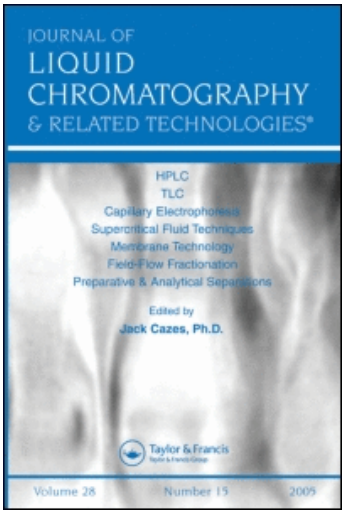

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Chromatographic Method Validation: A Review of Current Practices and Procedures. III. Ruggedness, Re-Validation and System Suitability

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CHROMATOGRAPHIC METHOD VALIDATION: A REVIEW OF CURRENT PRACTICES AND PROCEDURES. III. RUGGEDNESS, RE-VALIDATION AND SYSTEM SUITABILITY

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ABSTRACT

Validation of analytical methodologies is an important aspect of their development/utilization and is widely required in support of industrial product development and registration. In this manuscript, ruggedness as a validation parameter is considered in terms of its definition, appropriate evaluation procedures and acceptance criteria. Additionally, the re-validation of analytical methods is discussed, strategies for the effective development and utilization of system suitability tests are described and the term "stability indiating" is defined.

INTRODUCTION

Chromatographic methods are used for the quantitative and qualitative characterization of environmental and pharmaceutical samples. The object of the characterization is to generate a reliable, accurate and interpretable set of information describing the sample. To ensure that an analytical procedure fulfills this objective, it undergoes an evaluation loosely termed validation. In previous parts of this series,[1,2] primary validation parameters (e.g., accuracy, precision, specificity, linearity and sensitivity) were identified and discussed in

terms of their definition, scope, evaluation procedures and acceptance criteria. In this manuscript, the series is concluded with a consideration of ruggedness, re-validation and system suitability.

RUGGEDNESS

Definition

It is generally expected that an analytical method will perform in an acceptable manner each time it is used. A method which is difficult to implement is highly undesirable from the practical perspective of efficient resource utilization and is generally suspect in terms of the quality of the data generated. The ability to routinely implement an assay reflects its inherent ruggedness. While a consideration of method ruggedness is a necessary part of any method's validation, it's a critical issue for compendial methods because of their widespread use in many different laboratories.[3]

Ruggedness establishes a method's ability to perform effectively in the face of variations which can reasonably be expected to occur whenever the method is implemented. More specifically, ruggedness is the reproducibility of test results obtained by the analysis of samples under a variety of normal test conditions such as different laboratories, analysts, instruments, reagent lots, elapsed assay times, temperatures, etcetera.[3-7] Thus, ruggedness addresses ***unintentional*** variation in the method introduced by its application, at different times by different people at different locations using different instrumentation and materials. Ruggedness measures the extent to which a method is sensitive to small changes in procedures and circumstances.[8] A rugged method will be able to withstand minor operating or performance changes[9] and has built in buffers against typical procedural abuses, such as, differences in care, technique, equipment and conditions.[10]

Procedures

Clearly, ruggedness is assessed by implementing the analytical method under different operational conditions. The ruggedness test should be performed at several values of each operational parameter which affects method performance.[3] For chromatographic assays, these parameters might include mobile phase composition and flow rate, column vendor, column condition, detection wavelength, sample and standard preparation procedures and operating temperature. For a reverse phase HPLC method using an ion pairing reagent, for example, the following conditions can be evaluated for their effect on capacity factor(s) or resolution of a critical pair of analytes:[11]

* Mobile phase composition (pH, buffer concentration, ion pairing reagent concentration, percent organic phase),
* column temperature,
* injection volume,
* gradient dwell time, and
* column lots or column manufacturers.

The ruggedness test should be performed by analyzing aliquots from homogeneous sample lots using operational and environmental conditions that differ but are still within the method's specified operating range.[4,9,12] The ruggedness evaluation should be performed on a sample which has been previously characterized (especially in terms of its stability) by an experienced analyst[13] and should include any precision-related tests and requirements contained in the procedure's protocol or specification.[14]

In order to assess the magnitude of operator-related ruggedness, it has been suggested that four analysts perform one assay per day for three days.[5] The utilization of statistically designed experiments (e.g., Plackett-Burman, nested ANOVA, factorial plans) to establish the ruggedness of an assay is strongly recommended.[15,18]

A common source of performance variation in chromatographic methods is the separation column. Performance variation is introduced into the method by the age and care of the column, inherent column non-reproducibility resulting from production variations within a manufacturer's process (batch of stationary phase, packing procedure) and variation in selectivity and performance between columns of similar generic type supplied from different vendors in different configurations.

In order to assess column ruggedness, it is recommended that the specificity of at least three columns, each one from a different batche produced by the recommended column manufacturer and at least one column from a different manufacturer be checked.[3,10,14]

Acceptance Criteria

The quantitative measure of a method's ruggedness is the precision behavior it exhibits over the course of the various operational scenarios examined during the validation exercise. To determine the method's ruggedness, method reproducibility obtained throughout the changes tested, should be compared to the precision of the assay under normal conditions;[4] the reproducibility thus obtained should not be significantly different from the method's intermediate precision obtained under normal operating conditions. Generally, a rugged method's reproducibility is 2 to 3 times greater than the

method's repeatability [inherent method precision under "normal" controlled operating conditions].[17] For ruggedness determinations utilizing a factorial design, a ratio of the variances associated with ruggedness and reproducibility of greater than 1.5 is strongly indicative of one or more factors that adversely affect the method's performance.[18] For an evaluation of column to column ruggedness, it is required, in addition to a precision comparison, that the method pass the specificity test criteria on all columns tested.[14]

Related Considerations

Application of chromatographic procedures requires the use of liquid samples, standard and related analytical reagents. In most routine applications, solutions are not used immediately after preparation but may be stored under specified conditions prior to use. Verifying solution stability is an important aspect of method validation; specifically, a valid method is one for which all related and analytical solutions are stable over the period typically required for their utilization/analysis. To address stability, the analytical solutions should be prepared, assayed, allowed to stand (in accordance with the method's protocol or specification) for a length of time equal to the anticipated maximum analysis time and then re-assayed.[14,19] It has been suggested that, for analytical scenarios involving overnight runs, four sample solutions over the working concentration range should be analyzed repetitively over the course of at least sixteen hours.[20] In such evaluations, the analytical solution is stable if all concentration values obtained before and after storage agree to within three times the system precision.[14,19] Additionally, no new peaks should appear in, nor should existing peaks be lost, from the chromatograms of the first and last sample injection.[14]

While ruggedness is related to unintentional variation in a method due to its use in varying analytical situations, method robustness is a measure of a procedure's capacity to remain unaffected by small but ***deliberate*** variations in method parameters and thus is a measure of the procedure's reliability during normal usage.[4,13,21,22] Although time consuming to perform, thorough robustness studies will help avoid unexpected results in subsequent applications of the method. Thus, the robustness evaluation should serve as a prelude to assay transfer.[13] While data for robustness is not usually submitted in regulatory product applications, a robustness evaluation is recommended.[22]

It has been suggested that in order to determine robustness, a method's critical operational variables should be identified by breaking the testing process up into unit operations and then assessing the potential variability of each such operation.[13] Unit operations might include:

1. Analytical solution preparation: amount of material used, volumes of solvent used, dissolution times and conditions, solvent used.
2. Variation in the tested product (inhomogeneity, aging).
3. Instrumental analysis: detection wavelength, mobile phase composition and flow rate, column use history.

The intent of the robustness evaluation is to quantify the amount of method variation introduced by changing an operational variable by a known amount. Clearly, a robust method is one which is operationally immune to commonly encountered but relatively minor variations in its critical operating parameters.

METHOD RE-VALIDATION

If an analytical method exhibits any significant longevity, it invariably undergoes some change in procedure or implementation. It is possible that method performance, and thus the validity of the data generated by the method, could be adversely impacted by such changes. Re-validation, which may be required in such situations, is the reassessment of a validated analytical method in response to a change in some aspect of the method.

Issues associated with re-validation are two-fold: 1) how big of a change triggers a re-validation and 2) how extensive should the re-validation be? Considering the former, utilization of the most conservative approach minimizes the likelihood that even the most apparently innocuous change could produce a significant change in performance. Specifically, the investigator must avoid assumptions regarding the definition of a "major" change[13] and assume that any modification of the analytical method would require re-validation.[23] In essence, validation should be ongoing in the form of re-validation with method changes.[14] For chromatographic methods, significant changes could include:

* Changes in the product for which the method was validated,
* Use of the assay for a product different from that for which it was validated,
* Instrument changes,
* Reagent changes (type or vendor),
* Procedural changes,
* Personnel changes, and
* Technological changes (e.g., developments in column and/or instrumentation technology).

With regard to the extent of the re-validation exercise, it is clear that the greater the magnitude of the method change, the greater the need for and scope of the re-validation.[24] The decision regarding which parameters require re-

Table 1

Method Changes and Re-Validation Tests Required

Method Characteristics Changed	Performance Parameters to Re-validate
Instrument Changes	Linearity (working range), LOD, LOQ, system precision
Product Changes	Selectivity, accuracy, precision
Sample Preparation Procedure (same solvent, same concentration range)	Accuracy, recovery, precision, ruggedness
Sample Preparation Procedure (different solvent, different concentration range)	Complete-reassesment of all previously used validation parameters
Analyst Changes	Qualification testing (perform re-tests, side by side collaborative studies)
Chromatographic change (e.g., column, mobile phase)	Selectivity, linearity, LOD/LOQ, system precision

LOD = Limit of Detection, LOQ = Limit of Quantitation.
From reference 13.

validation should be based on a logical consideration of the specific validation parameters which are likely to be affected by the change.[16] Minimally, however, re-validation of chromatographic methods might include an assessment of accuracy and the absence of interference[4] or the running of a standard curve with new quality control samples to show that the response relationships and general characteristics of the "new" method are similar to the previous validation results.[23] For bio-analytical methods, precision, accuracy and limit of quantitation are considered to be the minimum re-validation tests.[24]

More specific recommendations for which method parameters should be re-validated in response to specific types of procedural changes are summarized in Tables 1 and 2.

SYSTEM SUITABILITY

Role

To obtain a good and acceptable analytical result, two requirements must be met; (1) the method has to be adequate and (2) the execution has to be adequate.[25] In its broadest sense, method validation addresses the former issue

Table 2

Additional Guidelines for Re-validation

Method Characteristics Changed	Performance Parameters to Re-Validate
Extraction solvent, buffer, back extraction matrix or injection solvent	Linearity, recovery, LOQ, intra-batch preciison and accuracy in process solution stability. Additionally, if injection solvent is changed, processed sample stability should be checked but recovery or in-process stability checks are not necessary.
Chromatographic conditions [column, mobile phase composition, detector type or monitoring condition (e.g. wavelength) change].	Linearity, selectivity, intra-batch precision and accuracy (recovery not necessary).
Extending the upper end or reducing the lower end of the calibration curve range.	Linearity, LOQ (if reduced), intra-batch precision and accuracy at revised upper or lower levels.
Internal standard	Selectivity, intra-batch precision and accuracy, recovery.

From reference 16.

but leaves the latter essentially unresolved. While the most rigorous verification of adequate execution would be re-validation at each use, such an approach suffers from serious practical shortcomings involving resource constraints. System suitability tests (SST) have been adopted by chromatographers to describe the process by which the execution of an analytical process is evaluated. SST typically represents a sub-set of the method validation procedures and obviates the need for a more rigorous re-validation[10] by serving as a surrogate for the more involved validation process.

SST tests, introduced by FDA chemists in the early 1970's,[26] were originally intended to prevent the known variability of chromatographic components from adversely affecting official methods. Even today, the USP monograph on Chromatography[27] indicates that the SST "are used to verify that the resolution and repeatability of the chromatographic system are adequate..." and that resolution, tailing factor and precision are the primary SST parameters. As SST procedures became a more common part of the method development/utilization process, their traditional role has been enumerated by

numerous authors. Such descriptions suggest that the role of SST testing is to:

* Confirm the method's continuing suitability for use.[28,29]
* Ensure that the method is performing properly, satisfactorily or as intended.[8,10,24,30.]
* Establish that the system meets criteria of historic norms, accepted operational standards, or performance requirements.[3,5,10]
* Provide the analyst with an early warning that an analytical process is likely to be out of control.[31]

Historically then, the SST has been implemented as a time of use procedure, whose sole purpose was to document acceptable system operation by comparing observed performance versus previously established guidelines. While it served an important role in such applications, the impact of the SST was both passive and reactive. Although the test identified a sub-optimal system, it provided no clue as to how to improve performance. Additionally, the performance of the SST was most commonly viewed as a one time event, with little or no effort made to interpret trends in SST data as a means of proactively recognizing decaying system performance. More recent manuscripts, have suggested more active roles for SST evaluations including:

* A correctly used SST should verify that the analysis has been performed consistently over time.[29]
* A SST should indicate which component in or step of an analytical procedure should be replaced or modified.[29]
* The data should be useful as a means of directing a non-compliance towards a compliance.[30]
* The SST must indicate what the analyst should do in the event of a test failure.[11]

Thus, in its evolving role, the SST serves not only as an indicator of adequate performance but also provides diagnostic information related to the source of the problem and prescriptive information related to the correction of the problem. Through the use of control charting, the SST database provides a picture of the system's historical capabilities and allows for the development of statistically based performance criteria. In this expanded role, the SST is a vital tool for the routine quality control of chromatographic assays.[30]

Previously, an effective validation plan was defined as one for which the user knows which performance parameters to assess (scope), how parameter evaluation is performed (procedure) and the appropriate acceptance criteria are.[2] A similar definition is appropriate for an effective SST. The following discussion considers these aspects of an SST evaluation in greater detail.

Table 3

ParametersWhich Should be Contained in a System Suitability Evaluation (Based on a Survey of Published Methods on the LC Analysis of Drug Substances and Dosage Forms)

Evaluation Parameter	Frequency of Citation (#)
Resolution	23
Precision of Standards	17
Standard Linearity	7
Tailing Factor	6
Theoretical Plates	5
Retention Time	2
Precision of Impurities	1
Capacity Factor	1
Peak Asymmetry	1

(#) Number of citations which mentioned this specific SST parameter. Of the 84 total references city in this manuscript, 28 provided system suitability test guidelines.
From reference 34.

Scope

Critical issues associated with performing a system suitability test include the identification of which performance characteristics need to be monitored and how frequently the test must be performed. The overriding issue here is efficiency; it is desired that the test provide the maximum measure of system performance with a minimum expenditure of time and effort.[29] The design of the system suitability test should balance the time to perform the test versus the risk of chromatographic failure during the run (and the resulting non-availablilty of the analytical data).[29] Historically, this balance has been heavily weighted against rigorous SST testing, which is often viewed as a formality to be overcome. However, evaluation of a system with a properly written SST may actually save more analytical time than is taken to perform the test by eliminating retesting.[14]

Ultimately, the amount of testing performed will depend on the purpose and nature of the test method.[22] While an SST should be considered for each parameter which was checked during method validation,[28] the implemented test procedure should incorporate only those key parameters that are crucial to the success of the method, as defined by its specific analytical objectives.[29] For example, while an SST for sensitivity might be quite applicable in an impurity

assay (where the ability to detect the impurity is important), such a test might have little application in situations wherein the intent of the assay is to accurately quantitate a formulation component present in the sample in large quantities.

Since this manuscript is limited to a consideration of chromatographic methods, appropriate SST parameters must reflect problems associated with the implementation specifically of chromatographic procedures. Problems a thorough system suitability test should surface include;[32]

* flow irregularity,
* injection irreproducibility,
* system plumbing problems,
* detector mis-alinement/malfunction,
* column malfunction, and
* mis-preparation of analytical solutions (mobile phase, sample diluent, derivatization reagent, standards, samples).

Numerous authors have outlined parameters which should be examined in a rigorous SST, e.g. references 2, 7-12, 22, 27, 28, 32 and 33. Two parameters mentioned in every manuscript examined for this review were resolution and repeatability (e.g., system precision). The universal use of these parameters is understandable since they touch on two important properties of the chromatographic assay, specificity and precision. A measure of peak shape (e.g., tailing factor, peak asymmetry) was also frequently cited as a necessary component of a rigorous suitability assessment. Assessment parameters which were less frequently noted included capacity factor (ratio), a measure of sensitivity (LOD or LOQ), linearity, column efficiency (plate count) and the analysis of controls. The use of multiple injections of a standard, made throughout the run to assess response stability, was also suggested.[11]

These observations are reinforced by two recently published surveys. In 1990, T. D. Wilson published the results of a survey of literature methods on the LC analysis of drug substances and dosage forms.[34] Of 84 references cited, 28 made specific mention of system suitability parameters. As shown in Table 3, the frequency with which specific SST parameters were mentioned mimics the general trend noted previously. Additionally, in 1994, G. S. Clarke surveyed most major pharmaceutical companies with research laboratories in the UK with respect to their method validation and system suitability procedures.[35] Data summarizing the frequency with which specific SST parameters were used are contained in Table 4. Parameters which were used by a majority of the companies included precision, selectivity (resolution) and chromatographic performance (e.g., resolution, efficiency) while accuracy, linearity, selectivity, ruggedness, solution stability and sensitivity (signal to noise ratio) were used less commonly.

Table 4

Parameters Which Sould be Contained in a System Suitability Evaluation (Based on a Survey of Practices Used by Major Research-Based Pharmaceutical Companies With Laboratories in the UK)

Parameter	Company Responses (*)	
	In Common Use	Not in Common Use
Selectivity (resolution)	20	0
	14	4
Chromatographic parameters (capacity factor, plate count, tailing factor)	11	9
Limit of Detection (LOD)	9	11
Accuracy	4	16
Linearity	3	15
Signal to Noise ration	1	19
Selectivity (peak homogeneity)	1	19
Solution Stability	1	9

(*) The total number of companies survey was 20.
From reference 35.

Wahlich and Carr[28] advocate the use of SST parameters which reflect each parameter which was considered as part of the method's validation process. These parameters, linked to typical validation parameters and contrasted to the more conventionally recommended SST parameters, are summarized in Table 5.

In reviewing the literature related to SST parameters, this author was struck by several points. Firstly, it is somewhat unusual, in this author's opinion, that some direct measure of accuracy was so infrequently cited as a necessary SST parameter. This is striking since accuracy is one of the most universally applied method validation parameters, Except in bioanalytical procedures, wherein analyzing QC samples is the most popular method for monitoring assay performance,[24] the direct assessment of method accuracy is rarely mentioned as a necessary SST parameter.

Secondly, there exists some discordance in terms of which of the chromatographic performance parameters are most useful. For example, several authors suggest that "it is questionable whether in absolute terms either tailing or column efficiency add anything to the suitability for use of a method".[28] They suggest this is true since little attempt is usually made to determine whether failure to comply with criteria for these parameters means that the method is any

Table 5

A Suggested Link Between Method Validation Parameters and System Suitability Tests

Method Validation Parameter	Traditional SST	Recommended SST
Ruggedness/robustness	None	Check on critical method parameters
Accuracy	None	Control sample, re-extraction or mass balance
Precision	RSD of replicate injections	RSD of replicate injections: RSD of replicate sample preparations
Selectivity	Resolution check	Resolution check (using impure standards or samples of the impurities)
Stability of the measurement system	None	Comparison of standards at the start and end of run
Linearity	None	Use of standard at different concentrations
Signal to Noise (LOD/ LOQ)	None	Calculation of H/s_B rato (*)
General Acceptability	None	Chromatogram compared to reference chromatogram
None	Tailing factor/peak asymmetry	None
None		None
None	Column efficiency/plate count	None

(*) H = peak height of a specified standard; s_B = standard deviation of the baseline.
From reference 28.

less valid. Additionally, neither peak tailing or efficiency has any direct link to a primary validation parameter. Considering tailing, it has been observed that as peak asymmetry increases, accuracy[22] and precision[27,29] suffer. Thus peak tailing acts a surrogate SST for accuracy and precision. Since precision is a routinely utilized SST parameter, and the SST assessment of accuracy is becoming more common, the usefullness of the peak shape SST is questionable.

A similar situation exists for efficiency. Efficiency is utilized as an SST to indirectly assess method specificity; that is, efficiency seeks to ensure that the column possess the ability to separate the analytical peak(s) of interest from all

possible interfering responses. While most investigators acknowledge that an assurance of specificity is an important SST component, it is frequently noted that efficiency (theoretical plates) is inferior to resolution[14] as a measure of specificity. Resolution may be considered to be a more powerful tool for testing chromatographic performance since it addresses efficiency (N), selectivity (alpha) and capacity (k') via the expression:[30]

$$R = 1/4 \times (\alpha - 1) \times N^{0.5} \times [k'/(1+k')]$$

Procedures

The first procedural aspect to be considered is the frequency with which an SST should be performed. Two timeframes are pertinent; within run repetition of SST testing and between run testing. Considering within run replication of SST testing, the current USP emphasis is to perform all system suitability injections prior to the analysis of actual samples.[27] However, such an approach can lead to erroneous results since it establishes only that the system performed within expectations at the beginning of the run and does not demonstrate that such performance was maintained throughout the run.[36] In general, intervals between tests should be shorter than the observed time in which the system drifts outside of acceptable levels.[37] In most cases, this means that the SST is performed at the beginning and end of the run. Such testing can take the form of a precision evaluation to ensure that the nature of the analytical response has not changed over time, or may involve nothing more complicated than a visual comparison of chromatograms generated at the beginning and end of the run, from the same sample.[28] More rigorously, it has been proposed that the appropriate frequency for the SST test, is to run one QC control per every ten samples or, for short runs, two QC controls minimum,[24] while tests for bias and/or response stability should include the repetitive analysis of a single solution throughout the run.[10,11,33] Additionally, an SST evaluation is performed each time an instrument malfunction has been identified during the course of a run.[27]

The decision of how frequently an SST is performed between analytical runs should be determined by experience and based on need, type of test and equipment and previous performance of the equipment.[37] Minimally, the SST should be performed in full each time the system is assembled for the assay. However, if the system is in continuous use for the same analysis, then it may be sufficient to perform an abbreviated SST check each day.[14]

Considering other procedural aspects of system suitability testing, several authors provide somewhat more quantitative guidelines on how the SST is to be performed. When utilizing QC samples to assess accuracy, it is suggested that duplicate injections be made of QC standards at three concentrations [below,

Table 6

Recommended System Suitability Test Acceptance Criteria

Parameter	Assay Type	Acceptance Criterion	
		Hsu and Chien(*)	CDER (#)
Capacity Factor			
	General	2 to 8	> 2
	Trace	1 - 3	N/A
	Stability Indicating	> 4	N/A
Selectivity	General	1.05 to 2.0	N/A
Resolution	General	>2.0	>2.0
	Quantitative Analysis	>1.5	N/A
	Biologicals	>1.2	N/A
Plate Count (N)	General	(a)	>2000
Precision	General	% RSD ≤ 1.5%	% RSD ≤ 1.0% (b)
	Biologicals	% RSD ≤ 5%	N/A
	Trace	% RSD, 5 to 15%	N/A
Tailing Factor	General	1.5 to 2.0	≤ 2.0

Notes: (a) = no criterion given, however, the analyst should look for decreases in this number as a sign of degrading system performance.
(b) = for 5 replicate injections.
N/A = no specific guidelines given for this situation.
(*) reference 40; (#) reference 22.

within (midpoint) and above] around the expected range.[23,28] To assess system precision, samples at both ends of the calibration curve should be injected at least five times, with six injections being required if the acceptance criterion is a %RSD greater than 2.0%.[14,27,29]

Acceptance Criteria

The acceptance criteria established for the SST evaluation must balance the need to insure adequate performance with the practical reality of performing chemical analyses. Thus, the criteria must be sufficiently tight that data quality is assured but not so restrictive that perfectly acceptable systems cannot readily pass all criteria. It is crucial that the acceptance criteria are designed to reflect method variances which affect the quality of the data generated.[11] To be useful to the analyst, the criteria should reflect minimum, as opposed to typical, performance.[30]

In general, setting the acceptance criteria involves an assessment of the chromatographic conditions to which the method is most sensitive and then using existing performance data (obtained perhaps during method development-validation), to help establish the criteria.[30] Typically, data obtained from ruggedness testing, can help define system suitability criteria.[39] It has been proposed[30] that the following three step process be used to develop meaningful system suitability criteria:

(1) Determine the sensitivity of the method to changes in chromatographic conditions.
(2) Identify suitable performance parameters that can monitor system functionality and determine their minimum or maximum acceptable value.
(3) Validate these criteria for each formulation, product or sample that is assayed by the method.

Specific recommendations for SST acceptance criteria include:

* For the repetitive injection of response stability samples, the %RSD of the repetitive injections should be ≤ 120% of the system precision.[33]
* Duplicate injections of a standard injected periodically throughout an assay should agree to within 0.5% of their average.[10]
* The %RSD of a series of standard injections interspersed throughout the run should have a %RSD ≤ 1%. Failure to comply with this criterion may be overcome by using standard bracketing to divide the run into "compliant" portions [i.e., portions which meet the criterion].[28]
* In using QC samples, the results are acceptable if they are within 10% of the known value.[38]
* For QC protocols involving the duplicate analysis of samples prepared at three concentrations (e.g., biological samples), 4 of the 6 QC values must be within 20% of expected, while those outside this range cannot be of the same concentration.[23]

More detailed acceptance criteria are provided for the common SST parameters by Hsu and Chien[40] and the Center for Drug Evaluation and Research [FDA][22] and are summarized in Table 6.

SST Failures

If a system fails an SST and the procedure specification or protocol describes the analytical procedure in great detail, the analyst is faced with the dilemma of what to do next. Fortunately, it is well recognized in the pharmacopeial literature, that the specification of definitive parameters in a

monograph (procedure) does not preclude the use of other suitable operating conditions and, thus, that adjustments of operating conditions to meet the system suitability requirements may be necessary and appropriate.[3,27] However, once the conditions have been adjusted, it is not adequate to test the new system only for that SST which was previously failed. Utilization of the adjusted system is predicated on the assumption that its is capable of meeting all SST requirements.

STABILITY INDICATING ASSAYS

Assays suitable for the determination of the stability and shelf life of pharmaceutical formulations and products share expected performance criteria which are somewhat more rigorous than those necessary for assays used in other applications. A stability indicating assay must be able to determine small changes in the concentration of the analyte of interest and exhibit no interference from other sample components (e.g., degradation products).[41] Special demands placed on stability indicating assays include:[33]

* The method should be able to accurately follow the decrease in active content during the period of the stability investigation,
* The desired resolutions between peaks are set higher (than in most other applications) in order to identify and quantitate degradation products,
* Reproducibility (day to day precision) must be better than 1% RSD in order that small decreases in active ingredients can be measured, and
* the peaks of the primary and secondary degradations products must be separated from one another, the active ingredient and other formulation impurities.

Stability indicating assays, typically quantitate analytes which include one or two major componentes and several impurities (<0.5%). These assays have resolution (between multiple peaks), accuracy, reproducibility and sensitivity as primary validation and system suitability parameters.[7] Thus, one can expect the acceptance criteria for these assays to be more stringent than for those assays used in other pharmaceutical situations.

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