Bhagyasree.et al. / Vol 4 / Issue 8/ 2014 /444-448.

International Journal of Pharmaceutical Research & Analysis

e-ISSN: 2249 – 7781
Print ISSN: 2249 – 779X

www.ijpra.com

A REVIEW ON ANALYTICAL METHOD DEVELOPMENT AND VALIDATION

Bhagyasree T*[1], Neelam I[1], Ajitha A[1], Uma Maheshwara Rao V[1]

Department of Pharmaceutical Analysis & Quality Assurance, CMR College of Pharmacy, JNTU (H) University, Hyderabad, Andhra Pradesh, India.

ABSTRACT

Analytical methods development must be validated to provide reliable data for regulatory submissions. Method development is the process of proving that an analytical method is acceptable for use to measure the concentration of an API in a specific compounded dosage form which allow simplified procedures to be employed to verify that an analysis procedure, accurately and consistently will deliver a reliable measurement of an active ingredient in a compounded preparation.These methods are essential for a number of purposes, including testing for quality control release, testing of stability sample, testing of reference materials and to degree of assurance and is an important process in the drug discovery. Although the drug shows good potency, lack of validated analytical method will not allow the drug to enter into the market. This is to ensure the quality and safety of he drug. This review gives ideas about various methods to check the stability of dug and various validation parameters as per various regulatory authorities.

Keywords: Method Development, Drug analysis, Validation, ICH.

INTRODUCTION

Analytical methods development and validation play important roles in the discovery, development, and manufacture of pharmaceuticals. Pharmaceutical products formulated with more than one drug, typically referred to as combination products, are intended to meet previously unmet patients need by combining the therapeutic effects of two or more drugs in one product [1]. These combination products can present daunting challenges to the analytical chemist responsible for the development and validation of analytical methods. There is a scope, therefore to develop newer analytical methods for such drugs. Also quality is important in every product or service but it is vital in medicines as it involves life [2].

BASIC CRITERIA FOR NEW METHOD DEVELOPMENT OF DRUG ANALYSIS:

- The drug or drug combination may not be official in any pharmacopoeias
- A proper analytical procedure for the drug may not be available in the literature due to patent Regulations
- Analytical methods may not be available for the drug in the form of a formulation due to the Interference caused by the formulation excipients
- Analytical methods for the quantization of the drug in biological fluids may not be available
- Analytical methods for a drug in combination with other drugs may not be available
- The existing analytical procedures may require expensive reagents and solvents. It may also involve cumbersome extraction and separation procedures and these may not be reliable [3].

METHOD DEVELOPMENT LIFE CYCLES

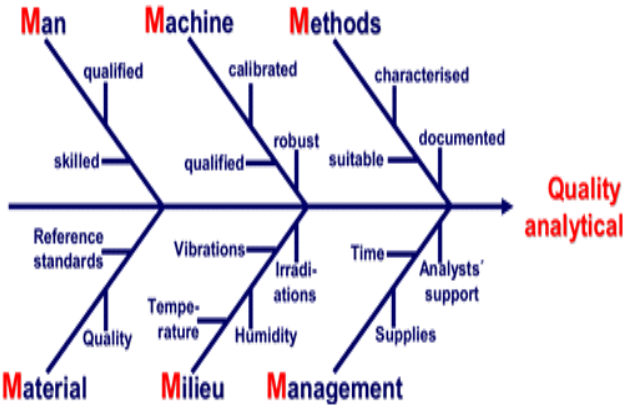
Steps in method development

The steps of method development and method validation

- Method development plan definition

Corresponding Author:-T.Bhagyasree Email:- bhagyasree63@gmail.com

- Background information gathering
- Laboratory method development, it includes various stages namely sample preparation, specific analytical method, detection and data processing [4]



Generation of test procedure

1. Sample information, define separation goals
2. Sample pre-treatment, need of special HPLC procedure
3. Selection of detector and detector settings
4. Selection of LC method; preliminary run; estimate best separation conditions
5. Optimize separation conditions
6. Check for problems or requirement for special procedure
7. Method validation [5]

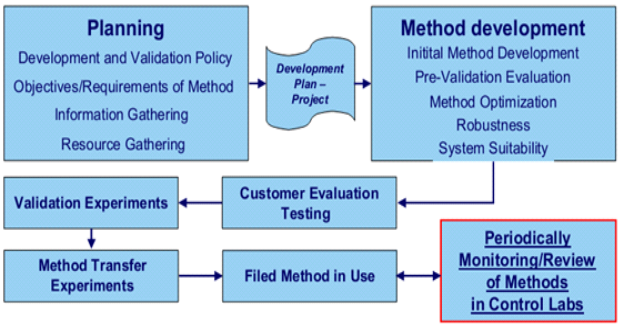
Sample information

1. Number of compounds present
2. Chemical structure of compounds
3. Chemical nature
4. Molecular weight of compounds
5. pKa Value(s) of compounds
6. Sample solubility
7. Sample stability and storage
8. Concentration range of compounds in sample
9. UV spectra of compounds or properties for detection of compounds [6].

A well-developed method should be easy to validate. A method should be developed with the goal to rapidly test preclinical samples, formulation prototypes, and commercial samples. There are five common types of analytical methods, each with its own set of validation requirements

- Identification tests
- Potency assays
- Quantitative tests for impurities
- Limit test for the control of impurities
- Specific tests [7]

The first four tests are universal tests, but the specific tests such as particle-size analysis and X ray diffraction are used to control specific properties of the active pharmaceutical ingredient(API) or the drug product.



Analytical Method Development Is Required for

- Herbal Products
- New process and reactions
- New molecules
- Active ingredients (Macro analysis)
- Residues (Microanalysis)
- Impurity Profiling
- Component of Interest in different matrices [8]

Analytical methodology contents

An Analytical Methodology consists of the following

- Techniques
- Method
- Procedure
- Protocol

Importance of analytical methodology to an analyst

- The required data for a given analytical problem
- The required sensitivity
- The required accuracy
- The required range of analysis
- The required precision

METHOD VALIDATION

Method validation is the process used to confirm that the analytical procedure employed for a specific test is suitable for its intended use. Results from method validation can be used to judge the quality, reliability and consistency of analytical results; it is an integral part of any good analytical practice. Use of equipment that is within specification, working correctly and adequately calibrated is fundamental to the method validation process. Analytical methods need to be validated or revalidated

• Before their introduction into routine use;

• whenever the conditions change for which the method has been validated (e.g., an

Instrument with different characteristics or samples with a different matrix); and

• whenever the method is changed [9].

Current Best Practice in Analytical Method Validation

Validation should not be seen separately from the development of a method. Therefore entire process of analytical method development and validation can be

considered in a totality as represented in the general scheme. The method's performance characteristics should be based on the intended use of the method. These include analyte, its expected concentration, sample matrix, possible interfering substances, regulatory requirement, application (qualitative/quantitative), requirement for robustness, detection and quantization limit, accuracy and precision expectation, different types of equipment and the locations where the method will be run, skill requirements for analyst, etc. Before an instrument is used to validate a method, its performance should be verified. But still after method development it needs to be validated as per requirement which gives certain level of confidence for its intended use [10].

Advantages of analytical method validation

The advantages of the analytical method validation are as follow:

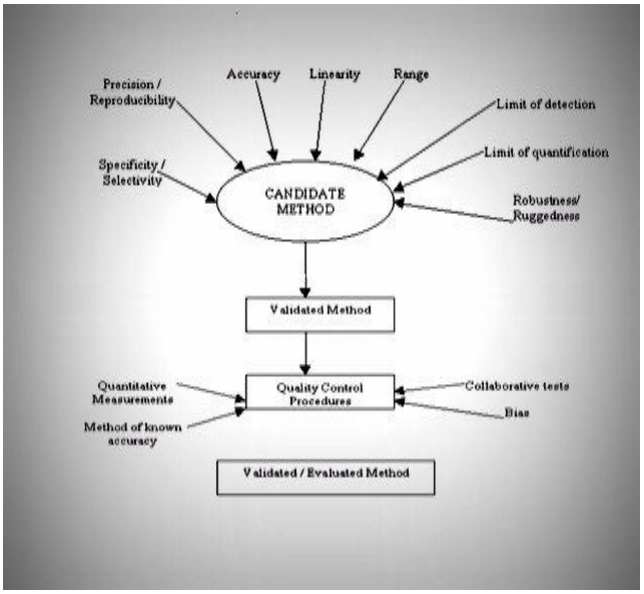
- The biggest advantage of method validation is that it builds a degree of confidence, not only for the developer but also to the user.
- Although the validation exercise may appear costly and time consuming, it results inexpensive, eliminates frustrating repetitions and leads to better time management in the end.
- Minor changes in the conditions such as reagent supplier or grade, analytical setup are unavoidable due to obvious reasons but the method validation absorbs the shock of such conditions and pays for more than invested on the process [11].

Requirements of analytical method validation

- Method Validation is required for the following reasons
- Assuring quality and achieving a levels
- Achieving acceptance of products by the international agencies.
- Mandatory requirement purposes for accreditation as per ISO 17025 guidelines.
- Mandatory requirement for registration of any pharmaceutical product or Pesticide formulation.
- Validated methods are only acceptable for undertaking proficiency testing [12].

When should methods be validated?

- A method should be validated when it is necessary to verify that its performance parameters are adequate for use for a particular analytical problem. For example
- Method just developed
- Revised method or established method adapted to a new problem
- When a review of quality control indicates an established method is changing with time
- When an established method is used in a different laboratory, with different analysts or with different equipment [13].



Typical parameters recommended by FDA, USP, and ICH are as follow

1. Specificity
2. Linearity & Range
3. Precision

(A) Method precision (Repeatability)
(B) Intermediate precision (Reproducibility)

4. Accuracy (Recovery)
5. Solution stability
6. Limit of Detection (LOD)
7. Limit of Quantification (LOQ)
8. Robustness

Selectivity/specificity

The terms selectivity and specificity are often used interchangeably, the term specific generally refers to a method that produces a response for a single analyte [14].

Linearity and Range

The linearity of an analytical procedure is its ability (within a given range) to obtain test results, which are directly proportional to the concentration (amount) of analyte in the sample. A linear relationship should be evaluated across the range of the analytical procedure. It is demonstrated directly on the drug substance by dilution of a standard stock solution of the drug product components, using the proposed procedure. For the establishment of linearity, minimum of five concentrations are recommended by ICH guideline. The range of an analytical method is the interval between the upper and lower levels (including these levels) that have been demonstrated to be determined with precision, accuracy and linearity using the method as written [15].

Precision

The precision of an analytical procedure expresses the closeness of agreement (degree of scatter) between a

series of measurements obtained from multiple sampling of the same homogeneous sample. Precision may be considered at two levels: repeatability and intermediate precision. The precision of an analytical procedure is usually expressed as the variance, standard deviation or coefficient of variation of a series of measurements.The two most common precision measures are 'repeatability' and reproducibility' [16].

Repeatability

Repeatability study is performed by preparing a minimum of 6 determinations a 100% of the test concentration and analyzed as per the respective methodology [17].

Intermediate Precision

The extent to which intermediate precision should be established depends on the circumstances under which the procedure is intended to be used. The analyst should establish the effects of random events on the precision of the analytical procedure. Typical variations to be studied include days, analysts, equipment, etc. It is not considered necessary to study these effects individually.

Accuracy

The accuracy of an analytical procedure expresses the closeness of agreement between the value which is accepted either as a conventional true value or an accepted reference value and the value found. The evaluation of accuracy has got very prime importance as it deliberately force the method to extract the drug and impurities at higher and lower level.If certified Reference materials or control samples are not available, a blank sample matrix of interest can be spiked with a known concentration by weight or volume [18].

Solution stability

Drug stability in pharmaceutical formulations/active pharmaceutical ingredients is a function of storage conditions and chemical properties of the drug, preservative and its impurities. Condition used in stability experiments should reflect situations likely to be encountered during actual sample handling and analysis. Stability data is required to show that the concentration and purity of analyte in the sample at the time of analysis corresponds to the concentration and purity of analyte at the time of sampling [19-20].

Limit of detection

The limit of detection (LOD) for an individual analytical procedure is the lowest amount of analyte in a sample, which can be detected but not necessarily quantitated as an exact value. Determination of the signal-to-noise ratio is performed by comparing measured signals from samples with known low concentrations of analyte with those of blank samples and establishing the minimum concentration at which the analytecan be reliably detected. A signal-to-noise ratio between 3 or 2:1 is generally considered acceptable for estimating the detection limit. The limit of detection is the point at which a measured value is larger than the uncertainty associated with it [21-23].

Limit of Quantitation

The quantitation limit of an individual analytical procedure is the lowest amount of analyte in a sample, which can be quantitatively determined with suitable precision and accuracy. The limit of quantitation (LOQ) is a parameter of quantitative assays for low levels of compounds in sample matrices. A typical signal-to-noise ratio is 10:1 [24].

Robustness

The robustness of an analytical procedure is a measure of its capacity to remain unaffected by small but deliberate variations in method parameters and provides an indication of its reliability during normal usage.In case of liquid chromatography the factors chosen for all the drugs under investigation were the flow rate, mobile phase composition, pH of a mobile phase and using different lot of LC column [25].

Ruggedness is normally evaluated during method development, typically by the originating Laboratory, before collaborating with other laboratories and is a measure how well a method stands up to less than perfect implementation [26].

CONCLUSION

In the recent years the introduction of new drug molecules are increased and for these there is no standards and analytical procedures in pharmacopeias. so, analytical method development and validation is playing an important role in most of the pharmaceutical industries. The method validation should be very useful for industries when introduction of new methods for their routine work . For reporting of new toxicities and patient resistance method development and validation should be very important criteria in pharmacy field. Analytical methodology provides to an analyst the required data for a given analytical problem, sensitivity, accuracy, range of analysis, precision i.e. the minimum requirements which essentially are the specifications of the method for the intended purpose to be able to analyse the desired analyte in different matrices with surety and certainty. The aim of this article is to provide a simple way to use approaches with a correct scientific background to improve the quality of the method development and validation. Method development involves a series of simple steps. All the conditions are optimized as required for the purpose of the separation and the method is validated using ICH guidelines.

REFERENCES

1. Skoog DA, West DM, Holler FJ. Fundamentals of analytical chemistry. 8th ed., Fort Worth: Saunders College Pub. 1996.
2. Shewiy DH, Kaale E, Risha PG, Dejaegher B, Verbeke JS, Heyden YV. *J. Pharmaceut. Biomed. Anal*, 66, 2012, 11–23.
3. 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.
4. FDA, Guidance for Industry Q1A(R2), stability testing of new drug substances and products. 2003.
5. http:// www.chem.agilent.com/cag/cabu/buffersel.html
6. http:// www. chromatography-online.org/HPLC.html
7. Zhoua S, Songb Q, Tangb Y, Weng N. Critical Review of Development, Validation, and Transfer for High Throughput Bioanalytical LCMS/MS Methods. *Current Pharmaceutical Analysis*, 1, 2005, 3-14.
8. U.S. Food and Drug Administration Guidance for Industry, ICH Q6A, Specifications: Test Procedure and Acceptance Criteria for New Drug Substances and New Drug Products: Chemical Substances, 1999.
9. Chan CC, Leo YC, Lam H. Analytical method validation and Instrument Perfomance Validation. Vol-I, Wiley Interscince, USA. 2004.
10. Jain HK, Agrawal RK. *Indian Journal of pharmaceutical sciences*, 64(1), 2002, 88-71.
11. Shankar MB, Mehta FA, Bhatt KK, Mehta RS and Geetha M. *Indian Journal of pharmaceutical sciences*, 65(2), 167-170.
12. Jenke DR. Chromatographic method validation: A review of current practices and procedures. Part II. Guidelines for primary validation parameters. *Instrum. Sci. Technol*, 26, 1998, 1-18.
13. Swarbrick J and Boylan J. Encyclopedia of pharmaceutical technology, Volume I, Marcel Dekker Inc., New York, 1998, 217 - 224.
14. ICH Q2 (R1). Validation of Analytical Procedures: Text and Methodology. International Conference on Harmonization, IFPMA, Geneva, 2005.
15. Zhoua S, Songb Q, Tangb Y, Weng N. Critical Review ofDevelopment, Validation, and Transfer for High Throughput Bioanalytical LCMS/MS Methods. *Current Pharmaceutical Analysis*, 1, 2005, 3-14.
16. Green JM. A practical guide to analytical method validation. *Anal. Chem. News & Features*, 1996, 305A–309A.
17. Rockville MD. General Tests, Chapter 621 – Chromatography System Suitability, United States Pharmacopeial Convention (USP), USP 31, 2009.
18. Shabir GA, Lough WJ, Arain SA, Bradshaw TK. Evaluation and application of best practice in analytical method validation. *J Chromatogr RT*, 30, 2007, 311-333.
19. ISO/IEC. Guide 99: International vocabulary of metrology – Basic and general concepts and associated terms (VIM). First edition, Geneva: International Organization for Standardization. 2007.
20. Causon R. Validation of chromatographic methods in biomedical analysis viewpoint and discussion. *Journal of Chromatography B*, 689, 1997, 175-180.
21. Hokanson GC. A life cycle approach to the validation of analytical methods during pharmaceutical product development, Part II: Changes and the need for additional validation, pharmaceutical product development, Part II: Changes and the need for additional validation. *Pharm.Tech.*, 1994, 92–100.
22. Wegscheider. Validation of analytical methods, in: Accreditation and quality assurance in analytical chemistry, edited by H. Guenzler, Springer Verlag, Berlin, 1996.
23. Seno S, Ohtake S and Kohno H. Analytical validation in practice at a quality control laboratory in the Japanese pharmaceutical industry. *Accred. Qual. Assur*., 2, 1997, 140–145.
24. AOAC Peer-Verified Methods Program, Manual on policies and procedures, Arlington, Va., USA, 1998. http://www.aoac.org/vmeth/PVM.pdf .
25. A report by Jay Breaux , Kevin Jones and Pierre Boulas AAI Development Services Analytical Method Development and Validation.
26. Beckett AH, Stenlake JB. Practical Pharmaceutical Chemistry, 4th ed., Part 2, CBS Publishers and distributors, 1997, 275-337.