *Pharmaceutical Dissolution Testing*. Taylor & Francis, Boca Raton, FL, 2005.
11. R. Hanson, and V. Gray. *Handbook of Dissolution Testing*, 3rd ed. Dissolution Technologies, Hockessin, DE, 2004.
12. J. Eaton, G. Deng, W. W. Hauck, W. Brown, R. G. Manning, and S. Wahab. Perturbation study of dissolution apparatus variables—a design of experiment approach. *Dissolution Technol.* **14**:20–26 (2007).
13. M. R. Liddell, G. Deng, W. W. Hauck, W. Brown, S. Wahab, and R. G. Manning. Evaluation of glass dissolution vessel dimensions and irregularities. *Dissolution Technol*. **14**:28–33 (2007).
14. S. A. Qureshi, and I. J. McGilveray. Typical variability in drug dissolution testing: study with USP and FDA calibrator tablets and a marketed drug (glibenclamide) product. *Eur. J. Pharm. Sci*. **7**:249–258 (1999).
15. S. A. Qureshi and J. Shabnam. Cause of high variability in drug dissolution testing and its impact on setting tolerances. *Eur. J. Pharm. Sci.* **12**:271–276 (2001).
16. Z. Gao, T. W. Moore, A. P. Smith, W. H. Doub, and B. J. Westenberger. Studies of variability in dissolution testing with USP apparatus 2. *J. Pharm. Sci*. **96**:1794–1801 (2007).
17. J. L. Baxter, J. Kukura, and F. J. Muzzio. Hydrodynamics-induced variability in the USP Apparatus 2 dissolution test. *Int. J. Pharm.* **292**:17–29 (2005).
18. J. Kukura, J. L. Baxter, and F. J. Muzzio. Shear distribution and variability in the USP apparatus 2 under turbulent conditions. *Int. J. Pharm.* **279**:9–17 (2004).
19. USP. The USP performance test: mechanical calibration *vs.* a periodic performance verification test with reference standard tablets (calibrators) [and links therein]. www.usp.org/USPNF/notices/calibratorsPublicStatement.html. Accessed October 19, 2007.
20. R. Hanson. Comments on mechanical and chemical calibration by an instrument manufacturer. *Dissolution Technol*. **14**:7(2007).