DEVELOPMENT AND VALIDATION OF A STABILITY INDICATING HPLC ASSAY METHOD FOR SIMULTANEOUS DETERMINATION OF TRANDOLAPRIL AND VERAPAMIL HCl IN TABLET DOSAGE FORM

INTRODUCTION

Analytical Chemistry is particularly concerned with the questions of "what chemicals are present, what are their characteristics and in what quantities are they present?" Modern Analytical Chemistry is dominated by instrumental analysis. These are extremely sensitive, providing precise and detailed information from small samples of material. They are for the most part rapidly applied, and in general are readily amenable to automation. For these reasons they are now in widespread use in product development, in the control of manufacture and formulation, as a check on stability during storage, and in monitoring the use of drugs and medicines.

Traditionally, analytical chemistry has been split into two main types, qualitative and quantitative. Qualitative Inorganic Analysis seeks to establish the presence of a given element or inorganic compound in a sample. Qualitative Organic Analysis seeks to establish the presence of a given functional group or organic compound in a sample. Quantitative analysis seeks to establish the amount of a given element or compound in a sample.

There are various techniques used for analysis of mixtures. Spectroscopy measures the interaction of the molecules with electromagnetic radiation. Chromatography (from Greek “chroma” means colour and "grafein" means to write) is the collective term for a family of laboratory techniques for the separation of mixtures. It involves passing a mixture dissolved in a "mobile phase" through a stationary phase, which separates the analyte to be measured from other molecules in the mixture and allows it to be isolated. Analytical Chromatography is used to determine the existence and possibly also the concentration of analyte(s) in a sample.

HPLC is a popular method of analysis because it is easy to learn and use and is not limited by the volatility or stability of the sample compound. Modern HPLC has many applications including separation, identification, purification, and quantification of various compounds. HPLC is widely considered to be a technique mainly for
biotechnological, biomedical and biochemical research as well as for the pharmaceutical industry.

Analytical chemistry has played critical roles in the understanding of basic science to a variety of practical applications, such as biomedical applications, environmental monitoring, quality control of industrial manufacturing, forensic science and so on.

Factors Affecting the Choice of Analytical Method

Analytical techniques have different degrees of sophistication, sensitivity and selectivity, as well as, different cost and time requirements. An important task for the analyst is to select best procedure for a given determination this will require careful consideration of the following criteria:

1. The type of analysis required: elemental or molecular, routine or occasional.
2. Problem arising from the nature of the material to be investigated: e.g. radioactive substance, corrosive substance, substances affected by water.
3. Possible interference from components of the material other than those of interest.
4. The concentration range to be investigated.
5. The accuracy required.
6. The facilities available, particularly the instrument.
7. The time required to complete the analysis.
8. The number of analysis of similar type which have to be performed.

Selection of Analytical Method

First stage in the selection or development of method is to establish what is to be measured and how accurately it should measured. Unless one has series of methods at hand to assess quality of the product, validation program may have limited validity. The selected method must have the following parameters:

1. As simple as possible.
2. Most specific.
3. Most productive, economical and convenient.
4. As accurate and precise as required.
5. Multiple source of key component (reagents, columns, TLC plates) should be avoided.

6. To be fully optimized before transfer for validation of its characteristics such as accuracy, precision, sensitivity, ruggedness etc.

**Classification of Analytical Methods**

The various methods of analysis can be broadly classified into two categories.

**Classical Methods:**

a. **Volumetric Methods:** In volumetric, also called titrimetric procedures, the volume or mass of a standard reagent required to react completely with the analyte was measured.

b. **Gravimetric Methods:** In gravimetric measurements, the mass of the analyte or some compound produced from the analyte was determined.

   The extent of their general application is, however, decreasing with the passage of time and with the advent of instrumental methods to supplant them.

**Instrumental Methods:**

These methods are based upon the measurement of some physical properties as conductivity, electrode potential, light absorption or emission, mass-to-charge ratio and fluorescence of substance. There are many techniques available for the analysis of analytes.

a. **Spectroscopic Analysis**

1. Ultraviolet and visible spectrophotometry
2. Fluorescence and phosphorescence spectrophotometry
3. Atomic spectrophotometry (emission & absorption)
4. Infra-red spectrophotometry
5. Raman spectroscopy
6. X-ray spectroscopy
7. Radio chemical techniques including activation analysis
8. NMR spectroscopy
9. ESR spectroscopy

b. **Electrochemical Techniques**

1. Potentiometry
2. Voltametry
3. Stripping techniques
4. Amperometric techniques
5. Coulometry
6. Electrogravimetry
7. Conductance techniques

c. Chromatographic Methods
   1. Gas chromatography (GC)
   2. High performance liquid chromatography (HPLC)
   3. High-performance thin layer chromatography (HPTLC)

d. Miscellaneous Techniques
   1. Thermal analysis
   2. Mass spectrometry
   3. Kinetic techniques

e. Hyphenated Methods
   1. GC-MS
   2. ICP-MS
   3. GC-IR
   4. MS-MS

Amongst all the techniques mentioned above UV-Visible spectrophotometry and High Performance Liquid Chromatography (HPLC) are the most widely used techniques for quantitative analysis of pharmaceutical substances.

The HPLC system is basically composed of 1) pump 2) an injector 3) a column 4) a column oven 5) a detector 6) a recorder. The pump keeps liquid flow rate constant. Sample is injected through the injector, it is carried into the column and separated into its components. Each component elutes from the column one by one and is detected and recorded.
Advantages of HPLC:
1. It provides specific, sensitive and precise method for analysis of various complicated sample.
2. The analysis by HPLC is specific, accurate and precise.
3. It offers advantage over gas chromatography in analysis of many polar, ionic substances, high molecular weight substances, metabolic products and thermolabile as well as nonvolatile substances.
4. It is helpful in impurity profiling along with analysis of combination of drugs.

Method Development In HPLC:
In developing HPLC method for the quantitative analysis of multicomponent formulation the following general requirements should be fulfilled.

1. The identity of the component to be analyzed should be established.
2. Separation of specific components should be achieved.
3. Sample preparation should be reproducible.
4. Standard of known purity should be available and accuracy will be directly related to the degree of purity of standards used in determination.
5. A stationary phase that separates the component in reproducible manner.
6. There must be a constant flow of mobile phase.
7. Sample application or injection should be reproducible.