2. Review of literature:

2.1 General review on methods of simultaneous determination of hypertensive drugs.

Still present many analytical methods are available on traditional or classical methods and these are not assuring more accuracy for drug analysis. As well HPLC assay methods are available for single hypertensive dosage form and combined of 2 molecules dosage forms. Very less assay method available for combined of 3 molecules dosage forms. No HPLC method available for combined of <3 molecules dosage forms from literature.

The regulatory guidelines mandated the need for establishing stability-indicating assay. The practical steps for establishing the stability indicating method are elusive in the regulatory guidelines and pharmacopoeias. Hence a literature was undertaken replete with the publications on the development of methods of drug substance and drug products.

Recent progress in methods development has been largely a result of improvements in analytical instrumentation. This is especially true for chromatographs and detectors. Isocratic and gradient reverse-phase HPLC have evolved as the primary techniques for the analysis of drugs. The HPLC detector of choice for many types of methods development is the photodiode array (PDA) detector because it can be used for both quantitative and qualitative analysis. The use of a PDA detector to determine peak purity of the active ingredient in stressed samples greatly facilitates the development of stability-indicating assays.

The ultraviolet (UV) absorbance detector remains the most common HPLC detector for potency and impurity analysis. Once specificity has been demonstrated, the PDA detector is replaced with a variable wavelength detector and the HPLC effluent is monitored at fixed wavelengths. Stability-indicating and impurity methods often are required to measure analytes within a wide concentration range.

The review of literature began with factual collection of large of number of methods reported over the past decades under the nomenclature.

2.2 Literature review from Pharmacopoeias:

All country having their own regulatory body to control the quality of pharmaceutical drug substances and drug products. The pharmacopoeias with respective regulatory body publishing the analytical methods for drug substances and drug products.

From review of Indian Pharmacopoeia (IP) the analytical method published for only single dosage form and combined two molecule dosage forms. The methods are available in IP given in tabular form [Table 2.1].

<table>
<thead>
<tr>
<th>Table 2.1 – Methods available for hypertensive drugs in IP</th>
</tr>
</thead>
<tbody>
<tr>
<td>Single dosage form</td>
</tr>
<tr>
<td>Dosage form of two molecules</td>
</tr>
</tbody>
</table>

From IP review no single method available for combination form more than 2 hypertensive drugs.
From review of *United state Pharmacopoeia* (USP) the analytical method published for only single dosage form and combined two molecule dosage forms. The methods are available in USP given in tabular form [2.2].

**Table 2.2 – Methods available for hypertensive drugs in USP**

<table>
<thead>
<tr>
<th>Single dosage form</th>
<th>Amlodipine Besylate API and Tablets, Chlorthalidone API and Tablets, Hydrochlorothiazide API and Tablets, Losartan potassium API and Tablets, Telmisartan Tablets, Irbesartan API and Tablets, Valsartan API, Atorvastatin Ca API</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dosage form of two molecules</td>
<td>Irbesartan + Amlodipine Tablets, Losartan potassium + Hydrochlorothiazide Tablets, Valsartan + Hydrochlorothiazide Tablets</td>
</tr>
</tbody>
</table>

From USP review no single method available for combination form more than 2 hypertensive drugs.

From review of *British Pharmacopoeia* (BP) and *European Pharmacopoeia* (Ph. Eur) the analytical method published for only single dosage form and combined two molecule dosage forms. The methods are available in BP and Ph. Eur given in tabular form [2.3].

**Table 2.3 – Methods available for hypertensive drugs in BP and Ph. Eur.**

| Single dosage form | Hydrochlorothiazide API and Tablets, Chlorthalidone API and Tablets, Amlodipine Besylate API, Losartan potassium API, Telmisartan API, Irbesartan API, Valsartan API, |

From BP and Ph. Eur review no single method available for combination form more than 2 hypertensive drugs. Also assay method for many hypertensive drugs developed on classical method like titration method.

From review of *Japanese Pharmacopoeia* (JP), the analytical method published for only single drug is Hydrochlorothiazide API.

Hence from overall pharmacopeias published methods available for single drugs drug or combined two drugs only.

**2.3 Literature review from Journals:**

Literature studies show various analytical methods reported for the estimation of individual, binary or tertiary combination of anti-hypertensive drugs or in combination with diuretics. Recently *HPLC method* with fluorescence detection for simultaneous determination of Chlorthalidone, Valsartan and Fluvastatin from human plasma is available from journal. The column-switching liquid chromatographic system with fluorescence detection has been reported for determination of Losartan, Telmisartan and Valsartan from human urine.
While from the literature, many HPLC methods has been developed and validated for different hypertensive drugs. Some HPLC methods reported for single dosage form and it given as below.

i Determination of Irbesartan in human plasma by HPLC method$^6$.

ii Stability-indicating assay for Chlorthalidone formulation: evaluation of the USP analysis$^{10}$.

iii UPLC method for determination of Valsartan and their degradation products in active pharmaceutical ingredient$^{11}$.


v Analysis of Amlodipine in Human Plasma by Liquid Chromatography–Mass Spectrometry$^{24}$.

vi Validated RP-HPLC method for the estimation of Telmisartan in serum samples$^{28}$.

vii Validated Spectrofluorometric methods for determination of Amlodipine Besylate in Tablets$^{29}$.

viii Quantitative determination of Chlorthalidone in pharmaceutical dosage forms by high-pressure liquid chromatography$^{50}$.

ix High-performance liquid chromatographic analysis of angiotensin II receptor antagonist Valsartan using a liquid extraction method$^{64}$.

x Development and validation of HPLC method for the estimation of Irbesartan in pharmaceutical dosage form$^{80}$.

xi Stability-indicating RP-HPLC method for analysis of Telmisartan in the bulk drug and in formulations$^{92}$.

xii Stability-indicating high performance liquid chromatographic determination of Atorvastatin calcium in pharmaceutical dosage form$^{102}$.

From literature HPLC methods has been developed and validated for combined dosage form also. Reported analytical methods given as below by HPLC.

i Simultaneous Estimation of Olmesartan Medoxomil and Hydrochlorothiazide by RP-HPLC Method from Combined Dosage Form$^1$.

ii Simultaneous Determination of Hydrochlorothiazide (HCT) and Losartan Potassium (LOS) from Tablets$^2$.

iii Simultaneous determination of Atenolol and Amlodipine in Tablets$^3$.

iv Simultaneous Determination of Candesartan Cilexetil and Hydrochlorothiazide$^4$.

v RP-HPLC determination of Atorvastatin calcium and Amlodipine Besylate combination in tablets$^6$.

vi Simultaneous Determination of Amiloride Hydrochloride, Atenolol, Hydrochlorothiazide, and Chlorthalidone in their Combined Mixtures$^{20}$.

vii Simultaneous quantitation of Hydrochlorothiazide and Metoprolol in human by liquid chromatography-tandem mass spectrometry$^{22}$.

viii Simultaneous Determination of Valsartan and Hydrochlorothiazide in Tablets by High-Performance Liquid Chromatography$^{27}$.
ix RP-HPLC method for simultaneous estimation of Bisoprolol Fumarate and Hydrochlorothiazide in tablet formulation$^{34}$.

x Development and Validation of New Method for Atenolol, Hydrochlorothiazide and Losartan potassium by RP-HPLC$^{40}$.

xi RP-HPLC method for the determination of Losartan potassium and Ramipril in combined dosage form$^{41}$.

xii RP-HPLC method for the simultaneous determination of Atorvastatin and Amlodipine in tablet dosage form$^{42}$.

xiii Determination of Losartan potassium, Ramipril and Hydrochlorothiazide in Pharmaceutical preparation$^{52}$.


xv Simultaneous Analysis of Candesartan Cilexetil and Hydrochlorothiazide in Human Plasma and Dosage Forms Using HPLC with a Photodiode Array Detector$^{58}$.

xvi Analysis of binary mixtures of Losartan potassium and hydrochlorothiazide by using high performance liquid chromatography, ratio derivative Spectrophotometric and compensation technique$^{60}$.

xvii Simultaneous HPLC Analysis of Olmesartan and Hydrochlorothiazide in Combined Tablets and in vitro Dissolution Studies$^{63}$.

xviii Simultaneous Estimation of Losartan potassium and Amlodipine Besylate in Tablet formulation$^{67}$.

xix Simultaneous estimation of Atorvastatin Calcium and Amlodipine Besylate from Tablets$^{69}$.

xx RP-HPLC method for Simultaneous estimation of Losartan Potassium and Atenolol$^{72}$.

xxi Simultaneous high-performance liquid chromatographic determination of Telmisartan and Hydrochlorothiazide in pharmaceutical preparation$^{76}$.

xxii Rapid simultaneous determination of Telmisartan, Amlodipine Besylate and Hydrochlorothiazide in a combined poly pill dosage form by stability indicating Ultra performance liquid chromatography$^{85}$.


xxiv Determination of Atenolol Combinations with Hydrochlorothiazide and Chlorthalidone in Tablet Formulations by Reverse-Phase HPLC$^{89}$.

xxv RP-HPLC method for simultaneous estimation of Telmisartan and Hydrochlorothiazide in tablet dosage$^{101}$.

**LC-MS** analytical method has been reported for identification and characterization of photolytic degradation of Telmisartan$^{78}$. Quantification of Irbesartan and major proteins in human plasma given by Mass spectrophotometer with time of flight analyzer$^{14}$. The LC-MS method is more applicable in determination drugs in plasma and in identification of structure of drugs whereas it is costly than HPLC.

**UPLC analytic method** has been reported for determination of Valsartan and their degradation products$^{11}$. The UPLC method is faster but it is high costly than HPLC.
Spectrophotometric method given for simultaneously determination of Olmesartan Medoxomil and Amlodipine Besylate from tablet dosage form. Development and validation of simultaneous estimation of Atorvastatin Calcium and Ramipril from Capsule dosage form given by first order derivative spectroscopy. Many UV-Spectrophotometric methods reported for single drug determination from dosage form. The Spectrophotometric method is less accurate than HPLC and it is not stability indication method. As well it cannot apply for matrix containing a more no of drugs for determination due to absorbance interference.

Many HPTLC methods available for simultaneous determination of hypertensive simultaneous estimation of Irbesartan and Hydrochlorothiazide, Telmisartan and Ramipril and has been reported and Telmisartan and Metoprolol Succinate in Pharmaceutical Formulation has been reported in journals. The HPTLC is less reliable and less accurate than HPLC.

Capillary Zone Electrophoresis method already developed for simultaneously determination of Losartan with Chlorthalidone or Hydrochlorothiazide from literature. S. Hillaert, W Van Den Bossche have been developed the method by capillary electrophoresis for simultaneous determination of hydrochlorothiazide and several angiotensin-II-receptor antagonists (Candesartan, Eprosartan mesylate, Irbesartan, Losartan potassium, Telmisartan and Valsartan).

On a details review of various monographs, book chapter, and research articles, it is observed that most of the reported methods included Titrimetry, spectrometric and chromatographic techniques developed only for single or less than three combined drugs. If objective of analysis is determination more number of drugs in matrix then Titrimetry and spectrometric method is inconvenient and less reliable.

The nature of requirement of separation of multiple components during analysis of samples drove chromatographic methods to take precedence over the conventional methods of analysis. HPTLC, HPLC and GC are separation method and use to determine the drugs in sample. The HPTLC is reliable and fast. But due to less accuracy and resolution capacity not popular to use for determination of more drugs in samples. The GC is better method, but not versatile technique, as drug substances may be non volatile or thermally unstable. Hence GC methods rarely use in assay analysis, where the option is not available on HPLC.

In comparison the HPLC has been widely employed. This technique has gained popularity in regular analysis and stability studies due to its high resolution capacity, sensitivity and specificity. Non-volatile, thermally unstable or polar, ionic compounds can also be analyzed by this technique. Therefore HPLC will be use to develop the methods for more combined drugs and use to stability study due high resolution capacity.

The parent guideline on drug stability testing Q1A (R2) issued by International Conference on Harmonization (ICH) stipulates stress studies to be carried out on a drug in order to establish the drug’s inherent stability characteristics. The parent guideline on validation of analytical procedure Q2A (R1) issued by International Conference on Harmonization (ICH) guideline provided the guidance for method validation procedure to prove the method is qualify and fit for use to regular analysis. On base of validation characteristics complies the method will be suitable for regular uses.
However, so far, no method is reported for the simultaneous determination in combination for Hydrochlorothiazide, Chlorthalidone, Amlodipine Besylate, Valsartan, Telmisartan, and its application to pharmaceutical samples. An attempt is made in this study to develop a rapid, economical, precise and accurate stability-indicating assay method for simultaneous estimation of Hydrochlorothiazide, Chlorthalidone, Amlodipine Besylate, Valsartan, and Telmisartan in tablet formulation.

As well, no method is reported for the simultaneous determination in combination for Atorvastatin Calcium, Olmesartan Medoxomil, Candesartan, Hydrochlorothiazide, Chlorthalidone and its application to pharmaceutical samples. An attempt is made in this study to develop a rapid, economical, precise and accurate stability-indicating assay method for simultaneous estimation of Atorvastatin Calcium, Olmesartan Medoxomil, Candesartan, Hydrochlorothiazide and Chlorthalidone in tablet formulation.

Also, no method is reported for the simultaneous determination in combination for Irbesartan, Losartan, Hydrochlorothiazide, Chlorthalidone and its application to pharmaceutical samples. An attempt is made in this study to develop a rapid, economical, precise and accurate stability-indicating assay method for simultaneous estimation of Irbesartan, Losartan, Hydrochlorothiazide and Chlorthalidone in tablet formulation.

All proposed methods will be rapid, simple, accurate, and reproducible. The methods will be successfully employed in the routine analysis of both drug substance and drug products.