1 Analytical Methods – An Overview

Introduction

In many paradigms of analytical chemistry, it is necessary to have reliable and identical data on recovery, detection and quantification of drugs. In the pharmaceutical research laboratories, and industries the quality control and assessment **(QC & A)** parameters are required to examine and evaluate the quality, safety and efficacy of the products or formulations manufactured and analyzed. In clinical analysis, (**QC & A)** is a vital aspect of safety and patient care, diagnosis and control of therapy of individual patient, and for biological analysis.

Quality control and assessment (**QC & A)** forms an essential part of the instruments or automated systems used for production in laboratories and industries. For an ***internal quality control*** **(IQC)** a continued observation is required for everyday calibration of instruments. ***External Quality Assessment*** **(EQA)** serves an important part of assuring laboratory compliance. To comprehend, it is necessary to understand the basic concept of quality, quality assurance and quality control to apply these parameters of check to a range of instrumental methods and the methods developed therewith.

Quality is the entirety of features and uniqueness of a product or a service that bears on its capability to assure stated or implied needs.

Quality Assurance **(QA)** is a term used to illustrate the means of ensuring the precision of the results in a laboratory. It is an internal analytical tool that is concerned with the prevention of quality problems through planned and systematic activities.

Quality Control **(QC)** is a feature of **QA** that is concerned with the activities and techniques employed to achieve and maintain the product quality.

The aim of **QC** is to ensure conformity with the quality specifications and provide economic effectiveness at relevant stages of operation of an analytical method.

The requirements which are necessary for (**QC & A)** fall within the sphere of quality assurance program and to seek out for international quality standards, laboratories must function in accordance with (**ISO / IEC 17025: 2000) (ISO / IEC 2000),** ***Good Laboratory Practices (*****GLP; OECD 1992)** or the ***Mandatory Health and Human Service Guidelines for federal workplace drug testing program*** **(DHHS 1994).**

Aims of Analytical Chemistry

The basic requirement for the study of the chemical and biological properties of a given molecule is that the analyte sample should be homogeneous.

In 1894, Wilhelm Ostwald wrote "Analytical chemistry, or the art of recognizing different substances and determining their constituents, takes a prominent position among the applications of science, since the questions which it enables us to answer arise wherever chemical processes are employed for scientific or technical purposes."

The aims of analytical chemistry encompass the following:

1. The advancement of theory and development of scientific validation of the existing analytical methods.
2. Detailing the scientific basis of new analytical methods which help them comply with the requirements of advancing science and modern production.
3. Analysis of products from natural substances, environment and industrial materials.

Selection of Method of Analysis

The selection of an analytical method is dependent upon many factors, such as: chemical properties of an analyte and its concentration, sample matrix, speed and cost of analysis, type of analysis, i.e., quantitative or qualitative and the number of samples.

A ***Qualitative Method*** gives information about the identity of a particular analyte in a sample. A ***Quantitative Method*** yields information about the relative amounts of one or more analytes present in a sample. A separation process is generally an indispensable step for both a qualitative and a quantitative process.

Criteria for Selection of a Method

- The technical model/method/instrumentation that is compatible with the properties of the sample to be analysed must be present.
- The method must meet the particular standards of precision, sensitivity and specificity.
- Operating cost must remain within reasonable economic limits
- Before starting with the method development one must have knowledge about the nature of the sample; the objectives of analysis should be defined and the number of samples must be known. The nature of the sample helps one to decide or conclude about the best method to proceed with the method development. Generally, chemical analysis is performed only on a small division of the product/sample whose composition is to be studied. But, the composition of the small fraction taken for analysis must be identical to the composition of the bulk material. The procedure of acquiring a representative sample is called 'sampling'. Some products/ samples need pre-treatment prior to analysis in order to avoid interferences or to concentrate the analytes. The aims of a separation process for example should be precise and detailed before starting with a method development. It may for example, be necessary to resolve all the constituents of a sample or it may be required that the analytes are separated from impurities and degradation products without the need to further separate these impurities and degradation products from each other. The concluding process should meet all the requirements specified in the beginning of the method and when the method for quantitative use is finalized it should be validated.

Various Methods of Analysis

Since few decades, different methods of analysis are used for quantitative analyses which include:

Spectroscopic Methods:

- UV – Visible Spectroscopic Method
- Infrared Spectroscopy
- Mass Spectroscopy
- NMR Spectroscopic method

Spectrometry Method:

- Mass Spectrometry

Chromatographic Methods: Most widely used chromatographic methods include

- High Performance Liquid Chromatography **(HPLC)**
- High Performance Thin Layer Chromatography **(HPTLC)**
- Gas Chromatography (also sometimes called as Gas Liquid Chromatography-**GLC,** vapor-phase chromatography **-VPC,** or gas–liquid partition chromatography-**GLPC)**
- GC-MS
- LC-MS

Electrochemical Methods:

- Coulometry
- Voltametry
- Potentiometry
- Atomic Spectrometry
- Emission (plasma) Spectrometry
- Conductometry
- Polarography
- Amperometry

Other Methods:

- Gravimetry
- Titrimetry

Hyphenated Techniques Adding New Life into the Analytical Instrumentation

Widespread research in the fields of biochemistry, drug discovery, environmental testing, and even space research, has increased the need for high-performance analytical equipment. Each new development offers much more than its forerunner. Traditional analytical approaches including HPLC (High-Performance Liquid Chromatography), GC (Gas Chromatograph), UV (Ultraviolet) detection, etc., have become inadequate to efficiently address the challenges in analyses of species-specificity and sensitivity. Modern analyses methods, whose developments are referred to as hyphenated techniques, commence from the traditional use of specific detection.

Hyphenated Techniques combine chromatographic and spectral methods to exploit the advantages of both. Chromatography techniques produce pure or nearly pure fractions of chemical components in a mixture. Spectroscopic techniques produce selective information for identification using standards or library spectra.

Hyphenated techniques and instruments have been around for over 15-20 years now. The GC-MS (Mass Spectrophotometer), ICP-MS (Inductive Coupled Plasma – Mass Spectrophotometer) have been finding applications in several research efforts across various fields. Stand-alone instruments could analyze various physical properties such as boiling point, toxicity, solubility and metabolic pathways in different living organisms. With integrated such the HPLC-ICP-MS or the GC-ICP-MS, sample preparation before analysis is minimized and 'on-line' species separation is rapid. These integrated techniques are becoming increasingly popular in applications where analyses of complex matrices with low detection limits and high specificity are expected.

The hyphenated techniques offer advantages of, shorter analysis time, higher degree of automation, increased sample throughput, better reproducibility, reduction of contamination due to a closed system unit and enhanced combined selectivity and higher degree of information output.

Researchers in various fields are already using in varied applications and in the coming era research will be greatly benefited by these hyphenated techniques.

The proceeding chapters would help gain insights into Chromatography, its basic principles and related hyphenated techniques which are of paramount importance in the field of analytical research worldwide.