Research Inventy: International Journal of Engineering And Science
Vol.11, Issue 4 (April 2021), PP 29-35
Issn (e): 2278-4721, Issn (p):2319-6483, www.researchinventy.com

A Review: Technique and Recent Treades for Simultaneous Estimation of Sacubitril/Valsartan in Combination of Pharmaceutical Dosage Forms.

Harshada Raosaheb Rokade[1*], Sagar Anil Daitkar, [2] Neha Sanjay Ahire [3], Sopan Subhash Jadhav[4].

[1*]*Department of Pharmaceutics, R.C. Patel Institute of Pharmaceutical Education and Research, Shirpur – 425 405, Maharashtra, India*
[2] *Department of Industrial Pharmacy, Loknete Dr. J. D. Pawar College of Pharmacy, Manur, - 423501, Maharashtra, India*
[3] *Department of Industrial Pharmacy, Loknete Dr. J. D. Pawar College of Pharmacy, Manur, - 423501, Maharashtra, India*
[4] *Department of Pharmaceutics, S.N.D College of Pharmacy, Babhulgaon – 423 401, Maharashtra, India.*
[*]***For Correspondence author:*** *Harshada Raosaheb Rokade[1], Department of Pharmaceutics, R.C. Patel Institute of Pharmaceutical Education and Research, Shirpur – 425 405, Maharashtra, India*

ABSTRACT:
The Present work was to develop a simple, fast, accurate, precise, reproducible, Reverse Phase High Performance Liquid Chromatographic Method for simultaneous estimation of Valsartan and Sacubitril in pure drug form. Valsartan passes acidic tetrazole systemic which acid group play important role bind to angiotensin II receptor (AT1 receptor subtype) and acts as antagonist. Therefore, it also known as angiotensin II Blocker. Sacubitril is prodrug metabolized by de-ethylation and gives active compound as Sacubitril at (ACE Inhibitors) inhibit the neurolysin enzyme necessary for lowering blood pressure. Valsartan/Sacubitril at are potent antihypertensive decrease risk of cardiovascular & hospitalization in patient with heart failure. These are several methods to develop simultaneous estimation of valsartan for stabile indicating RP-HPLC, HPTLC, Selective LC, Isocratic system, Classical least square, Principal component regression, chromatographic separation achieved by changing different mobile phase. Regard to improvement of effectiveness and stability has been developed simple, accurate, Precise, highly sensitive, rapidly, economical selective UV Spectrophotometric method for simultaneous determination of Valsartan in combination of pharmaceutical dosage form validate by ICH guideline.

KEYWORDS: *Sacubitril, valsartan, UV-spectrophotometric, ICH guideline.*

Date of Submission: 27-05-2021 Date of Acceptance: 09-06-2021

I. INTRODUCTION:

Sacubitril is chemically 4- [[(2S, 4R)-5-ethoxy-4-methyl-5-oxo-1-(4-phenyl-phenyl) pentan-2-yl] amino]-4-oxobutanoic acid.[1, 2, 3] Sacubitril is prodrug undergo de-ethylation by esterase enzyme to give Sacubitrilat which inhibite the neprilysin enzyme (Degradation of atrial and brain natriuretic peptide) used as antihypertensive drug used in combination with valsartan (Angiotensin Receptor Blocker) for the treatment of heart failure.[4, 5]

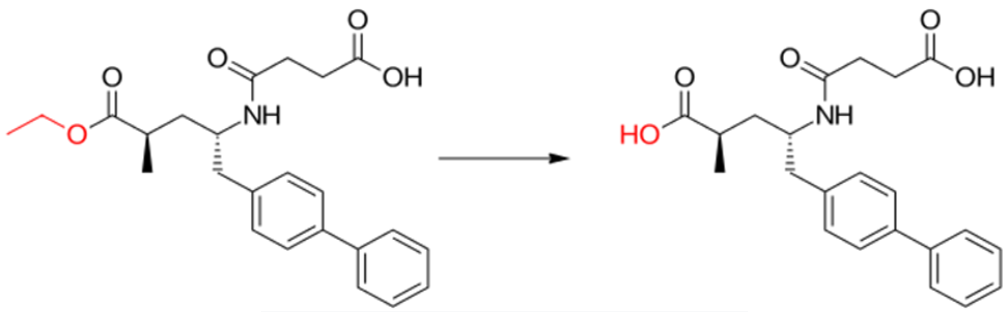

Figure 1: Sacubitril activation to sacubitrilat.

In addition, neprilysin degrade a variety of peptide including bradykinin and inflammatory mediator exerting potent vasodilatory effect.[6, 7]

Valsartan (Diovan) is nonpeptide, orally active and specific angiotensin II receptor blocker acting on the AT1 receptor subtype.

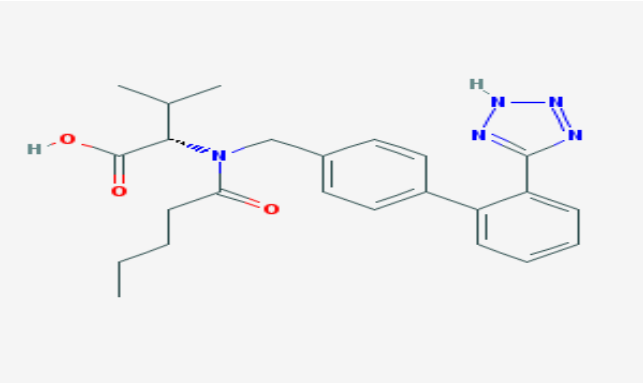

Figure 2: Structure of Valsartan

Valsartan is chemically N-(1-oxopentyl)-N-[[2'(1Htetrazol-5yl) [1, 1'-biphenyl]-4-yl] methyl]-Lvaline.[8, 9, 10]

Methods such as RP-HPLC, HPTLC, selective LC-MS, protein precipitation, Classical leas square, Principal regression, Isocratic HPLC and simultaneous Spectrophotometric methods are reported for estimation of valsartan are in combination with other agents one of the such as sacubitril.[11, 12, 13] A literature search reveals that only two analytical methods were reported for simultaneous estimation of sacubitril and valsartan from synthetic mixture using RP-HPLC and UV-spectrophotometric method. There is no stability indicating analytical methods was reported for simultaneous estimation of sacubitril/valsartan combination (LCZ696).[14,15,16] Hence simple, rapid, sensitive and selective, economic, accurate precise stability indicating UV-spectrophotometric and RP-HPLC method was developed for the simultaneous estimation of sacubitril and valsartan from API and pharmaceutical dosage form validated to ICH guideline. Entresto (sacubitril/valsartan) have been approved for decrease risk of cardiovascular death as well as hospitalization for heart failure in patient also reduced left ventricle ejection fraction.[17, 18.19]

Mechanism of Action:

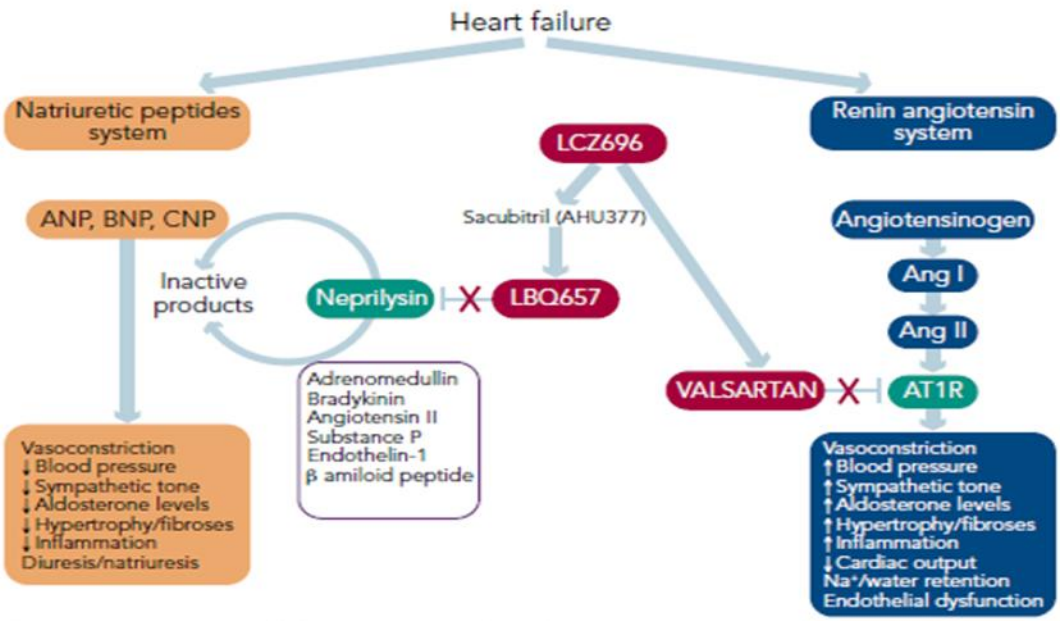

Figure 3: Role of natriuretic peptides in Heart Failure

The Role of the Renin-Angiotensin-Aldosterone System in Heart Failure Neurohumoral activation of the renin- angiotensin aldosterone system (RAAS) and nervous system play important role in the development and progression of Heart Failure. The RAAS is most essential for the regulation of cardiovascular homeostasis that exerts its action through the renin-angiotensin system. The sympathetic system is play important role in Heart Failure. These are necessary for to regulate blood pressure and maintain water and sodium balance. Abnormalities in cardiac function in HF activate the RAAS and sympathetic nervous system in order to maintain perfusion of vital organs. However, prolonged activation of RASS system increases systemic vascular resistance and causes sodium and water retention, myocardial hypertrophy, fibrosis and apoptosis, which accelerates the progression of HF and promotes end-organ damage. The blockage of beta-adrenergic receptors leads to symptomatic improvement and reduced morbidity and mortality in patients with HFrEF. In addition, the central role of the RAAS system in HF has led to the therapeutic use of RAAS inhibitors including angiotensin-converting enzyme (ACE) inhibitors,14angiotensin receptor blockers (ARBs) in patients who cannot tolerate ACE inhibitors15 and mineralocorticoid receptor antagonists in the treatment of chronic HF.[16,17] ARBs competitively inhibit the binding of angiotensin II to its AT1 receptors located on blood vessels and other tissues, and improve symptoms, haemodynamics and outcomes in chronic HF. These beneficial effects are attributed to the inhibition of the deleterious effects of AT1 receptor stimulation, i.e., vasoconstriction, Na+ and water retention, aldosterone and vasopressin release, stimulation of sympathetic tone, inflammation, and fibrosis and cell growth. However, ace inhibitors, ARBS, aldosterone receptor antagonists and combinations of drugs in these classes are limited in their ability to fully inhibit the activity of the RAAS. Furthermore, ace inhibitors and ARBS induce a reactive rise in plasma renin activity that may eventually surpass their RAAS-inhibitory effect, and plasma aldosterone levels remain elevated in a subset of patients despite therapy, a phenomenon known as aldosterone escape or aldosterone breakthrough in addition, ARBS do not enhance bradykinin-mediated vasodilation and are considered less effective than ace inhibitors.[20,21,22,23]

Ideal properties of solvents:

- It should not interact with the solute.
- It should not significant absorption.
- It should be good solvent.
- It should be stable at various temperature.[24,25]

Analytical Method:

- UV-spectrophotometric method
- Chromatographic method; RP- HPLC method
- HPTLC
- Selective LC-MS
- Classical least square
- Principal component regression.[26,27]

Chromatographic method:

Type of HPLC Techniques:

- **Based on polarity of stationary phase and mobile phase:**

1. Normal phase mode:

Stationary phase is polar e.g. Silica gel and mobile phase is non polar. In Normal phase mode, nonpolar compound travel faster and eluted first. This is because of more affinity towards mobile phase and less affinity towards stationary phase. Polar compound is retained longer time in the column because more affinity towards stationary phase and take more time to elute.

2. Reverse phase mode:

Stationary phase is Non polar and mobile phase is polar. In Reverse phase mode, polar compound gets eluted first and non-polar compound are retained for longer time. Since most of the drugs and pharmaceuticals are polar in nature and not retained for longer time and eluted faster.

- **Based on separation principle:**

1. Adsorption chromatography:

Separation of compound based on the difference in affinity of compound towards stationary phase. Most widely usede stationary phase is unmodified silica which having high efficiency and high-resolution power. The silanol group are responsible for adsorbtion which react whith solute and to form Hydrogen bon

2. Ion exchange chromatography:

Resin is used for separation of mixture of compound having charge ions. Compound based on exchange of ion of functional group. The retention of ion on the basis of column packing material and ionic strength and pH of the mobile phase.[28, 29]

- **Type of exchanger**

a. Polystyrene resins: The allow cross linkage which increase stability of chain.

b. Controlled: Pore glass: Binding of ions of higher charge and smaller radius.

c. Cellulose and dextran ion porous silica: Process involved larger pore size and low charge densities making them suitable for protein separation.[30,31,32]

3. Ion pair chromatography:

In that revers phase, column is converted temporarily into ion exchange column using compound such as pentane or hexane or heptane with sulphonic acid sodium salt.

4. Chiral phase chromatography:

Separation is based on optical isomer chairal stationary phases.

5. Affinity chromatography:

Separation depends on affinity of the sample towards the stationary phase.

- **Based on elution techniques:**

1. Isocratic separation: Same mobile phase combination is used through the process of separation.

2. Gradient separation: Mobile phase combination of lower elution strength is used by gradually increasing the elution.[33,34]

- **Based on operation:**

a. Analytical HPLC

Example: Analysis of sample (microgram).

b. Preparative HPLC

Example: Individual fraction of sample are analyte (milligram).

- **Based on type of analysis**:

1. Qualitative analysis:

Example: Identification of compound, detection of impurities.

2. Quantitative analysis:

Example: Quantity of sample.

Simultaneous estimation of Sacubitril and Valsartan by using UV- Visible Spectrophotometer:

On survey, it was found that the only RP-HPLC methods was found that estimation of the Sacubitril and Valsartan are combined dosage forms using simultaneous equation method up to no any one method is available in the pharmacopoeias. Few analytical methods have been developed for the determination of Sacubitril and Valsartan individually, and in combination with other drugs. So, we have developed a novel, simple and highly sensitive UV spectrophotometric methods for assay of Sacubitril and valsartan in bulk and pharmaceutical formulations and validated according to ICH guidelines.[35, 36]

Validated Eco-Friendly Chromatographic Methods for Simultaneous Determination of Sacubitril and Valsartan:

HPLC and HPTLC are used for routine analytical separation techniques that are applied for qualitative and quantitative purposes but unfortunately, majority of these conventional techniques produce dangerous organic solvents that harmfully affect humans and the surrounding environment. So, the development of greener methods became favorable which is accomplished by developing analytical methods in a safe way that decreases or disposes of unwanted toxic substances that are produced by the method to be safer to nature. However, the greater part of the applied chromatographic methods doesn't concern about the green viewpoints, some modern analytical methods presented examples of the application of an ecofriendly approach that was developed for separation of fingerprinting complex matrices.[37]

Cost-Effectiveness of Sacubitril-Valsartan Combination Therapy:

In 1980s and early 2000s, the development and use of angiotensin converting enzyme inhibitors (ACEIs) also angiotensin II receptor blockers (ARBs), beta-blockers (BBs), and aldosterone receptor antagonists have decreased disease state and death rate and to improve the life of most of patients with heart failure (HF) and a decrease ejection fraction (HFrEF). The most recent pharmacotherapy to demonstrate a mortality benefit in HFrEF is the dual-acting angiotensin receptor neprilysin inhibitor (ARNI) sacubitril valsartan. In 2015 July, sacubitril-valsartan was approved by the U.S. Food and Drug Administration for use in patients with New York Heart Association (NYHA) functional class II to IV HFrEF based on the results of the PARADIGM-HF (Prospective Comparison of ARNI with ACEI to Determine Impact on Global Mortality and Morbidity in Heart Failure) trial. Sacubitril-valsartan treatment resulted in 20% reduction in the primary outcome of a composite of death of cardiovascular causes or hospitalization for HF. The approval of sacubitril-valsartan marked the first new medication with a demonstrated mortality benefit in HFrEF in more than 10 years. Because the current cornerstone of HFrEF pharmacotherapy revolves around low-cost generic medications such as ACEIs and BBs, it is unclear how the cost of sacubitril-valsartan ($4,560 per year) will effect on its clinical utility. One of several considerations in medical decision making is value. Cost-effectiveness analysis (CEA) is one approach to determining medication value by quantifying the benefits and costs of different treatment options. With the goal

of aiding decision making, we estimated the incremental costs and cost-effectiveness of sacubitril-valsartan relative to ACEIs for the treatment of HFrEF.[38]

Effect of food on the oral bioavailability of the angiotensin receptor-neprilysin inhibitor sacubitril/valsartan (LCZ696) in healthy subjects:

Sacubitril/valsartan (LCZ696) has been approved to reduce the risk of cardiovascular death and hospitalization for heart failure in patients with HF and reduced left ventricular ejection fraction. LCZ696 is a salt complex containing neprilysin inhibitor prodrug sacubitril and angiotensin receptor blocker valsartan in 1: 1 molar ratio. Chemically, sacubitril is (2R, 4S)-5-biphenyl4-yl-4-(3-carboxy-propionylamino)-2-methyl-pentanoic acid ethyl ester, and valsartan is (2S)-3-methyl-2-[pentanoyl-[[4-[2-(2Htetrazol-5-yl) phenyl] phenyl] methyl] amino] butanoic acid. LCZ696 provides simultaneous inhibition of neprilysin and blocking of the angiotensin II type-1 receptor, resulting in complementary effects on the CV and renal system that are considered beneficial in patients with HF. LCZ696 is highly soluble in water (>100 mg/mL). While sacubitril has moderate to high permeability (>50×10–5 cm/min in Caco-2 cells) across the intestinal membrane, valsartan is poorly permeable, potentially limiting its bioavailability. Upon oral administration, LCZ696 delivers systemic exposure to sacubitril and valsartan. Sacubitril is rapidly hydrolyzed by esterase action to sacubitrilat, the active neprilysin inhibitor. The peak plasma concentrations of sacubitril, sacubitrilat, and valsartan are reached within 0.5-2.0 hours of oral administration. Sacubitrilat is not metabolized further and is eliminated unchanged.[39, 40]

II. CONCLUSION:

The number of methods developed for Simultaneous estimation of Sacubitrile and Valsartan such as Spectrophotometry, Chromatographic method, Classical least square, principal component regression. These methods required more time for analysis, High cost and not give accuracy & precision in result. The new developed method RP-HPLC Chromatographic method for Simultaneous estimation of Sacubitrile and Valsartan was found to be simple, precise, accurate and high resolution and shorter retention time therefore more acceptable and cost effective also eco-friendly it should be applied for routine analysis in research institute and quality control department.

Abbreviations:

ACE Inhibitors	:	Angiotensin-converting enzyme
RP-HPLC	:	Reverse Phase High Liquid Chromatography
HPTLC	:	High Performance Thin Layer Chromatography
LC	:	Liquid Chromatography
UV	:	Ultra-Visible Spectrophotometry
ICH	:	International Council for Harmonization
LC-MS	:	Liquid Chromatography-Mass Spectrometry
LCZ696	:	(sacubitril/valsartan) is the first approved agent of a novel class of agents combining a neprilysin inhibitor and an ARB
RAAS	:	Renin Angiotensin-Aldosterone System
API	:	Active Pharmaceutical Ingredients
HF-REF	:	Heart Failure with Reduced Ejection Fraction
ARBs	:	Angiotensin Receptor Blockers
ARNI	:	Angiotensin Receptor Neprilysin Inhibitor
BBs	:	Beta-Blockers
NYHA	:	New York Heart Association
PARADIGM-HF	:	Prospective Comparison of ARNI With ACEI To Determine Impact On Global Mortality and Morbidity in Heart Failure.
CEA	:	Cost-Effectiveness Analysis

REFERENCES:

[1]. Murugan S, Vetrichelvan T. Absorbance Ratio and First Order Derivative Spectroscopic Methods for Simultaneous Determination of Sacubitril and Valsartan in Bulk and Tablet Dosage Form. Research Journal of Pharmacy and Technology. 2019 Nov 30; 12(11):5251-4.

[2]. Moussa BA, Hashem HM, Mahrouse MA, Tarek Mahmoud S. A validated RP- HPLC method for the evaluation of the influence of grapefruit juice on liver S9 activation of sacubitril. Biomedical Chromatography. 2019 Aug; 33(8): e4553.

[3]. De Vecchis R, Ariano C, Di Biase G, Noutsias M. Sacubitril/valsartan for heart failure with reduced left ventricular ejection fraction. Herz. 2019 Aug; 44(5):425-32.

[4]. De Vecchis, Renato, and Carmelina Ariano. "Sacubitril-Valsartan for Heart Failure with Reduced Left Ventricular Ejection Fraction: A Retrospective Study." (2017).

[5]. Maurer M, Bader M, Bas M, Bossi F, Cicardi M, Cugno M, Howarth P, Kaplan A, Kojda G, Leeb- Lundberg F, Lötvall J. New topics in bradykinin research. Allergy. 2011 Nov; 66(11):1397-406.

[6]. Arendse, Lauren B., AH Jan Danser, Marko Poglitsch, Rhian M. Touyz, John C. Burnett, Catherine Llorens-Cortes, Mario R. Ehlers, and Edward D. Sturrock. "Novel therapeutic approaches targeting the renin-angiotensin system and associated peptides in hypertension and heart failure." Pharmacological reviews 71, no. 4 (2019): 539-570.

[7]. Sharma, J. N., & Al-Sherif, G. J. (2006). Pharmacologic targets and prototype therapeutics in the kallikrein-kinin system: bradykinin receptor agonists or antagonists. The Scientific World JOURNAL, 6, 1247-1261.

[8]. Meyers KE, Behar B. Valsartan use in pediatric hypertension. Future cardiology. 2011 Mar; 7(2):151-9.

[9]. Tajane TS, Code QR. Estimation of Valsartan in Pharmaceutical Formulation by Area under Curve Spectrophotometric Method.

[10]. Karanam SR, Swarupa V. Stability Indicating Analytical Method Development and Validation for Simultaneous Estimation of Valsartan and Hydrochlorothiazide in Tablet Dosage Form. Asian Journal of Pharmaceutical Analysis. 2016; 6(1):7-14.

[11]. Alaguraj, K. "Development and Validation of RP-HPLC Method and Uv–Spectrophotometric Simultaneous Equation Method of Aceclofenac and Diacerein in Combined Dosage form, Development and Validation of RP-HPLC Method and Uv-Spectrophotometric Simultaneous Equation Method of Mosapride and Pantoprazole in Combined Dosage Form. Development and Validation of RP-HPLC Method and Uv-Specrophotometric Simultaneous Equation Method of Mosapride and Pantoprazole in Combined Dosage Form." PhD diss., Madurai Medical College, Madurai, 2009.

[12]. Kalaimagal A. Quantification of Valsartan by Uv-Visible and NMR Spectroscopy (Doctoral dissertation, College of Pharmacy, Madras Medical College, Chennai).

[13]. Sathishkumar, S. "Development and Validation of Analytical Methods for the Simultaneous Estimation of Atorvastatin Calcium, Aspirin, Ramipril and Metoprolol Tartrate in Bulk and in Pharmaceutical Dosage form by UV Spectroscopy and HPTLC." PhD diss., Adhiparasakthi College of Pharmacy, Melmaruvathur, 2012.

[14]. Naazneen S, Sridevi A. Development of assay method and forced degradation study of valsartan and sacubitril by RP-HPLC in tablet formulation. Int J App Pharm. 2017; 9(1):9-15.

[15]. Eissa MS, Abou Al Alamein AM. Innovative spectrophotometric methods for simultaneous estimation of the novel two-drug combination: sacubitril/valsartan through two manipulation approaches and a comparative statistical study. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy. 2018 Mar 15; 193:365-74.

[16]. Ragab MA, Galal SM, Korany MA, Ahmed AR. First derivative emission spectrofluorimetric method for the determination of LCZ696, a newly approved FDA supramolecular complex of valsartan and sacubitril in tablets. Luminescence. 2017 Dec; 32(8):1417-25.

[17]. King JB, Shah RU, Bress AP, Nelson RE, Bellows BK. Cost-effectiveness of sacubitril-valsartan combination therapy compared with enalapril for the treatment of heart failure with reduced ejection fraction. JACC: Heart Failure. 2016 May;4(5):392-402.

[18]. Gaziano TA, Fonarow GC, Claggett B, Chan WW, Deschaseaux-Voinet C, Turner SJ, Rouleau JL, Zile MR, McMurray JJ, Solomon SD. Cost-effectiveness analysis of sacubitril/valsartan vs enalapril in patients with heart failure and reduced ejection fraction. JAMA cardiology. 2016 Sep 1;1(6):666-72.

[19]. McCormack PL. Sacubitril/valsartan: a review in chronic heart failure with reduced ejection fraction. Drugs. 2016 Mar 1;76(3):387-96.

[20]. Iborra-Egea O, Gálvez-Montón C, Roura S, Perea-Gil I, Prat-Vidal C, Soler-Botija C, Bayes-Genis A. Mechanisms of action of sacubitril/valsartan on cardiac remodeling: a systems biology approach. NPJ systems biology and applications. 2017 Apr 18;3(1):1-9.

[21]. Singh, Jagdeep SS, Louise M. Burrell, Myriam Cherif, Iain B. Squire, Andrew L. Clark, and Chim C. Lang. "Sacubitril/valsartan: beyond natriuretic peptides." Heart 103, no. 20 (2017): 1569-1577.

[22]. Sarrias A, Bayes-Genis A. Is sacubitril/valsartan (also) an antiarrhythmic drug?. Circulation. 2018 Aug 7;138(6):551-3.

[23]. Gori, Mauro, Emilia D'Elia, and Michele Senni. "Sacubitril/valsartan therapeutic strategy in HFpEF: clinical insights and perspectives." International journal of cardiology 281 (2019): 158-165.

[24]. Mennucci B, Cances E, Tomasi J. Evaluation of solvent effects in isotropic and anisotropic dielectrics and in ionic solutions with a unified integral equation method: theoretical bases, computational implementation, and numerical applications. The Journal of Physical Chemistry B. 1997 Dec 4;101(49):10506-17.

[25]. Poole CF. Chromatographic and spectroscopic methods for the determination of solvent properties of room temperature ionic liquids. Journal of chromatography A. 2004 May 28; 1037(1-2):49-82.

[26]. Swartz ME, Krull IS, editors. Analytical method development and validation. CRC press; 2018 Oct 3.

[27]. Perez-Lozano P, Garcıa-Montoya E, Orriols A, Minarro M, Tico JR, Sune-Negre JM. Development and validation of a new HPLC analytical method for the determination of alprazolam in tablets. Journal of pharmaceutical and biomedical analysis. 2004 Mar 10; 34(5):979-87.

[28]. Mitulovic. G, Mechtler K. HPLC techniques for proteomics analysis—a short overview of latest developments. Briefings in Functional Genomics. 2006 Dec 1; 5(4):249-60.

[29]. Pineiro C, Sotelo CG, Medina I, Gallardo JM, Perez-Martín RI. Reversed-phase HPLC as a method for the identification of gadoid fish species. Zeitschrift fur Leben smittel untersuchung und-Forschung A. 1997 Jun; 204(6):411-6.

[30]. Haddad PR, Jackson PE. Ion chromatography. Elsevier; 1990 Jul 16.

[31]. Dorfner K. 1.1 Introduction to Ion Exchange and Ion Exchangers. In Ion Exchangers 2011 Jun 15 (pp. 7-188). De Gruyter.

[32]. Kumar SD. An overview of Ion Exchange: Classical to the Current Status. .21.

[33]. Jandera P, Churacek J. Gradient elution in column liquid chromatography: theory and practice. Elsevier; 1985 May 1.

[34]. Jandera P, Cesla P, Hajek T, Vohralík G, Vynuchalova K, Fischer J. Optimization of separation in two-dimensional high-performance liquid chromatography by adjusting phase system selectivity and using programmed elution techniques. Journal of Chromatography A. 2008 May 2; 1189(1-2):207-20.

[35]. Vaka S, Parthiban P. New method development and validation for the simultaneous estimation of sacubitril and valsartan in a bulk and pharmaceutical dosage forms. Int J Res. 2017;4(1):17-24.

[36]. Eissa MS, Abou Al Alamein AM. Innovative spectrophotometric methods for simultaneous estimation of the novel two-drug combination: sacubitril/valsartan through two manipulation approaches and a comparative statistical study. Spectrochimica Acta Part A: Molecular and Biomolecular Spectroscopy. 2018 Mar 15; 193:365-74.

[37]. Abou Al Alamein AM. Validated eco-friendly chromatographic methods for simultaneous determination of sacubitril and valsartan in spiked human plasma and in pharmaceutical formulation. JAPS. 2018 Feb;8(2):011-7.

[38]. King JB, Shah RU, Bress AP, Nelson RE, Bellows BK. Cost-effectiveness of sacubitril-valsartan combination therapy compared with enalapril for the treatment of heart failure with reduced ejection fraction. JACC: Heart Failure. 2016 May;4(5):392-402.

[39]. Ayala somayajula S, Langenickel T, Chandra P, Wolfson E, Albrecht D, Zhou W, Pal P, Rajman I, Sunkara G. Effect of food on the oral bioavailability of the angiotensin receptor neprilysin inhibitor sacubitril/valsartan (LCZ696) in healthy subjects. International journal of clinical pharmacology and therapeutics. 2016 Dec 1;54(12):1012.

[40]. Flarakos J, Du Y, Bedman T, Al-Share Q, Jordaan P, Chandra P, Albrecht D, Wang L, Gu H, Einolf HJ, Huskey SE. Disposition and metabolism of [14C] Sacubitril/Valsartan (formerly LCZ696) an angiotensin receptor neprilysin inhibitor, in healthy subjects. Xenobiotica. 2016 Nov 1; 46(11):986-1000.

Harshada Raosaheb Rokade, et. al " A Review: Technique And Recent Treades For Simultaneous Estimation Of Sacubitril/Valsartan In Combination Of Pharmaceutical Dosage Forms." *International Journal of Engineering and Science*, vol. 11, no. 4, 2021, pp. 29-35.