www.pharmaerudition.org

ISSN: 2249-3875

International Journal of Pharmaceutical Erudition

Research for Present and Next Generation

NOV 2025

Vol: 15 Issue:03

(18--24)

Review Article

A REVIEW ON HPLC METHOD DEVELOPMENT AND VALIDATION

Soni Neha *, Goyal Rakesh

Dept. of Pharmaceutical Quality Assurance, Mahatma Gandhi College of Pharmaceutical Sciences,

Jaipur - 302022

Analytical technique development, validation, and transfer are critical components of any pharmaceutical development effort. Effective method development optimizes laboratory resources and ensures techniques meet drug development objectives at all stages. High performance liquid chromatography is a reliable technology for analyzing medicinal products, both qualitatively and quantitatively. Developing and validating analytical methods is crucial for drug discovery, development, and manufacturing. It comprises determining the purity and toxicity of a pharmacological substance. Method development for the interested component in finished product or in process tests and the sample preparation of drug product and to provide practical approaches for determining selectivity, specificity, limit of detection, limit of quantitation, linearity, range accuracy, precision, recovery solution stability, ruggedness, and robustness of liquid chromatographic methods to support the Routine, in process and stability analysis.

Keywords: Analytical Method Development, Method Validation, HPLC, specificity, stability analysis.

INTRODUCTION

Any product or service needs analysis, but since drugs involve human life, they require it much more. The study of separation, measurement, and identification of chemical additives is known as analytical chemistry.[1]

Materials made of herbs and synthetics that contain one or more substances or ingredients. Two main classes make up analytical chemistry: qualitative evaluation, which identifies the chemical additives present in the sample, and quantitative evaluation, which calculates the quantity of positive detail or compound present in the substance, or sample.[2]

Pharmaceutical analysis plays a very outstanding role in the examination of pharmaceutical formulations and bulk drugs regarding the quality control and assurance.[3]

The development of analytical tools has led to advances in scientific and practical analytical approaches. The time and cost of analysis have decreased and precision and accuracy have increased due to advancements in analytical technique development and analytical instrumentation. An essential component of the requirements for regulatory organizations is the development and validation of analytical techniques for active pharmaceutical ingredients, excipients, related substances, drug products, degradation products, residual solvents, etc.[4]

Analytical method development finally results in official test method. Consequently quality control laboratories used these methods to check the efficacy, identity, purity, safety as well as

performance of products of the drug. The significance of analytical procedures in production is highly valued by regulatory bodies. Drug approval by regulatory authorities requires the applicant to prove control of the entire process of drug development by using validated analytical methods.[5-6]

Definition On Validation

The FDA (FOOD AND DRUG ADMINISTRATION) defines validation as a production and process control procedure that ensures the identity, strength, quality, and purity of drug products. FDA guidelines from May 1987 state that the validation package needs to include all the data and test procedures needed to demonstrate that the system and process satisfy the standards.[7-8]

Analytical Method Development

For the analysis of novel products, new methods are being developed when authoritative methods are not accessible. Novel techniques are created to examine the current pharmacopoeial or non-pharmacopoeial products and save costs while improving robustness and precision.[9]

These methods are optimized and validated through trial runs. With all available benefits and drawbacks, alternative approaches are suggested and implemented to replace the current strategy in the comparative laboratory data.[10-11]

Purpose Of Analytical Method Development

The identification, characterization, and determination of pharmaceuticals in mixtures, such as dosage forms and biological fluids, are revealed by drug analysis. The primary goal of analytical methods in the manufacturing process and drug development is to provide information about potency (which can be directly related to the need for a known dose), impurity (related to the drug's safety profile), bioavailability (which includes important drug characteristics like crystal form, drug uniformity, and drug release), stability (which indicates the products of degradation), and the impact of manufacturing parameters to guarantee consistent drug product production.[12-13]

The goal of quality control is to evaluate and identify a true and correct product through a set of procedures meant to prevent and eliminate mistakes at various production stages. A product's release or disposal decision is based on one or more types of control actions. Ensuring a straightforward and analytical procedure for diverse complex formulations is an extremely significant topic. The need for new analytical techniques in the pharmaceutical industry has quickly increased due to the industries' rapid growth and continuous drug production across the globe. As a result, developing analytical methods has become the fundamental analysis task in a quality control laboratory.[14]

Need Of Analytical Method Development Validation-

- Available method can be too costly, time

ingesting or power extensive, or that won't be without problems computerized.

- Present approach can be too much errors, infection susceptible or they may be unreliable.
- Present approach may not offer adequate sensitivity.
- For requirements related to regulations, it's necessary.
- The main criteria for choosing drugs while developing a new analytical method.
- The drug or drug combination might not be dependable according to any pharmacopoeias.
- Because of the interference caused by the excipients in the formula, analytical procedures may not be available for the drug in the form of a formula.
- There won't be an analytical method available for quantifying the medication in biological fluids.
- There won't be analytical methods accessible for a medicine when combined with other drugs.[15-16]

Analytical Method Validation[17-18]

Method validation, as defined by ICH Q2 (R1), involves establishing written proof that a process consistently produces desired results within predetermined parameters and quality characteristics.

Analytical processes must be appropriate for their intended application and support the identity, quality, purity, and potency of pharmacological substances and products. Method validation is necessary for both new and established methods utilized across several laboratories and analysts.

The performance characteristics required to validate various methods by using various guidelines such as USP, ICH, FDA, European guidelines etc.

1. According to USP

The analytical parameters can be validated are accuracy, precision, specificity, detection of limit, quantitation limit, linearity, range, ruggedness and robustness.

2. According to ICH

The analytical parameters can be validated are accuracy, precision, specificity, detection of limit, quantitation limit, linearity, range, system suitability and robustness.

3. According to FDA

The analytical parameters can be validated are accuracy, precision, specificity/selectivity, detection of limit, quantitation limit, linearity, range, system suitability, reproducibility, sample solution stability and robustness.

4. According to European guidelines

The analytical parameters can be validated are accuracy, precision, specificity, detection of limit, quantitation limit, linearity and range

Analytical methods need to be validated, verified, or revalidated in the following instances[19]

- Before initial use in routine testing
- When transferred to another laboratory
- Whenever the conditions or method parameters for which the method has been validated change (for example, an instrument with different characteristics or samples with different matrix).

Types of analytical procedures to be validated[20]

The following types analytical procedures to be validated.

- Identification tests
- Quantitative tests for impurities content
- Limit tests for the control of impurities
- Quantitative tests of the active moiety in samples of drug substance or drug product.

1. Identification Test –

Identification tests are used to ensure the identity of an analyte in a sample. This is normally achieved by comparison of a property of the sample to that of a reference standard

2. Quantitative tests and Limit tests for impurity control-

Testing of impurities can be performed by using a quantitative test or a limit test for the impurity in a sample. Different validation parameters are required for a quantitative test than for a limit test

3. Quantitative tests of the active moiety in samples of drug substance or drug product-

In this type, assay procedures are used to measure the analyte present in a given sample. The assay represents a quantitative measurement of the major component(s) in the drug substance.

Objectives Of Method Validation[21-22]

- To obtain consistent, reliable and true data.
- To demonstrate that it is suitable for its intended purpose.
- To form a base for written procedure for production and process control which are designed to assure that the drug products have the identity, strength, quality and purity.
- To hold the quality, safety and efficacy in final product.
- To control each step of manufacturing process.
- To produce the best analytical results possible.
- It decreases risk of regulatory noncompliance
- Critical parameters of the process can be fully understood due to analytical method.
- Minimization of interference on accuracy and precision

Advantages of Method Validation[23-24]

- It builds a degree of confidence, not only for the developer but also to the user.
- Produces quality products.
- Reduce the product cost by increasing efficacy, few reject and longer equipment life.

- Helps in optimization of process or method.
- Helps in process improvement, technology transfer related products validation and increased employee awareness. It eliminates testing repetitions and leads to better time management in the end.

In high-performance liquid chromatography, a compound with lower affinity for the stationary phase travels faster and covers a longer distance, while a compound with higher affinity moves slower and covers a shorter distance. This differential migration facilitates effective separation and analysis of sample components. High performance liquid chromatography (HPLC) proves invaluable in pharmaceutical analysis, efficiently isolating and quantifying major medications, reaction impurities, synthesis intermediates, and degradants. As a preeminent analytical tool, HPLC excels in identifying, measuring, and separating diverse sample components soluble in liquid. Its precision is paramount for both quantitative and qualitative drug product analysis, playing a pivotal role in determining drug product stability. By offering a meticulous approach to characterizing pharmaceutical samples, HPLC stands as an indispensable technique in ensuring the quality and safety of medicinal formulations in the field of analytical chemistry.[25-26]

CONCLUSION

This review article gives idea that what is validation, its type, their purpose and why it is necessary and gives information about all validation parameter such as linearity, accuracy, precision, Range, LOD, LOQ, specificity etc. Validation is necessary technique in the pharma department and it is used to assure that the quality is worked into the procedure supporting the development of drug and production. The main objective of this review article is to improve the quality of analytical method development and validation.

REFERENCE

1. Noman A, Bukhaiti ALWedad Q, Alfarga A, Abed Sherif M, Mahdi AA. And Waleed AA. HPLC technique used in food analysis-Review. International Journal of Agriculture Innovations and Research. 2021; 5(2):181-88.
2. Snyder LR, Kirkland JJ, Dolan JW. Introduction to modern liquid chromatography. John Wiley & Sons. New York. 2020.
3. Xiang Y, Liu Y, Lee ML. Ultrahigh pressure liquid chromatography using elevated temperature. Journal of Chromatography. 2016; 1104(1): 198-202.
4. Horvath CG, Preiss BA, Lipsky SR. Fast liquid chromatography. Investigation of operating parameters and the separation of nucleotides on pellicular ion exchangers. Analytical chemistry, 1967; 39(12): 1422-28.
5. Malviya R, Bansal V, Palo P, and Sharma PK. High Performance Liquid Chromatography: A Short Review. Journal of Global Pharma Technology. 2023; 2(5):22-23.

6. Pratap B. et al. Importance of RP-HPLC in Analytical method development: A review. 2022.
7. International journal of novel trends in pharmaceutical sciences 2023; 3(1): 15-23.
8. Lindholm J. Development and Validation of HPLC method for Analytical and Preparative Purpose. Acta Universities Upsaliensis Uppsala. 2014; 13-14.
9. Chetta N. et.al. Development and validation of a stability indicating high performance liquid chromatographic (HPLC) method for Atenolol and hydrochlorothiazide in bulk drug and tablet formulation. Int J Chem tech res. 2013; 1(3): 654-662.
10. Kumar GS. et.al. Development and validation of RP-HPLC method for simultaneous estimation of Atenolol and Chlorthalidone in Bulk and dosage form. Int Res J Pharma 2013; 3(10): 215-19.
11. FDA Guidance for Industry -Analytical Procedures and Method Validation, Chemistry, Manufacturing, and Controls Documentation, Center for Drug Evaluation and Research (CDER) and Center for Biologics Evaluation and Research (CBER). 2000.
12. Julia T, Mena AJ, Aucoin MG, Kamen AA. Development and validation of a HPLC methodfor the quantification of baculovirus particles. J Chromatogr B. 2021; 879: 61-68.
13. Nevado JJB et.al. Reliable and Sensitive SPE-HPLC-DAD Screening of Endocrine Disruptors Atrazine, Simazine and their Major Multi residues in Natural Surface Waters: Analytical Validation and Robustness Study Performance. J Chromatograph Separat Techniq 2014; 5: 215.
14. Nia Y et.al. Determination of Ti from TiO2 Nano particles in Biological Materials by Different ICP-MS Instruments: Method Validation and Applications. J Nano med Nanotechnol.2015; 6:269.
15. A. Shrivastava, V.B. Gupta, HPLC: Isocratic or Gradient Elution and Assessment of Linearity in Analytical Methods, J Adv Scient Res. 2020; 3(2) 12-20.
16. Weston A, Brown PR. HPLC and CE Principles and Practice. Academic press California; 1997.
17. Ngwa G. Forced Degradation Studies. Forced Degradation as an Integral part of HPLC Stability Indicating Method Development. Drug Delivery Technology. 2010; 10(5).
18. Gad S. Pharmaceutical manufacturing handbook of regulations and quality. John wiley and sons; 2006.
19. Webster P. Analytical procedures and method validation. Environmental protection agency; 2001.
20. Mohamad T, Mohamad MA, Chattopadhyay M. Particle size role, Importance and Strategy of HPLC Analysis An update. International Archives of Bio Medical and Clinical Research. 2016; 2(2): 5-11.
21. Weston A, Brown PR. HPLC and CE Principles and Practice. Academic press California; 1997.

22. Ngwa G. Forced Degradation Studies. Forced Degradation as an Integral part of HPLC Stability Indicating Method Development. Drug Delivery Technology. 2010; 10(5).
23. Christopher R M, Thomas W R. Quality systems approach to pharmaceutical cGMP. Development and Validation of Analytical Methods, 11th Edition. 2021.
Sharma, B., Instrumental Methods of Chemical Analysis, thirteen ed. Goel publisher house, Meerut, 2018, 184-186.
24. Devalarao, G., Text Book of Pharmaceutical Analysis, second ed. Birla publications, New Delhi, 2015, 1-5.
25. Ewing, G., Instrumental Methods of Chemical Analysis, fifth ed. Mc graw- hill book company, New York, 2005, 1-7.