



ANALYTICAL METHOD VALIDATION REQUIREMENTS: A COMPARATIVE REVIEW OF ICH, USP, CHINESE PHARMACOPEIA, AND ANVISA (BRAZIL) REGULATORY GUIDELINES

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ABSTRACT

Analytical method validation is an essential component of pharmaceutical quality assurance, ensuring that analytical procedures are suitable for their intended purposes and consistently generate reliable results. Regulatory authorities and pharmacopoeial organizations have established validation requirements to support pharmaceutical development, manufacturing, quality control, and regulatory submissions. However, differences in validation terminology, experimental design, acceptance criteria, and documentation practices may complicate global regulatory compliance. This review critically compares the analytical method validation requirements described in the International Council for Harmonization (ICH) Q2(R2) guideline, United States Pharmacopeia (USP) General Chapter <1225>, Chinese Pharmacopeia (ChP), and Brazilian Health Regulatory Agency (ANVISA) Resolution RDC No. 166/2017. This review focuses on specificity/selectivity, accuracy, precision, linearity, range, detection limit, quantitation limit, robustness, and system suitability. The review also examines how analytical quality by design, risk-based validation, lifecycle management, and statistical evaluation are integrated into contemporary analytical practice. Although the four frameworks share the fundamental objective of establishing analytical procedure fitness for purpose, they differ in regulatory status, procedural prescription, and how they interpret validation evidence. ICH Q2(R2) provides an internationally harmonized scientific framework, USP <1225> establishes compendial validation principles, the Chinese Pharmacopeia addresses validation within China's pharmacopoeial system, and ANVISA RDC 166/2017 specifies detailed requirements for analytical methods used in Brazilian pharmaceutical regulatory activities. A scientifically justified validation strategy that considers these differences can reduce redundant experimentation, facilitate international submissions, and improve analytical reliability. Future harmonization should emphasize performance-based acceptance criteria, analytical procedure lifecycle management, and transparent documentation.

Keywords: Analytical method validation; ICH Q2(R2); USP <1225>; Chinese Pharmacopeia; ANVISA; Regulatory harmonization; Analytical quality by design.

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INTRODUCTION

Analytical method validation is a systematic process that demonstrates an analytical procedure is suitable for its intended application. It plays a critical role in pharmaceutical quality control by establishing confidence in the identification, quantification, purity assessment, and stability evaluation of pharmaceutical

substances and finished dosage forms. Reliable analytical procedures are essential for ensuring product quality, patient safety, manufacturing consistency, and compliance with regulatory requirements [1–3]. The increasing globalization of pharmaceutical research, manufacturing, and distribution has created a need for analytical procedures that satisfy multiple regulatory jurisdictions. Pharmaceutical companies frequently submit analytical validation data to authorities in different countries, making harmonization of validation expectations an important consideration. Although international guidelines have established common scientific principles, regional differences in validation requirements may influence experimental design, statistical evaluation, documentation, and regulatory acceptance [4, 5].

The International Council for Harmonization introduced ICH Q2(R2) to modernize analytical procedure validation and align it with current analytical technologies and scientific approaches. The guideline addresses validation characteristics according to the intended purpose of an analytical procedure and is complemented by ICH Q14, which describes analytical procedure development and associated lifecycle concepts.

The United States Pharmacopeia provides validation principles through General Chapter <1225>, Validation of Compendial Procedures, while other chapters address verification of compendial procedures, chromatography, and analytical procedure lifecycle considerations [6]. In China, the Chinese Pharmacopeia establishes pharmacopoeial requirements and general guidance for analytical procedures used in pharmaceutical quality assessment [7,8]. Brazil's Agência Nacional de Vigilância Sanitária (ANVISA) introduced Resolution RDC No. 166/2017, which specifies criteria for analytical method validation and supports regulatory evaluation of pharmaceutical analytical procedures [9].

Despite their common scientific foundation, these frameworks differ in regulatory scope, technical detail, and approaches to validation planning. Understanding these similarities and differences is particularly important for laboratories developing analytical procedures intended for international regulatory submissions. The present review aims to critically compare analytical method validation requirements under ICH Q2(R2), USP <1225>, the Chinese Pharmacopoeia, and ANVISA RDC No. 166/2017. The discussion emphasizes validation characteristics, experimental considerations, statistical assessment, regulatory differences, and opportunities for international harmonization.

REGULATORY FRAMEWORKS GOVERNING ANALYTICAL METHOD VALIDATION

International Council for Harmonisation (ICH)

ICH Q2(R2), finalized in November 2023, provides a harmonized framework for validating analytical procedures used in pharmaceutical quality assessment. It describes validation characteristics, including specificity/selectivity, range, accuracy, precision, detection limit, quantitation limit, and robustness-related considerations. The selection of characteristics depends on the analytical procedure's intended purpose and the measurement being performed [10,11]. The revised guideline recognizes that validation evidence can be generated through scientifically justified experimental approaches. It accommodates modern analytical technologies and encourages appropriate use of prior knowledge, statistical evaluation, and development information. ICH Q14 complements this framework by describing systematic analytical procedure development, analytical target profiles, risk assessment, and analytical procedure control strategies [12,13].

An important feature of ICH Q2(R2) is its emphasis on demonstrating analytical performance rather than applying identical experimental designs to every procedure. This flexibility allows laboratories to establish scientifically appropriate validation strategies while maintaining regulatory confidence.

United States Pharmacopeia (USP)

USP General Chapter <1225> provides a framework for validating compendial analytical procedures. It describes the principal validation characteristics and their relevance to different analytical applications, including identification, impurity testing, and assay procedures [14]. The USP framework distinguishes between validating analytical procedures and verifying established compendial procedures. General Chapter <1226> addresses verification, which demonstrates that a laboratory can perform an existing compendial procedure under its actual conditions of use. USP <621> provides chromatographic requirements and system suitability considerations relevant to chromatographic analysis [15]. Consequently, USP validation practices must be interpreted according to whether a laboratory is developing a new procedure, applying an official compendial procedure, or modifying an established procedure.

Chinese Pharmacopoeia (ChP)

The Chinese Pharmacopeia provides official pharmaceutical quality standards and general technical requirements for medicines marketed in China. Its analytical framework includes requirements and guidance concerning analytical procedures, method validation, pharmaceutical specifications, and quality control testing [16]. Validation generally addresses characteristics such as specificity, accuracy, precision, linearity, range, detection limit, quantitation limit, and robustness, where applicable to the intended analytical purpose. However, laboratories must determine the exact experimental requirements from the applicable edition, general chapter, and individual monograph. For international submissions, laboratories should verify the requirements of the legally applicable Chinese Pharmacopeia edition and any supplementary guidance issued by the National Medical Products Administration (NMPA).

Brazilian Health Regulatory Agency (ANVISA)

ANVISA Resolution RDC No. 166/2017 establishes requirements for analytical method validation in Brazil. It addresses validation characteristics including selectivity, linearity, analytical range, precision, accuracy, detection limit, quantitation limit, and robustness, as appropriate to the analytical application [17]. Compared with the general scientific flexibility of ICH Q2(R2), RDC 166/2017 contains more explicit procedural and statistical expectations for several validation characteristics. These include aspects of calibration evaluation, replicate measurements, and documentation of method performance. The regulation also distinguishes complete validation from partial validation under applicable circumstances. Laboratories preparing submissions for Brazil must therefore

evaluate the specific provisions of RDC 166/2017 rather than assuming that compliance with ICH Q2(R2) automatically demonstrates full compliance with Brazilian requirements.

Table 01: Comparative overview of the four analytical method validation frameworks

Regulatory framework	Principal document	Main application	Distinctive features
ICH	Q2(R2), supported by Q14	Validation of pharmaceutical analytical procedures	Harmonized, science-based and flexible approach; integration with analytical development
USP	General Chapter (1225)	Validation of compendial analytical procedures	Validation characteristics categorized by analytical application; separate verification guidance
Chinese Pharmacopoeia	Applicable ChP edition and general chapters	Pharmaceutical testing and quality control in China	Pharmacopoeial validation requirements linked to official standards and monographs
ANVISA	RDC No. 166/2017	Validation of analytical methods within Brazilian regulatory scope	Detailed experimental and statistical provisions; complete and partial validation concepts

Comparative Evaluation of Analytical Method Validation Characteristics Specificity and Selectivity

Specificity and selectivity describe an analytical procedure's ability to assess the analyte of interest in the presence of potentially interfering components. These may include excipients, impurities, degradation products, metabolites, and matrix constituents.

ICH Q2(R2) recognizes different approaches for demonstrating specificity or selectivity, including the absence of interference, comparison with an orthogonal analytical procedure, and scientifically

justified technology-inherent discrimination. USP (1225) similarly emphasizes the reliable identification or measurement of an analyte in complex pharmaceutical samples.

The Chinese Pharmacopoeia considers the intended analytical application when establishing specificity requirements. For chromatographic procedures, relevant evidence may include separation of analyte peaks from interfering substances and degradation products

ANVISA RDC 166/2017 addresses selectivity by evaluating potential matrix interference and other relevant components. For stability-indicating procedures, demonstration of analyte discrimination from degradation products is particularly important.

Across all four frameworks, specificity or selectivity is fundamental to analytical reliability. However, the required experimental evidence depends on the procedure, sample matrix, analytical technology, and regulatory context [18,19].

Linearity and Calibration Model

Linearity refers to the relationship between analyte concentration and analytical response over a specified interval. Although linear calibration is common in pharmaceutical analysis, not every analytical technology produces an inherently linear response.

ICH Q2(R2) emphasizes establishing a suitable calibration model for the intended reportable range. Linear regression may be appropriate, but other mathematical relationships can be justified when analytical data support them.

USP (1225) traditionally describes evaluation of linearity using appropriate concentration levels and regression analysis. The Chinese Pharmacopoeia also incorporates calibration assessment within analytical procedure validation, with its 2025 revision reflecting updated international validation principles.

ANVISA RDC 166/2017 is more prescriptive. Article 25 specifies at least five different reference-standard concentrations, with solutions prepared at least in triplicate. Article 27 requires evaluation of regression, residuals, correlation, slope significance, and homoscedasticity. It also specifies a correlation coefficient greater than 0.990.

These requirements demonstrate why a high correlation coefficient alone should not be regarded as sufficient evidence of an appropriate calibration model. Residual distribution, variance behavior, calibration uncertainty, and performance at range limits are also relevant [20].

Accuracy

Accuracy expresses how close analytical results are to an accepted reference value. It is commonly investigated through recovery experiments, reference material analysis, or comparison with an established procedure.

ICH Q2(R2) permits scientifically justified approaches to accuracy assessment, including recovery studies, reference materials, and comparison with orthogonal

procedures. USP (1225) describes accuracy assessment according to the analytical test category. The Chinese Pharmacopeia incorporates accuracy into its validation framework, while the applicable acceptance criteria depend on the analyte, concentration, matrix, and analytical purpose. ANVISA RDC 166/2017 provides more detailed expectations for accuracy experiments, including assessment across appropriate concentration levels and replicate determinations [9]. A common limitation in analytical publications is applying a universal recovery interval without considering the intended analytical purpose. Recovery criteria suitable for major-component assay methods may not apply to trace-level impurity analysis [21].

Precision

Precision describes the closeness of agreement among independently obtained analytical results under specified conditions. It is generally evaluated through repeatability, intermediate precision, and, where appropriate, reproducibility. Repeatability evaluates variation under closely controlled operating conditions. Intermediate precision considers relevant variations within a laboratory, such as different days, analysts, instruments, or analytical runs. Reproducibility concerns variation between laboratories and is particularly relevant to collaborative studies and method standardization.

All four frameworks recognize precision as a fundamental performance characteristic. Differences primarily concern experimental prescription, selection of sources of variation, and the extent of statistical evaluation.

Precision is commonly expressed using standard deviation or relative standard deviation:

$$\%RSD = \frac{SD}{Mean} \times 100$$

However, the acceptance limit should be established based on the analytical purpose, analyte concentration, and applicable regulatory requirements [22].

Detection Limit and Quantitation Limit

The detection limit (DL or LOD) represents the lowest amount of analyte that can be detected, although not necessarily quantified with acceptable accuracy and precision. The quantitation limit (QL or LOQ) represents the lowest amount that can be quantitatively determined with suitable performance.

ICH Q2(R2) describes several scientifically justified approaches, including signal-to-noise evaluation, calibration-based estimation, and, where applicable, direct experimental assessment.

USP and Chinese Pharmacopeia requirements similarly depend on the analytical application, whereas ANVISA RDC 166/2017 specifies approaches to detection and quantitation limit evaluation within its regulatory framework [23,24].

Analytical Range

The analytical range is the interval over which a procedure has demonstrated suitable analytical performance for its intended purpose.

ICH Q2(R2) emphasizes the reportable range and the validation evidence supporting performance across that interval. This includes the appropriate relationship between calibration, accuracy, and precision.

USP (1225) addresses range in relation to the analytical application and the relevant performance characteristics. Chinese Pharmacopoeia guidance also links the range to the analytical purpose, while ANVISA requires evaluation of the working interval according to the applicable method category.

Importantly, an instrument calibration interval should not automatically be considered the validated reportable range. The reportable range must be supported by evidence of acceptable method performance [25,26].

Robustness

Robustness evaluates an analytical procedure's ability to remain suitable when small, deliberate variations are introduced into method parameters. Typical chromatographic factors include mobile-phase composition, flow rate, column temperature, detection conditions, and buffer pH.

ICH Q14 positions robustness evaluation primarily within analytical procedure development, and ICH Q2(R2) allows relevant development evidence to support validation. USP and Chinese Pharmacopeia frameworks also recognize robustness as important for demonstrating method reliability.

ANVISA RDC 166/2017 includes robustness within its validation framework and expects evaluation of relevant method parameters according to the analytical procedure.

Risk assessment and design of experiments can improve robustness investigations by identifying critical parameters and interactions more efficiently than one-factor-at-a-time changes [27,28].

Table 02: Comparison of principal analytical validation characteristics and regulatory expectations

Parameter	ICH Q2(R2)	USP (1225)	ChP 2025	ANVISA RDC 166/2017
Specificity/selectivity	Absence of interference, orthogonal comparison, or justified alternative	Appropriate discrimination of analyte	Application-specific specificity assessment	Selectivity evaluation addressing relevant interference
Linearity/calibration	Appropriate calibration model; scientific justification	Linearity assessment where applicable	Calibration assessment aligned with	≥5 concentrations, triplicate preparations; r

			analytical purpose	>0.990
Accuracy	Reference material, spiking, or procedure comparison	Accuracy demonstrated by suitable experimental studies	Accuracy appropriate to test category	Defined experimental and replicate requirements
Precision	Repeatability and intermediate precision; reproducibility when relevant	Precision appropriate to analytical category	Repeatability and other relevant precision levels	Repeatability and intermediate precision assessment
Detection limit	Suitable experimental or statistical approach	Required for relevant low-level tests	Applicable to detection-oriented tests	DL determination where relevant
Quantitation limit	Demonstrated suitability at lower quantitative boundary	Relevant to quantitative impurity procedures	Applicable to low-level quantitative tests	QL evaluation under relevant provisions
Range	Performance demonstrated over reportable interval	Supported by accuracy, precision, and response behavior	Application-specific validated range	Working interval supported by validation
Robustness	Generally investigated during development	Evaluated as appropriate	Included within procedure performance assessment	Robustness evaluation under applicable provisions
System suitability	Integral to procedure control strategy	Linked to method and compendial requirements	Method- and monograph-specific	Suitability of analytical system demonstrated as appropriate

Statistical Considerations in Analytical Method Validation

Statistical evaluation is essential for determining whether validation data adequately demonstrate analytical procedure performance. Traditional validation practices have frequently relied on correlation coefficients, percentage recovery, and relative standard deviation. Although these parameters remain useful, they may not independently characterize the complete analytical measurement process. Modern validation increasingly emphasizes residual analysis, variance evaluation, confidence intervals, measurement uncertainty, and the relationship between accuracy and precision. Calibration models should be assessed for systematic deviation, inappropriate weighting, heteroscedasticity, and potential lack of fit.

ICH Q2(R2) provides flexibility to select statistical tools according to the analytical procedure and its intended purpose. ANVISA RDC 166/2017 explicitly requires several statistical evaluations for linearity, making statistical documentation a particularly important component of Brazilian regulatory submissions.

The 2025 Chinese Pharmacopeia revision of General Chapter 9101 also expands statistical considerations and introduces a combined assessment of accuracy and precision.

For pharmaceutical laboratories, the most effective approach is to establish statistical acceptance criteria before executing validation experiments. This reduces the risk of retrospectively selecting favorable criteria and supports objective evaluation of analytical performance [29].

Analytical Quality by Design and Lifecycle Management

Analytical Quality by Design (AQbD) provides a systematic approach to analytical procedure development based on predefined objectives, scientific understanding, risk assessment, and experimental optimization.

ICH Q14 establishes a framework for analytical procedure development, including the analytical target profile, parameter evaluation, control strategy, and lifecycle-related considerations [2]. ICH Q2(R2) complements this approach by providing the principles for demonstrating that the developed procedure meets its intended performance requirements.

The USP analytical procedure lifecycle framework similarly supports integrating development, validation, and ongoing procedure performance assessment.

The Chinese Pharmacopeia's 2025 revision incorporates analytical lifecycle concepts and modern validation approaches, demonstrating convergence with international scientific principles.

Although ANVISA RDC 166/2017 predates the finalization of ICH Q14, risk-based development and experimental design can provide additional scientific justification for methods intended for Brazilian regulatory submissions. Such approaches supplement rather than replace the applicable regulatory requirements.

AQbD is particularly valuable for chromatographic procedures because interacting factors may influence method performance, including mobile-phase composition, pH, temperature, flow rate, and stationary-phase characteristics. Multivariate experimental designs can identify method operating regions associated with acceptable analytical performance [30].

Regulatory Harmonization and Practical Challenges

Regulatory harmonization seeks to reduce unnecessary differences between jurisdictions while maintaining scientifically appropriate analytical performance standards. The four frameworks examined in this review share several fundamental principles: analytical procedures must be fit for purpose, validation characteristics should reflect intended use, and the resulting data must support reliable pharmaceutical quality decisions. However, differences remain in the degree of regulatory prescription, experimental design, statistical documentation, and application of compendial standards. A significant challenge arises when laboratories use a single validation protocol for multiple jurisdictions without reviewing local requirements. For example, an ICH-compliant calibration study may require additional experimental or statistical documentation to satisfy the explicit provisions of ANVISA RDC 166/2017.

Similarly, when implementing USP procedures, consider the distinction between method validation and compendial method verification. USP (1226) makes clear that verification involves demonstrating suitability under actual laboratory conditions rather than unnecessarily repeating complete compendial validation [39,40]. An additional challenge is the transition between regulatory editions. The 2025 Chinese Pharmacopoeia revision of General Chapter 9101 reflects substantial modernization, including lifecycle principles, multivariate methods, statistical assessment, and the relationship between robustness and system suitability. Accordingly, comparisons based exclusively on older pharmacopoeial editions may no longer represent current expectations.

Table 03: Major regulatory differences, potential challenges, and recommended harmonization strategies

Regulatory issue	Principal difference	Practical challenge	Recommended strategy
Regulatory scope	ICH guidance, USP compendial standards, ChP requirements, and ANVISA regulation have different legal roles	Incorrect assumptions about cross-jurisdiction acceptance	Establish a jurisdiction-specific compliance matrix
Calibration design	ANVISA specifies detailed replicate and statistical requirements	Insufficient calibration evidence	Design calibration studies to satisfy the most demanding applicable requirements
Acceptance criteria	Criteria vary with method purpose and regulatory context	Inappropriate universal limits	Predefine scientifically justified performance criteria
Statistical assessment	Different levels of prescription for regression and residual analysis	Incomplete statistical documentation	Include regression diagnostics and appropriate uncertainty evaluation
Validation versus verification	USP distinguishes these activities	Unnecessary repetition or insufficient testing	Classify the procedure and its intended use before planning studies
Robustness	Development-based versus validation-focused approaches	Duplication of experiments	Integrate robustness studies with risk-based development
Lifecycle management	Different levels of formal integration	Inconsistent postvalidation monitoring	Establish continued procedure performance monitoring
Regulatory updates	Guidelines and pharmacopoeias undergo periodic revision	Use of superseded requirements	Confirm the applicable edition and implementation status before submission

Proposed Harmonized Validation Strategy

A practical international validation strategy should begin with defining the analytical procedure's intended use and identifying all relevant regulatory requirements. The analytical target profile can then be used to establish measurable performance expectations, including specificity, accuracy, precision, reportable range, and appropriate detection capability.

During method development, identify critical analytical parameters using prior knowledge, risk assessment, and experimental investigation. Robustness studies should support selection of suitable operating conditions and the establishment of an analytical

procedure control strategy. The validation protocol should subsequently be designed to satisfy the applicable requirements of each target jurisdiction. Where requirements differ, laboratories may adopt an experimental design that satisfies the more prescriptive provisions, provided it remains scientifically appropriate for the method.

For example, a chromatographic assay intended for international registration may use a calibration study that incorporates sufficient concentration levels, independent preparations, replicate measurements, residual evaluation, and appropriate regression diagnostics. Such a study may provide evidence useful

across several regulatory frameworks. After validation, evaluate routine analytical performance through system suitability testing, quality control samples, trend analysis, and investigation of atypical results. Assess method changes based on their potential impact on analytical performance and applicable regulatory requirements. This integrated approach can improve validation efficiency without compromising analytical reliability.

FUTURE PERSPECTIVES

The future of analytical method validation is expected to emphasize science-based regulatory convergence, advanced statistical evaluation, and integration of analytical procedure lifecycle management. Greater adoption of analytical quality by design may improve understanding of method variability and support more efficient validation strategies. Emerging analytical technologies, including spectroscopic platforms, multivariate calibration models, automated chromatography, and process analytical technology, create additional opportunities for improving pharmaceutical quality assessment. However, these technologies also require validation approaches that can evaluate complex analytical responses and data-processing procedures.

Regulatory convergence may be supported by wider acceptance of performance-based criteria, scientifically justified experimental designs, and structured risk assessments. The modernization of the Chinese Pharmacopoeia and proposed evolution of USP validation guidance indicate increasing alignment with contemporary international principles. Further progress will depend on transparent regulatory implementation, consistent interpretation of validation terminology, and continued collaboration among regulatory authorities, pharmacopoeial organizations, academic researchers, and pharmaceutical manufacturers.

CONCLUSION

Analytical method validation remains fundamental to pharmaceutical quality assurance and regulatory compliance. This review demonstrates substantial agreement among ICH Q2(R2), USP (1225), the Chinese Pharmacopoeia, and ANVISA RDC 166/2017 regarding the need to establish analytical procedures that are suitable for their intended purposes. Nevertheless, differences persist in experimental requirements, statistical evaluation, documentation, and regulatory implementation. ICH Q2(R2) emphasizes scientific flexibility and performance-based validation, whereas USP provides compendial validation and verification principles. The Chinese Pharmacopoeia incorporates updated analytical lifecycle concepts, while ANVISA specifies comparatively detailed experimental and statistical requirements. A harmonized validation strategy should therefore integrate common scientific principles with jurisdiction-specific expectations. Combining risk-based method

development, appropriate statistical assessment, predefined acceptance criteria, and continued performance monitoring can reduce redundant validation activities and improve international regulatory readiness. Further convergence of validation practices will support analytical reliability, pharmaceutical quality, and efficient global drug development.

CONFLICT OF INTEREST

The authors declare no conflicts of interest.

AUTHOR CONTRIBUTIONS

All authors contributed to the literature review, manuscript preparation, and critical revision and approved the final manuscript.

ETHICAL APPROVAL

Not applicable.

REFERENCES

1. International Council for Harmonisation. ICH Q2(R2): Validation of Analytical Procedures. Geneva: ICH; 2023. Official guideline.
2. International Council for Harmonisation. ICH Q14: Analytical Procedure Development. Geneva: ICH; 2023. FDA publication.
3. United States Pharmacopeia. General Chapter (1225): Validation of Compendial Procedures. USP–NF. Rockville (MD): United States Pharmacopeial Convention; 2017. doi:10.31003/USPNF_M99945_04_01.
4. International Council for Harmonisation. ICH Q2(R1): Validation of Analytical Procedures: Text and Methodology. Geneva: ICH; 2005.
5. United States Pharmacopeia. General Chapter (1226): Verification of Compendial Procedures. USP–NF. Rockville (MD): United States Pharmacopeial Convention; 2019. doi:10.31003/USPNF_M870_03_01.
6. United States Pharmacopeia. General Chapter (621): Chromatography. USP–NF. Rockville (MD): United States Pharmacopeial Convention.
7. Chinese Pharmacopoeia Commission. Pharmacopoeia of the People's Republic of China. 2025 ed. Vol IV. Beijing: China Medical Science Press; 2025.
8. Xu X, Wei S, Zhang Q, He L, Zhang J, Ma S. Additions and revisions of general chapters of physical and chemical analysis in the Chinese Pharmacopoeia 2025 Edition Volume IV. Drug Standards of China. 2025. Publication record.
9. Agência Nacional de Vigilância Sanitária. Resolução da Diretoria Colegiada RDC nº 166, de 24 de julho de 2017: Dispõe sobre a validação de métodos analíticos e dá outras providências. Brasília: ANVISA; 2017. Official regulation.
10. Ermer J, Miller JHMcB, editors. Method Validation in Pharmaceutical Analysis: A Guide to Best Practice. Weinheim: Wiley-VCH; 2005.

11. Miller JN, Miller JC. Statistics and Chemometrics for Analytical Chemistry. 6th ed. Harlow: Pearson Education; 2010.
12. United States Pharmacopeia. General Chapter (1220): The Analytical Procedure Lifecycle. USP–NF. Rockville (MD): United States Pharmacopeial Convention.
13. Vogt FG, Kord AS. Development of quality-by-design analytical methods. *J Pharm Sci*. 2011;100(3):797–812. doi:10.1002/jps.22325.
14. Borman P, Nethercote P, Chatfield M, Thompson D, Truman K. The application of quality by design to analytical methods. *Pharm Technol*. 2007;31(12):142–152.
15. United States Food and Drug Administration. PAT—A Framework for Innovative Pharmaceutical Development, Manufacturing, and Quality Assurance. Silver Spring (MD): FDA; 2004.
16. Murali Krishnam Raju P, Shyamala P, Venkata Narayana B, Dantuluri HSNR, Bhupatiraju RV. A fast, validated UPLC method coupled with PDA-QDa detectors for impurity profiling in betamethasone acetate and betamethasone phosphate injectable suspension and isolation, identification, characterisation of two thermal impurities. *Annales Pharmaceutiques Françaises*. 2022 Nov;80(6):837–52.
17. Byram KK, Patel C, Patel S, Muthyam SK. A Review of the Role of FTIR, NMR, and Raman Spectroscopy in Drug Product Characterisation. *UPI Journal of Pharmaceutical, Medical and Health Sciences*. 2026 May 6;1–7.
18. Kumar A. T. “Development and validation of RP-LC-UV method for determination of ursodeoxycholic acid in drug substance and drug product”. *Journal of Global Trends in Pharmaceutical Sciences*. 2016;7(3):3429-35.
19. Tallam AK, Alapati S, Nuli MV. RP-HPLC method for analytical method development and validation of multi-kinase inhibitor. *Journal of Integral Sciences*. 2023 Feb 25;20–6.
20. Narayanan BV, Sreenivas J, Rao VS. Spectrophotometric Determination of Trace Amounts of Molybdenum (VI) Using 3,4-Dihydroxybenzaldehyde Thiosemicarbazone. *Research Journal of Chemistry and Environment*. 2006;10:67-70.
21. Mukuntha Kumar M, Rao SV, Abbulu K, Narayana BV, Sukumar I. Simultaneous Estimation of Telmisartan and Chlorthalidone in Tablet Dosage Form by Using Reversed Phase High-Performance Liquid Chromatographic Method. *American Journal of Pharm Tech Research*. 2014;4(5):696-710.
22. Murali Krishnam Raju P, Shyamala P, Venkata Narayana B, Kondra S, Mantena BPV. Assay of tirofiban and identification of oxidative impurity in aqueous injection by using UPLC-PDA/QDa detectors. *Annales Pharmaceutiques Françaises*. 2022 Jan;80(1):35–47.
23. Kumar Byram K, Patel S, Patel C, Kumar Muthyam S. Forced Degradation Studies in Analytical Method Development for Pharmaceuticals: A Comprehensive Review. *International Journal of Pharmacognosy and Chemistry*. 2026 Feb 28;1–8.
24. Mukuntha Kumar N, Rao SV. RP-HPLC method development and validation for the simultaneous determination of Telmisartan and Atorvastatin in bulk and pharmaceutical dosage form. *International Journal of Scientific Research*. 2014;3(11):357-9.
25. Mohan Kk, Patrudu Tb, Burle Gs, Salakolusu S, Raju Pvn, Jonnalagadda Sb, et al. Isolation and structural elucidation of an unknown novel impurity in sulfasalazine by high-performance liquid chromatography coupled to mass spectroscopy and toxicology prediction. *Chinese Journal of Analytical Chemistry* [Internet]. 2025 Jul 19;53(11):100601.
26. Kallam SDM, Rudrapal M, Bodapati A. Development and validation of a stability-indicating UPLC-based method for simultaneous quantification of trifluridine, tipiracil hydrochloride and their impurities/related substances with characterisation of degradation products by UPLC-MS/MS. *Accreditation and Quality Assurance*. 2025 Sept 9;30(6):735–50.
27. Pokuri Chiranjeevi, Rao S, V. Rithvika, Anoop Bodapati, HariHaran K, Kanamaneni Chinna Kondanna, et al. Current Advances in Pharmaceutical Analytical Methods for the Aminoglycoside Antibiotic: Gentamicin. *International Journal of Advanced Science and Engineering*. 2025 Sept 27;12(1):5335–56.
28. Bodapati A, Pagala B. An Optimised UHPLC Technique for the Identification and Quantification of the Related Impurities for Cedazuridine/Decitabine Combination and the Characterisation of the Forced Degradation Products Through MS/MS. *Separation Science Plus*. 2025 May;8(5).
29. Burle GS, Kakullamarri PR, Kothamasu SB, Kallam SDM, Bodapati A. Case Study on Regulatory Approaches for New Degradation Impurity Exceeding ICH Thresholds in Solubilised Ibuprofen Capsules During Stability Testing. *Journal of International Research in Medical and Pharmaceutical Sciences*. 2024 Nov 7;19(3):70–82.
30. P. Laxmiprasanna, C. R. S., M. Vamsi Nuli, U.V. Naga Venkata Arjun, P. Vidiyala, B. Sasivardhan Reddy, J. Gopiseti, K. Krishnan. Analytical Method Development and Validation for a Fixed-Dose ALS Combination Using RP-HPLC: A QbD and Response Surface Optimisation Approach. *Adv. J. Chem. A*, 2025, 8(11), 1789-1809.