OPEN ACCESS
Eurasian Journal of Analytical Chemistry
ISSN: 1306-3057
2017 12(4):337-364
DOI 10.12973/ejac.2017.00174a

A Concise Review on Analytical Profile of Valsartan

Atul A. Shirkhedkar
R. C. Patel Institute of Pharmaceutical Education and Research, INDIA
Suraj R. Chaudhari
R. C. Patel Institute of Pharmaceutical Education and Research, INDIA
Amod S. Patil
R. C. Patel Institute of Pharmaceutical Education and Research, INDIA
Sanjay J. Surana
R. C. Patel Institute of Pharmaceutical Education and Research, INDIA

Received 18 August 2016 ▪ Revised 26 September 2016 ▪ Accepted 26 September 2016

ABSTRACT
Valsartan (VAL) is an orally active angiotensin-II receptor type-I antagonist. VAL is available alone in dosage form as well as multicomponent formulation with various antihypertensive drugs like nifedipine, hydrochlorothiazide, ramipril, amlodipine and nebivolol hydrochloride, for the management of hypertension. The present investigation assesses the various approaches for analysis of VAL in bulk drug as well as formulated products. A concise review represents the compilation and discussion of about more than 90 analytical methods which includes HPLC, HPTLC, capillary electrophoresis, electrochemical methods and UV-Spectrophotometry methods implemented for investigation of VAL in biological matrices, bulk samples and in different dosage formulations. The review describes the percentage utilization of the various approaches for analysis of VAL. The statistical data regarding the utility of these methods for estimation of VAL published during 2001 to 2016 have been included.
Keywords: chromatography, valsartan, method validation, review article, bioanalysis

INTRODUCTION

Valsartan (VAL) is chemically N-(1-oxopentyl)-N-[[2'-(1H-tetrazol-5-yl) [1, 1'-biphenyl]-4-*yl*] methyl]-L-valine (**Figure 1**). It is crystalline in nature with melting point in the range of 116-117ºC; It is soluble in water [1]. VAL is an angiotensin-II receptor antagonist used in the management of hypertension. It may be used in patients with heart failure who are unable to tolerate ACE inhibitors [2]. VAL lowers blood pressure by antagonizing the Renin-Angiotensin-Aldosterone System (RAAS); it competes with angiotensin-II for binding to the type-1 angiotensin-II receptor (AT1) subtype and prevents the blood pressure increasing effects of angiotensin II [3]. VAL may be used to treat hypertension, isolated systolic hypertension, left ventricular hypertrophy and diabetic nephropathy. It may also be used as

Correspondence: Atul A. Shirkhedkar, *Department of Pharmaceutical Chemistry, R. C. Patel Institute of Pharmaceutical Education and Research, Shirpur Dist: Dhule(MS) India 425 405.*
shirkhedkar@gmail.com

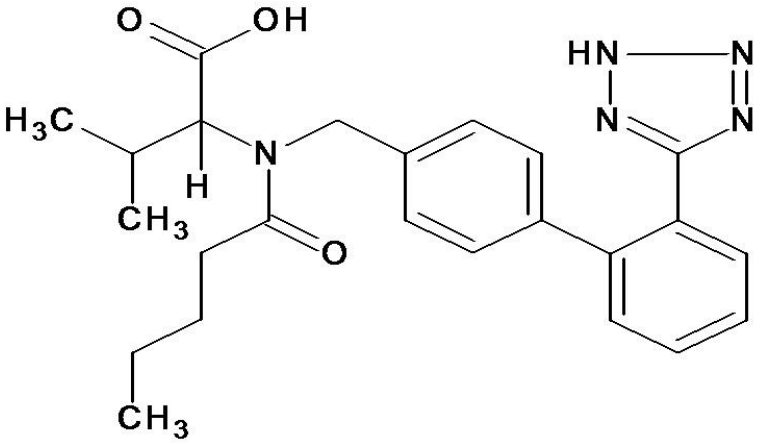

Figure 1. Chemical Structure of VAL

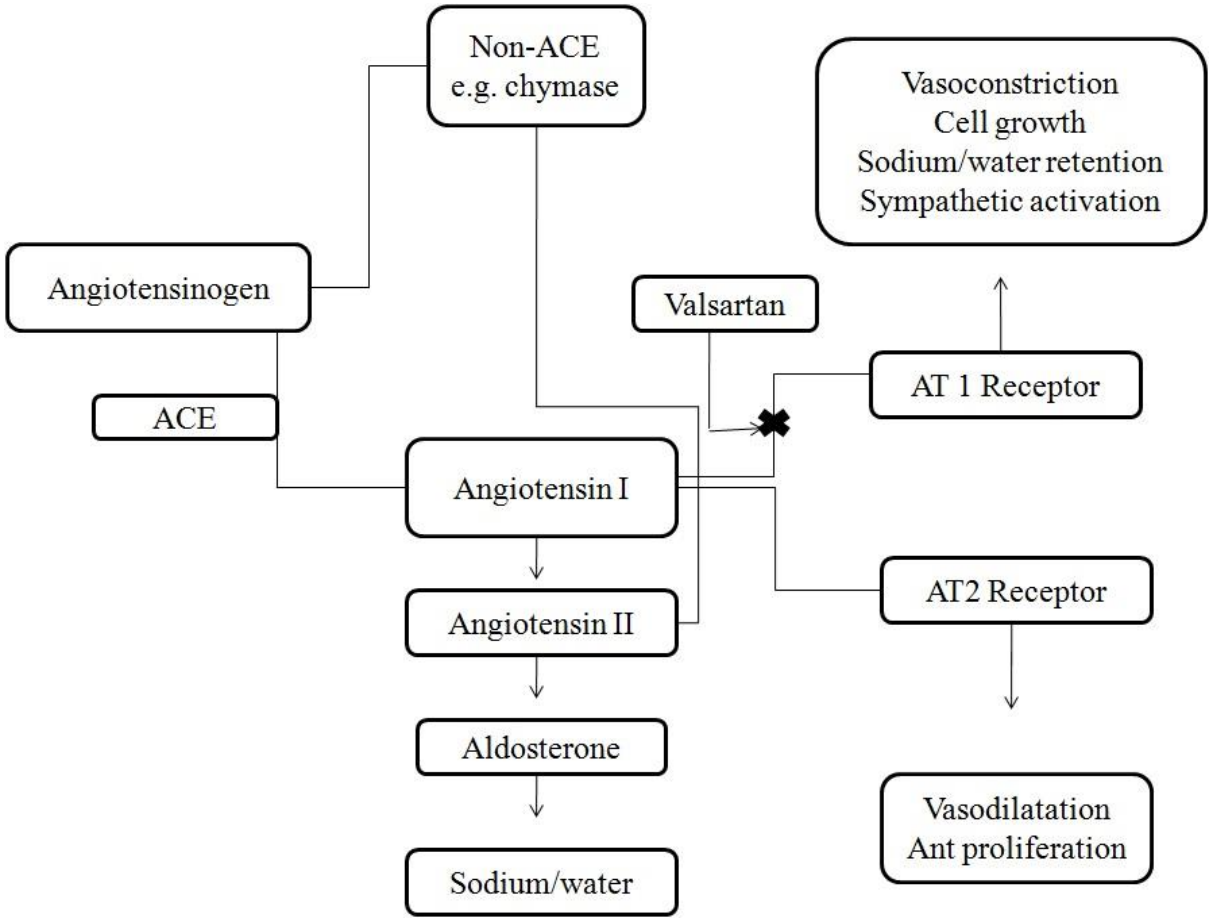

Figure 2. Schematic diagram of mode of action of VAL

an alternative agent for the treatment of heart failure, systolic dysfunction, myocardial infarction and coronary artery diseases [4].

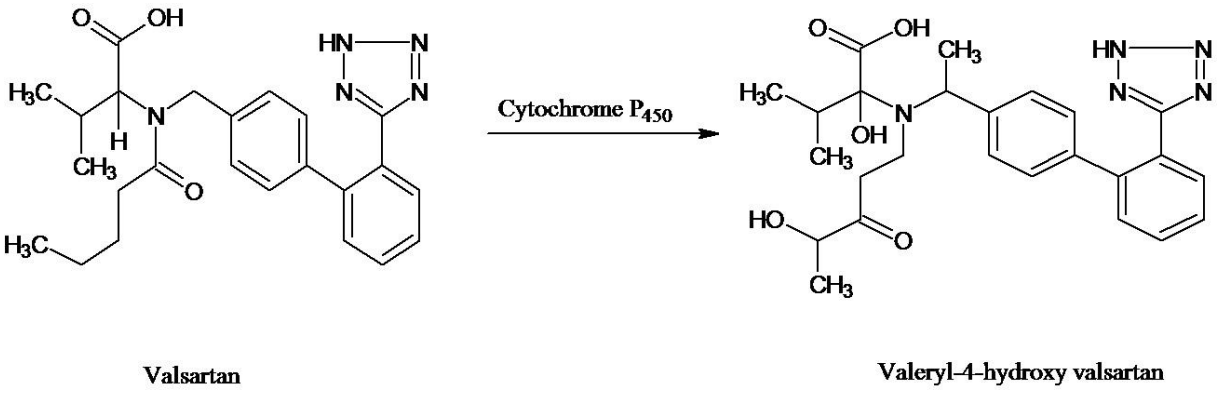

Figure 3. Metabolism of VAL to valeryl-4-hydroxy VAL

The schematic diagram shows the mode of action of VAL, **Figure 2**. Antagonism of angiotensin II receptor leads to blood pressure (BP) reduction, as well as decreases vascular smooth muscle contraction [6]. Pharmacokinetics reported that VAL is normally bound to serum protein - primarily serum albumin (94 - 97 %.). VAL is excreted largely as unchanged drug (80 %) and is minimally metabolized in humans. Metabolism of VAL gives valeryl 4-hydroxy metabolite is shown in **Figure 3** [7]. VAL is available in various doses, i.e. 10, 20,30,40,80,160 and 320 mg. It has also been reported that all these doses of VAL have been found to be safe and tolerable [8]. VAL is also available in combination with other antihypertensive agents such as nifidipine, hydrochlorothiazide, ramipril, amlodipine, nebivolol hydrochloride and antihyperlipidic agent vice ezetimibe. The information regarding dosage forms, route of administration and recommended dose of VAL is summarized in **Table 1**. VAL contraindicated in a person suffering from the renal artery, abnormally low blood pressure, liver problem, serious kidney problem and during pregnancy. The most common side effects of VAL are dizziness, low blood pressure, diarrhea, joint and back pain, hypotension, impaired renal function, hyperkalemia-Some patients with heart failure have developed increases in potassium [9].

Analytical accounts on VAL

The extensive literature survey revealed, several analytical techniques viz UV/Visible-Spectrophotometry, Spectrofluorimetry, HPLC, HPTLC and LC-MS for the determination of VAL in bulk and pharmaceutical formulations. The reported methods describe the estimation of VAL in various dosage forms as single constituent and in combination with amlodipine, hydrochlorothiazide, propranolol, nifedipine, ezetimibe, aliskiren, losartan, irbesartan, ramipril, nebivolol hydrochloride, atorvastatin, fluvastatin, simvastatin acid, ketoprofen, pentaprazole, chlorthalidone and with cilnidipine. **Figure 4** shows different analytical methods implemented for estimation of VAL.

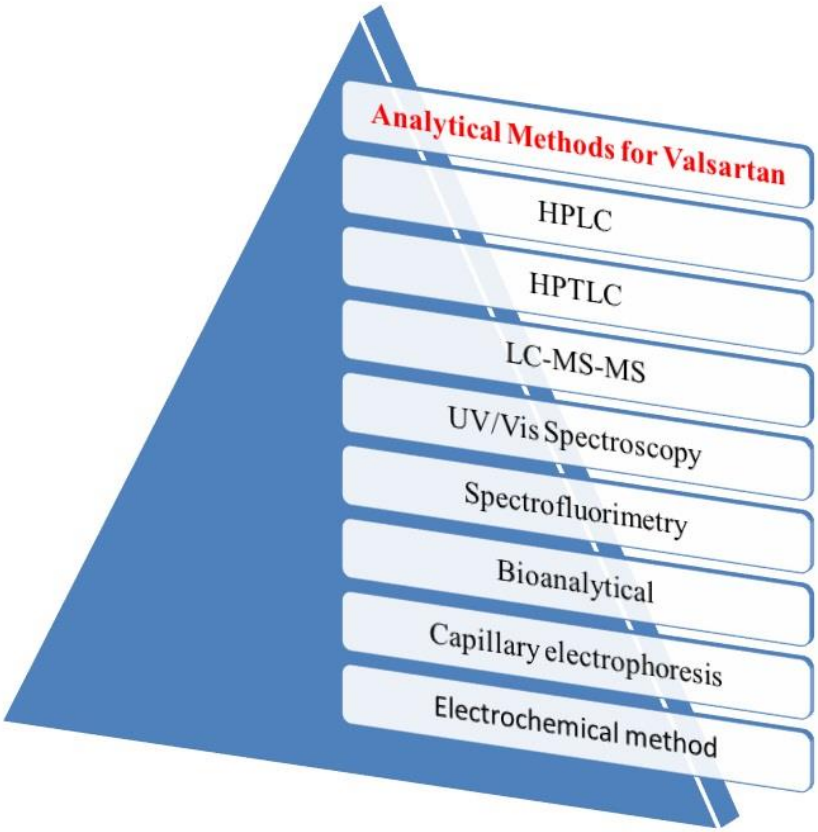

Figure 4. Analytical Methods of VAL

Table 1. Dosage forms, route of administration and recommended dose of VAL

Dosage forms	Route of Administration	Indication/Dose
Capsule	Oral	Pediatric Hypertension 160 mg daily
Capsule		Adult Hypertension 80 mg daily
Tablet		Pediatric Hypertension 6 to years of Age 40 mg daily
Tablet		Adult Hypertension -80 mg daily Heart failure- 80 mg daily
Tablet		Adult Hypertension-160 mg daily Heart failure- 160 mg daily Post myocardial infarction- 160mg twice a daily
Tablet		Heart failure 320 mg daily

Pharmacopoeial Status

VAL is the official drug in Indian Pharmacopoeia (IP) - 2010, the British Pharmacopoeia (BP) – 2012 and United States pharmacopeia (USP) - 32.

USP reported HPLC assay method using C18 (12.5 cm × 3 mm, 5µm) column as a stationary phase and a mobile phase consisted of acetonitrile, water and glacial acetic acid (500:500:1*v/v/v*) with a flow rate of 0.4 mL/min. Column effluent was monitored at 273 nm [10].

IP depicted HPLC assay method using C18 (25 cm × 4.6 mm, 5µm) column as a stationary phase and mobile phase consisted of acetonitrile, water and glacial acetic acid (50:50:0.1*v/v/v*) with a flow rate of 1 mL/min. Column effluent was monitored at 273 nm [11].

BP describe the potentiometric procedure in which 0.170 g of VAL is dissolved in 70 ml of 2-propranol, titrate with 0.1M tetrabutylammonium hydroxide and end point determine by potentiometrically [12].

Accounts on Bio-analytical Method for Determination of VAL

Bio-analysis is a sub-discipline of analytical chemistry covering the quantitative measurement of xenobiotics (drugs and their metabolites, and biological molecules in unnatural locations or concentrations) and biotic (macromolecules, proteins, DNA, large molecule drugs, metabolites) in biological systems. [13]

Literature survey revealed that HPLC is predominantly used for the bio-analysis of VAL.

Zong-Zhu Piaov et al. (2008) established a validated simple and sensitive method for determining VAL concentration in human plasma, the given sample was extracted by simple protein precipitation using methanol as a solvent. The analyst was separated using acetonitrile with 15 mm potassium dihydrogen phosphate in water (42:58 *v/v*) (pH 2.0; adjusted with phosphoric acid) with the flow rate of 1.2 mL/min [14].

Oskar Gonzalez et al. (2009) investigated the geometry optimization and the validation of a quantitative high-performance liquid-chromatography-photodiode array-fluorescence (HPLC-PDA-Fluo) method for the simultaneous analysis of combined drugs used in the treatment of cardiovascular diseases from human plasma. Separation of chlorthalidone (CLTD), VAL (VAL), VAL-M1 (VAL-M1) and fluvastatin (FLUV), using the mobile phase consisted of a mixture of acetonitrile and water containing 0.01% of formic acid and 10 mm of ammonium formate at (pH 4.1) [15].

D. R. Brunetto et al. (2009) studied Column-switching HPLC method and validation of it for quantification of losartan, telmisartan, and VAL in human urine. While analysts were extracted from the matrix using an on-line solid-phase extraction using solution 2% methanol in 5mM phosphate buffer (pH 3.8) at a flow-rate of 0.8 mL/min [16].

Table 2. Bioanalytical determination of VAL

Sr. No	Drug	Sample Matrix	Method	Colum	Detection	Internal Standard	Ref
1	VAL	Human Plasma	HPLC-UV	Phenomenex Luna C18 column	215 nm	Spironolactone	14
2	CLTD+VAL+VAL-M1+FLUV	Human Plasma	HPLC-PDA-Fluo	dC18 Atlantis column	CLTD-229 nm VAL- 254 nm FLUV-236 nm	Candesartan Cilexetil	15
3	VAL+LST+TMT	Human urine	HPLC	Chromolith RP-18e monolithic column	259 nm 399 nm	Candesartan M1	16
4	.VAL+ 4-OH VAL	Human Plasma	SPE-HPLC	RP C18 Atlantis	254 nm	Candesartan M1	17
5	VAL	Human Plasma	LC-MS/MS	XTerra MS C18	291.2 nm	Candesartan M1	18
6	VAL	Human Plasma	HPLC	Octadecylsilica column (mm,	234/374 nm excitation/emission wavelength	Candesartan M1	19
7	SA+AML+VAL	Human Plasma	LC-MS/MS	X-Terra C18 column	291.2 nm	Simvastatin acid	20
8	KTP+VAL+PTP	Human Plasma	HPLC	Chromasil C18 column	225 and 272 nm	Rofecoxib	21
9	VAL	Rat Plasma	HPLC-MS/MS	Thermo Hypurity C18	VAL -235 nm	Candesartan M1	22

Gorka Iriarte et al. (2007) reported a simple and fast method for the simultaneous determination of the antihypertensive drug VAL and its metabolite in human plasma. The proposed method deals with SPE, followed by an HPLC separation coupled with fluorimetric and photometric detection, the separation was performed on an RP C18 Atlantis 100 mm 63.9 mm column. The mobile phase consisted of a mixture of ACN 0.025% TFA and phosphate buffer (5 mm, pH = 2.5) 0.025% TFA and was delivered in gradient mode at a flow rate of 1.30 mL/min [17].

Nozomu Koseki et al. (2007) established a validated sensitive liquid chromatography-tandem mass spectrometry (LC-MS/MS) method for the determination of VAL in human plasma; analysts were extracted by solid-phase extraction using MeOH/H2O (50:50 *v/v*) [18]. Bioanalytical methods for determination of VAL are summarized in **Table 2**.

CHROMATOGRAPHY OVERVIEW

HPLC

Apart from pharmacopeial methods many HPLC methods were reported for determination for VAL in pharmaceutical formulations. The summary of the reported HPLC methods particularizing the mobile phase used for determination, sample matrix, λ_{max} and linearity is shown in **Table 3.** Instrumentation of HPLC methods for determination of VAL is summarized in **Table 4**.

Table 3. HPLC methods for VAL

Sr. No	Name of drug/ Formulation	Mobile phase composition	Detection (nm)	Discussion	Ref
1	VAL (Tablet)	Phosphate buffer: Acetonitrile (55:45 *v/v*)	233	VAL following linearity in the range of 1-6 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 3.94 min.	23
2	VAL (Tablet)	Water: Acetonitrile: Glacial acetic acid (500:500:01 *v/v/v*)	273	VAL obeyed linearity in the range of 40-140 µg/mL, Coefficient correlation was found out to be 0.999 and retention time was 4.6 min.	24
3	VAL (Tablet)	0.1M Phosphate buffer: Acetonitrile (20: 80 *v/v*)	273	VAL showed linearity in the range of 50-150 µg/mL, Coefficient correlation was found to be 0.9991 and retention time was 4.95 min.	25
4	VAL (Tablet)	Acetonitrile: Phosphate buffer (70:30 *v/v*)	273	VAL exhibited linearity in the range of 10-50 µg/mL, Coefficient correlation was found to be 0.9993 and retention time was 3.5 min.	26
5	VAL (Tablet)	Water: Acetonitrile : Glacial acetic acid (550:450:1*v/v*)	248	VAL having linearity in the range of 4-12 µg/mL, Coefficient correlation was found to be 0.9992 and retention time was 2.53 min	27
6	VAL (Tablet)	Acetonitrile : Phosphate buffer of pH 3.5: Triethylamine (40:60 *v/v*)	250	VAL having linearity in the range of 1-100 µg/mL, Coefficient correlation was found to be 0.996 and retention time was 5.19 min	28
7	VAL (Tablet)	Phosphate buffer : Acetonitrile (50:50 *v/v*)	210	VAL having linearity in the range of 5-25 µg/mL, Coefficient correlation was found to be 0.9998 and retention time was 4.45 min	29
8	VAL (Tablet)	Acetate buffer pH 4.6: Acetonitrile: Methanol (38:24:38 *v/v/v*)	248	VAL having linearity in the range of 10-30 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 4.6 min	30
9	VAL (Tablet)	Methanol : Phosphate buffer pH 3.0 (65:35 *v/v*)	210	VAL having linearity in the range of 10-100 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 6.22 min	31
10	VAL+ ALK (Tablet)	Methanol : Potassium Di Hydrogen Phosphate buffer : Acetonitrile pH 3.01 % orthophosphoric acid (50:30:20 *v/v/v*)	271	VAL and ALK having linearity in the range of 10-50 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 7.91 and 6.92 min	32

Table 3. HPLC methods for VAL (continued)

Sr. No	Name of drug/ Formulation	Mobile phase composition	Detection (nm)	Discussion	Ref
11	VAL+AML+ HCTZ (Tablet)	Potassium dihydrogen orthophosphate buffer (50 mM, pH 3.7) with 0.2% triethylamine : Acetonitrile (56:44 *v/v*)	232	VAL, AML and HCTZ having linearity in the range of 20-150 µg/mL 2-25 µg/mL and 5-45 µg/mL, Coefficient correlation was found to be VAL- 0.9971, AML-0.9945 and HCTZ-0.9967 and retention time was 10.15 min, 4.2 min and 3.56 min	33
12	VAL+ HCTZ (Tablet)	Potassium dihydrogen Orthophosphate : Methanol: Triethylamine (25:75:0.5 *v/v/v*)	259	VAL and HCTZ followed linearity in the range of VAL-32-80 µg/mL and HCTZ-2.5-12.5 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 4.15 and 3.20 min	34
13	VAL+LST+IRB (Tablet)	Acetonitrile: phosphate potassium buffer (pH 3) (40:60 *v/v*)	254	VAL, LST and IRB followed linearity in the range of 40-120 µg/mL, Coefficient correlation was found to be VAL-0.999 IRB and LST-0.999 and retention time was VAL-15.7 and 3.20 min. LST-8.31 and IRB-11.23 min	35
14	VAL + PROP (Tablet)	Acetonitrile: Methanol: 0.01 M disodium hydrogen phosphate (pH 3.5) (50:35:15 *v/v*)	250	VAL and PROP obeyed linearity in the range of VAL- 4-32 µg/mL and PROP-5-50 µg/mL, Coefficient correlation was found to be VAL-0.9966 and PROP-0.9988 retention time was 9.76 and 6.62 min	36
15	VAL+HCTZ (Tablet)	Methanol: isopropyl alcohol: n-hexane (50:25:25 *v/v/v*)	265	VAL and HCTZ obeyed linearity in the range of VAL-40-120 µg/mL and HCTZ- 6-18 µg/mL, Coefficient correlation was found to be 0.997 and 0.9997 and retention time was 1.5 and 3.5 min	37
16	VAL+ ALK (Tablet)	Acetonitrile: 0.05M Potassium dihydrogen phosphate buffer, (pH 3.5) adjusted with O-Phosphoric acid (45:55 *v/v*)	224	VAL and ALK having linearity in the range of 10-50 µg/mL, Coefficient correlation was found to be 0.9985 and 0.999 and retention time was 6.5 and 3.14 min	38
17	AML+ VAL+ HCTZ (Tablet)	Mixture of 30mM phosphate buffer (pH 5.5) : Methanol (38:62 *v/v*)	234	VAL, AML and HCTZ having linearity in the range of 17.6-32.8 µg/mL, 7-13 µg/mL and 5-45 µg/mL, Coefficient correlation was found to be VAL- 0.996, AML-0.999 and HCTZ-0.997 and retention time was 1.5 min, 4.2 min and 3.5 min	39

Table 3. HPLC methods for VAL (continued)

Sr. No	Name of drug/ Formulation	Mobile phase composition	Detection (nm)	Discussion	Ref
18	AML+ VAL+ HCTZ (Tablet)	Acetonitrile: Phosphate buffer (0.05 M) with (pH 2.8) (40/60 *v/v*)	227	VAL, AML and HCTZ having linearity in the range of 5-40 µg/mL, 4-28 µg/mL and 1-12 µg/mL, Coefficient correlation was found to be VAL- 0.996, AML-0.999 and HCTZ-0.997 and retention time was 11.19 min, 3.16 min and 2.26 min	40
19	AML+ VAL+ HCTZ (Tablet)	Water: Acetonitrile: Tri-Fluoroaetic acid (55:45:0.1 *v/v/v*)	AML-237 VAL-237 HCTZ -265	VAL, AML and HCTZ having linearity in the range of 80-240 µg/mL, 5-15 µg/mL and 18-75 µg/mL, Coefficient correlation was found to be VAL- 0.998, AML-0.999 and HCTZ-0.999 and retention time was 9.63 min, 6.83 min and 3.241 min	41
20	AML+VAL (Tablet)	Phosphate buffer (pH 3.6, 0.01 mol L-1): Acetonitrile: Methanol (46:44:10 *v/v/v*)	240	VAL and AML having linearity in the range of 10-50 µg/mL, Coefficient correlation was found to be 0.999 and retention time was 7.91 and 6.92 min	42

HPTLC

Six simple HPTLC methods have been studied for simultaneous estimation of VAL in combined dosage form with CLN, RMP, HCTZ and NBH. The summary of the reported HPTLC methods is shown in **Table 5**.

Ritesh P. Bhole et al. (2015) developed and validated a simple method for VAL and CLN in combined dosage form, standard solution of VAL and CLN were applied to precoated silica gel 60F 254, and mobile phase used for development toluene: methanol: ethyl acetate: glacial acetic acid in the ratio of (8:1:1:0.1 *v/v/v*) and Rf value was found to be 0.29 and 0.56, respectively. Accuracy and precision of the proposed method were evaluated by recovery studied and % recovery for VAL and CLN was 99.03 % and 99.86 % [43].

Della Grace Thomas Parambi et al. (2011) investigated a simple, accurate and precise method for quantitative estimation of VAL in tablet matrix. Standard solution of VAL was applied to pre-coated silica gel 60F 254, mobile phase used for development chloroform: acetonitrile: toluene: glacial acetic acid, in the ratio (1:8:1:0.1 *v/v/v/v*), and Rf value was found to be 0.65. The method showed good repeatability and recovery with relative standard deviation less than 2 [44].

Table 4. HPLC chromatographic columns and optimized analytical parameters

Sr. No	Name of drug	Column	Internal diameter and partical Size	Detector	Flow rate	Mode of analysis	Diluents	Ref
1	VAL	Kromasil C18 column	250×4.6, 5µm	UV detector	1 mL/min	Isocratic mode	Acetonitrile	23
2	VAL	Thermo hypersil ODS column	150 × 4.6, 5µm	UV detector	1.0 mL/min	Isocratic mode	Acetonitrile	24
3	VAL	A Venusil XBP C-18	250 × 4.6, 5µm	UV detector	1.0 mL / min	Isocratic mode	Acetonitrile	25
4	VAL	Agilent ODS UG 5 column C18 column	250 x 4.5, 5µm	UV detector	1.0 mL / min	Isocratic mode	Acetonitrile	26
5	VAL	X terra,RP-18	100 x 4.6, 5µm	UV-Visible	2.0 mL/min	Isocratic mode	Acetonitrile	27
6	VAL	Kromasil C-18	250 × 4.6 5µm	UV/VIS detector	1.0 mL/min	Gradient mode	Triethylamine	28
7	VAL	Xterra C18 column	100 × 4.6, 5µm	UV detection	1 mL/min.	Isocratic mode	Acetonitrile	29
8	VAL	ODS C18	250 × 4.6, 5µm	PDA detector	1.2 mL min	Isocratic mode	Methanol	30
9	VAL	Phenomenox C18	25 × 4.6, 5µm	PDA and UV detector	1mL min-1	Isocratic mode	Methanol	31
10	VAL+ ALN	Hiber Lichrosphere C18	250 × 4.6, 5µm	UV Detector	1.0 mL/min	Isocratic mode,	Acetonitrile	32
11	VAL+AMB+ HCTZ	Kromasil KR-5 C18 column	250 x 4.6, 5µm	PDA Detector	1.0 mL/min	Isocratic mode	Water	33
12	VAL+ HCTZ	C-18 intersil	250 x 4.6, 10µm	UV detector	1.0 mL/min	Isocratic mode	Triethylamine	34
13	VAL+LST+ IRB	C18, Eurospher	250 x 4.6, 5µm	UV detector	1.5 mL/min	Isocratic mode	Acetonitrile	35

Table 4. HPLC chromatographic columns and optimized analytical parameters (continued)

Sr. No	Name of drug	Column	Internal diameter and partical Size	Detector	Flow rate	Mode of analysis	Diluents	Ref
14	VAL + PROP	Hypersil ODS C-18 column	250 x 4.6, 5µm	UV detector	1.0 mL/min	Isocratic mode	Acetonitrile	36
15	VAL+HCTZ	Lichrosphere CN column	250 x 4.0, 5µm	UV detector	1.0 mL/min	Isocratic mode	Isopropyl alcohol	37
16	VAL+ ALK	Hyper ODS2, Column C18,	250 x 4.6, 5µm	UV detector	1 mL/min	Isocratic mode	Acetonitrile	38
17	AML+HCTZ VAL	Luna C18 column	250 x 4.6 5µm	UV detector	1.0 mL/min	Isocratic mode	Methanol	39
18	AML+VAL+ HCTZ	Phenomenex Kinetex RP-C18	150 x 4.6, 5µm	UV detector	0.8 mL/min	Isocratic mode	Acetonitrile	40
19	AML+ VAL+ HCTZ	Hypersil BDS C18 column	250 x 4.6 5µm	UV detector	1.0 mL/min	Isocratic mode	MP	41
20	AML+VAL	ODS 2,C18	200 x 4.6, 10µm	UV detector	1 mL/ min	Isocratic mode	MP	42
21	VAL	Waters 2695 using Symmetry C18	250 × 4.6, 5µm	PDA detector	1 mL/ min	Isocratic mode	MP	49
22	VAL+EZM	Symmetry C18 column	250× 4.6, 5µm	PDA detector	0.8 mL/ min	Isocratic mode	MP	50
23	VAL+ PRP	A Hypersil C18 column	250 × 4.6, 5µm	UV detector	1mL/min	Isocratic mode	MP	51
24	VAL+ HCTZ	Xterra column	25 ×4.6, 5µm	PDA detector	1.5 mL/min	Isocratic mode	MP	52
25	VAL+ATV	Hypersil BDS C18	250 × 4.6, 5µm,	UV detector	2.0 mL/ min	Isocratic mode	MP	53
26	AML+VAL+HCTZ	Zorbax SB-C8 column	250 × 4.6, 5 µm	PDA detector	1 mL/min	Isocratic mode	MP	54
27	VAL	C18 column	250 × 4.6, 5µm	UV detector	1.2 mL/min	Isocratic mode	Acetonitrile	55

Stability-Indicating Methods (SIM) for Determination of VAL

About seven stability-indicating methods have been studied so for determination of VAL in bulk substances and pharmaceutical formulations implementing different analytical techniques. Amongst these, three methods are for estimation of VAL alone and four of them described in stability studies of VAL in its combined dosage form with other drugs. The reported stability-indicating methods for VAL, illustrating sample matrix, λ_{max}, linearity range and retention time/factor presented in **Table 6**.

Table 5. HPTLC methods for determination of VAL

Dr. No	Name of drug	Formulation	Stationary Phase plates	Mobile phase Composition	Detection (nm)	Linearity	Rf	Ref
1	VAL+CLN	Tablet	Silica gel 60 F 254	Toluene: Methanol: Ethyl acetate: Glacial Acetic acid (8:1:1:0.1 *v/v/v/v*)	240	CLN-1000- 6000 ng/band VAL-8-48 µg/band	CLN-0.56 VAL-0.29	43
2	VAL	Tablet	Silica gel 60 F 254	Chloroform: Acetonitrile: Toluene: Glacial acetic acid (1:8:1:0.1 *v/v/v/v*)	254	VAL-50-500ng/band	VAL-0.65	44
3	VAL + RMP	Tablet	Silica gel 60 F 254	Ethyl acetate : Chloroform: Glacial acetic acid (8:2:0.2 *v/v*)	220	RMP- 800 – 4000 ng/spot VAL-50-500ng/band	RMP-0.15 VAL-0.49	45
4	VAL+HCTZ	Tablet	Silica gel 60 F 254	Chloroform: Ethyl acetate: Acetic acid	248	VAL-800-5600 ng/spot HCTZ-125-875 ng/spot	VAL-0.25 HCTZ-0.46	46
5	NBH + VAL	Tablet	Silica gel 60 F 254	Ethyl Acetate: Methanol: Acetic acid (6:1:0.5 *v/v/v*)	280 240	NBH-1200-2800ng/band VAL-600-1400ng/band	NBH-0.14 VAL-0.89	47
6	VAL + HCTZ	Tablet	Silica gel 60 F 254	Chloroform: Methanol: Formic acid (4:1:0.05 *v/v/v*)	264	VAL-1000 – 7000 ng/spot HCTZ-200 – 1000 ng/spot	VAL-0.76 HCTZ -0.44	48

Spectrophotometry Methods

Till the date, twenty-two UV-Spectrophotometry methods have been established for determination of VAL alone and in combined dosage forms. Also, two Spectrofluorimetry methods have been investigated analysis of VAL in tablets. The details Spectrophotometry and Spectrofluorimetry designating the basic principle, sample matrix, λ_{max} and solvent and linearity range is summarized in **Table 7**.

Table 6. Stability-indicating HPLC and HPTLC methods for determination of VAL

Sr. No	Name of drug	Sample Matrix	Mobile phase	Detection (nm)	Linearity	Rt/Rf	Ref
			HPLC Methods				
1	VAL	Tablet	0.02 mM sodium dihydrogen ortho-phosphate: (pH 2.5): Acetonitrile (58:42 *v/v*)	250	1–200 µg/mL	9.17-9.24 min	49
2	VAL+ EZT	Tablet	Phosphate buffer: Acetonitrile (pH 3.15) (58:42 *v/v*)	230	1- 200µg/mL	EZE- 0.14 and 1.80 min VAL- 0.12 min	50
3	VAL	Tablet	Ammonium dihydrogen phosphate buffer: Methanol (pH 3) with formic acid. (33.5:66.5 *v/v*)	265	1–200 µg/mL	VAL-11.9 Min	51
4	VAL+ HTZ	Tablet	0.20 M ammonium acetate, adjusted to pH 5.6 with Glacial acetic acid: Acetonitrile. (88:12 *v/v*)	265	VAL- 2.5–32 µg/mL HCTZ- 17.5-224 µg/mL	HCTZ- 5.00 VAL- 6.837 min	52
5	VAL+ ATV	Tablet	0.1% Acetic acid: Acetonitrile (50:50 *v/v*)	VAL-225 ATV-246	ATV- 5-15 µg/mL VAL- 40-120 µg/mL	VAL- 3.33 min ATV- 5.44 min	53
6	AML+ VAL+ HCTZ	Tablet	0.025M phosphoric acid: Acetonitrile (75:25 *v/v*)	AML-238 VAL- HCTZ- 225	AML- 5–200 µg/mL VAL- 5–200 µg/mL HCTZ- 10–200 µg/mL	HCTZ- 4.9 min AML- 6.4 min VAL- 8.3 min	54
7	VAL	Bulk	▪ Acid hydrolysis methanol: water (pH 7.2) (70:30 *v/v*) ▪ Oxidion methanol: water (pH 7.2) (60:40 *v/v*)	250	5-45 µg/mL 20-100 µg/mL	1.9.57 min 2.2.24 min	55
			HPTLC method				
1	NBH + VAL	Tablet	Ethyl Acetate: Methanol: Acetic acid (6:1:0.5 *v/v/v*)	NBH-280 VAL-240	NBH- 1200-2800ng/band VAL- 6001400ng/band	NBH- 0.14 VAL- 0.89	47

Table 7. Spectrophotometric and Spectrofluorimetric methods used for determination of VAL alone and in combined dosage form

Sr. No	Name of Drug	Sample matrix	Methods	Detection (nm)	Linearity	Correlation coefficient (r^2) value	Ref
1	VAL	Tablet	Zero order Second order	VAL-250 VAL-241	10-50 µg/mL	0.9998 0.9987	56
2	VAL	Tablet	Zero order Second Order	VAL-250 VAL-220	5-30 µg/mL	0.9995 0.9989	57
3	VAL+NIF	Synthetic Mixture	Zero order	VAL-262.6 NIF-327.5	2-20 µg/mL	0.9994 0.9976	58
4	VAL+ HCTZ	Tablet	First order	VAL-270.6 HCTZ-335	12-36 µg/mL 4.0-12 µg/mL	0.9975 0.9995	59
5	VAL	Tablet	Zero order	VAL-250	2-20µg/mL	0.9968	60
6	VAL	Capsule	Zero order Second order	VAL-205.6 VAL-231.2	2-10 µg/mL	0.9997	61
7	VAL+AML	Tablet	SEM ACM	VAL-250 AML-238	5-30 µg/mL	0.998 0.999	62
8	VAL + HCTZ	Tablet	ACM	VAL-231.5 HCTZ-270.5	2-20 µg/mL	0.9993 0.9986	63
9	VAL+HCTZ	Tablet	First order	VAL-250.20 HCTZ-270.60	VAL-4-20 µg/mL HCTZ-2-14 µg/mL	0.998 0.998	64
10	VAL+ AML	Tablet	DDI	VAL-282 AMD-247	5-40 µg/mL 1-100 µg/mL	0.9991 0.9991	65
11	VAL+AML+ HCTZ	Tablet	First order	VAL-245 AMD-265 HCTZ-279	VAL-8-80 µg/mL AMD-1-10 µg/mL HCTZ-2-20 µg/mL	0.9994 0.9996 0.9998	66
12	VAL+ HCTZ	Tablet	SEM ACM	VAL-249.9 HCTZ-272.6 VAL-258.4 HCTZ-272.6	5-30 µg/mL 4-24 µg/mL	0.998 0.998 0.999 0.999	67
13	VAL+ HCTZ	Tablet	SEM	VAL-248.5 HCTZ-271	VAL-0.5-3.5 mg/mL HCTZ- 0.5-1.4 mg/mL	0.9991 0.9998	68
14	AML+VAL	Tablet	Zero order First order	AML-360.5 VAL-290	AML-10-80 µg/mL VAL-20-180 µg/mL	0.999 0.999	69
15	CIL+ VAL	Synthetic Mixture	SEM	CIL-240 VAL-250	2-10 µg/mL 16-80 µg/mL	0.999 0.999	70

Table 7. Spectrophotometric and Spectrofluorimetric methods used for determination of VAL alone and in combined dosage form *(continued)*

Sr. No	Name of Drug	Sample matrix	Methods	Detection (nm)	Linearity	Correlation coefficient (r^2) value	Ref
16	VAL+ EZT	Tablet	Zero order	VAL-423 EZT-250	5-40 µg/mL 1-50 µg/mL	0.995 0.999	71
17	AML+ VAL	Capsule	First order ACM	AML-250 VAL-237.5 AML-250 VAL- 360	AML-10-50 µg/mL VAL-0-80 µg/mL	0.9992 0.9982 0.9996 0.9986	72
18	VAL+ EZT	Capsule	Second order	VAL-289.36 EZT- 226.89	VAL-2-16 µg/mL EZT-2-16 µg/mL	0.9973 0.9987	73
19	VAL+ HCTZ	Tablet	SEM	VAL-250 HCTZ-270	VAL-6-36 µg/mL HCTZ-2-12 µg/mL	0.998 0.999	74
20	VAL	Tablet	Std, Abs. AUC First order Q Abs Ratio	VAL-249 AUC-238-254 VAL-249 VAL-235-250	5-30 µg/mL	0.998	75
21	VAL+ NEB+ HCTZ	Capsule	SEM	VAL-246.6 NEB-275 HCTZ-280.2	VAL-1-20 µg/mL NEB- 0.5-2.5 µg/mL HCTZ- 1-3 µg/mL	0.999 0.998 0.999	76
22	VAL + AML + HCTZ	Table	SEM	VAL-250 AMB-239 HCTZ-272	VAL -4-40 µg/mL AMB-1-32µg/mL HCTZ- 2-20 µg/mL	0.99971 0.99992 0.99990	77
23	VAL + AML	Tablet	Spectrofluorimetry	VAL-227 AML-390	VAL-10-22 µg/mL AML-04-14 µg/mL	0.9997 0.9997	78
24	VAL+ AML	Tablet	Spectrofluorimetry	VAL-475 AML378	VAL-0.2-3.6 µg/mL AML- 0.008-0.080 µg/mL	0.99975 0.99985	79

Approaches for Analysis of VAL as a Single Component

Gupta et al. (2010) has described zero order and second order UV-spectrophotometry method for determination of VAL in tablets using methanol as solvent. The zero order and second order derivative method involve the calculation of absorbance at 250 nm and 241 nm respectively [56].

Tarkase et al. (2012) also performed similar UV- spectrophotometry methods for VAL tablets using phosphate buffer for dissolution of VAL and absorbance was recorded at 220 nm in second order derivative method [57].

Tatar et al. (2002) has employed ethanol for the solubilisation of VAL. The article described second order UV-spectrophotometry in which the distance between two extremum

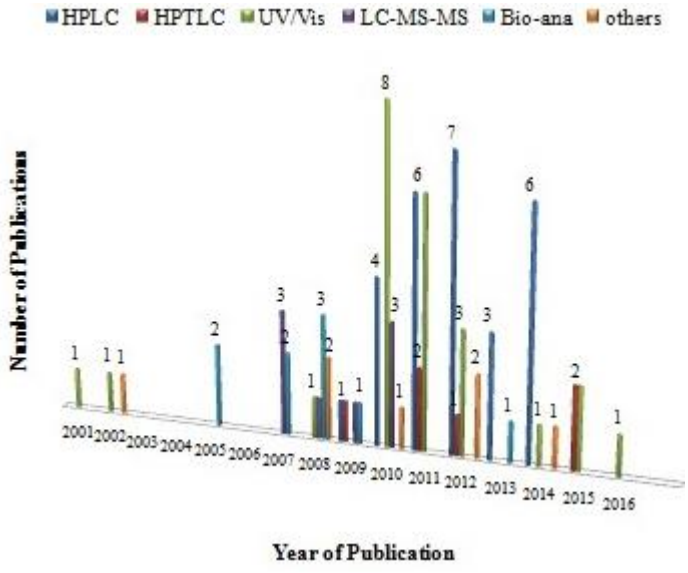

Figure 5. Statistics of research paper for estimation of VAL published during 2001 to 2016

values peak-to-peak amplitudes 221.6 nm and 231.2 nm were measured for determination of VAL in capsules and calibration curves were constructed by plotting $d^2A/d\lambda^2$ against concentrations of VAL solutions [61].

Kalaimagal et al. (2012) has reported standard absorbance method, Area under Curve method, first order derivative and Q-absorbance method using 0.1 N NaOH as solvent for VAL with good recovery in the range of 98.6% to 102.26% [75].

Approaches for analysis of VAL in combined dosage form with other drugs

VAL is available in combination with many antihypertensive, diuretics and antihyperlipidic agents. Few UV-Spectrophotometry methods have been stated for simultaneous determination of VAL in dosage forms and simple, rapid, accurate and economical methods have been developed for the assessment of VAL and HCTZ in tablet dosage form.

Satana E et al. (2001) developed a simple first order derivative method for analysis of VAL and HCTZ at wavelength 270.6 nm and 335 nm [59].

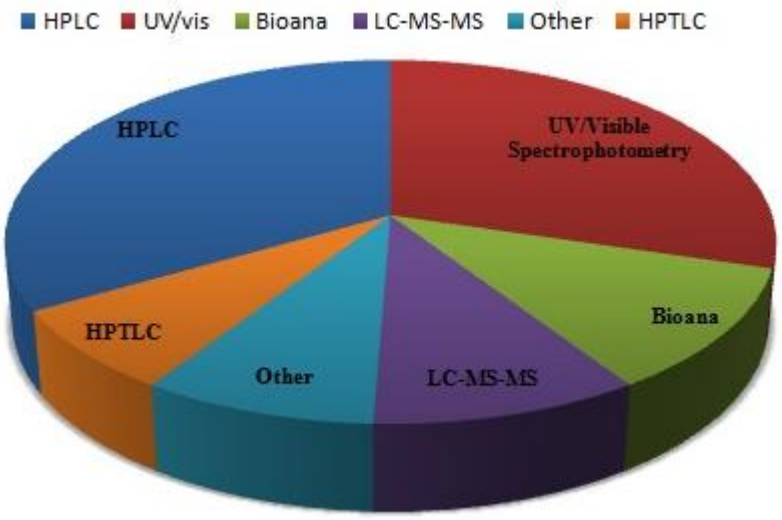

Figure 6. Percentage Utility of Analytical Approaches used for estimation of VAL

Patel S. A. et al. (2016) reported two simple, accurate and precise methods have been studied and validated for the simultaneous estimation of VAL and AML and in their combined dosage form of UV- Spectrophotometric methods. Simultaneous equation method (SEM) employs investigation of VAL and AML using 250.0 nm and 238.0 nm i.e. λ_{max} values of VAL and AML, respectively. Absorption Correction method (ACM) employs the estimation of VAL and AML at 360.0 nm i.e. λ_{max} values of one drug and 236.0 nm an isobestic wavelength [58].

Abdallah O.M. et al. (2011) validated a first order derivative method for measurements of the amplitudes of 234.5 nm and 247 nm for AML using 30 µg/mL of VAL as a divisor and at 282 nm and 292 nm for VAL [65].

Chaudhary A.B. et al. (2010) described two UV-Spectrophotometry methods have been developed and validated for simultaneous estimation of VAL and HCTZ in a tablet dosage form. The first method employed solving of simultaneous equations based on the measurement of absorbance at two wavelengths, 249.4 nm and 272.6 nm, λ_{max} for VAL and hydrochlorothiazide, respectively. The second method was the absorbance ratio method, which involves the studies of Q-absorbance equation at 258.4 nm (isoabsorptive point) and also at 272.6 nm λ_{max} of hydrochlorothiazide [63].

Meyyanathan S.N. et al. (2010) reported two simple, precise and reproducible UV-spectrophotometry methods, simultaneous equation method and Q-value analysis method, have been developed and validated for the simultaneous estimation of NEB, HCTZ and VAL 246.6 nm, 280.2 nm and 275 nm, respectively [76].

Jothieswari D. et al. (2010) illustrated simple, accurate, precise and reproducible UV spectrophotometric method has been developed for the simultaneous estimation of AML, VAL and HCTZ 239 nm, 250 nm, 272 nm, respectively [77].

Spectrofluorimetric Methods

Mohammed et al. (2015) reported two different Spectrofluorimetry methods, the first method depends on measurement of native fluorescence intensity of both drugs at emission 460 nm and 385 nm is using excitation 390 nm and 227 nm for AML and VAL, respectively in water. The second method utilizes a synchronous fluorimetric quantitative screening of the emission spectra of AML and VAL at 375 nm and 285 nm, respectively using Δλ of 80 nm [78].

Shaalan R. A. et al. (2010) described a simple, sensitive and reliable spectrofluorimetry method for the simultaneous determination of the two antihypertensive drugs; AML and VAL 360 nm, 245 nm respectively [79].

Liquid Chromatography–Mass spectrometric Methods

Nerea Ferreiro et al. (2007) investigated a validated quantitation of angiotensin-II receptor antagonists (VAL) (ARA-II) in human plasma, a method using liquid-chromatography (LC)-electro-spray ionization tandem mass spectrometry (MS/MS) has been reported with respect to simple sample clean-up and investigation of ion suppression effects. Sample prepared method used protein precipitation by using zinc sulphate and methanol [80].

Oskar Gonzalez et al.(2010) studied validated a simple fast method simultaneous analysis, in human plasma of VAL using high-performance liquid chromatography–tandem mass spectrometry (LC–MS/MS) with electro-spray ionization (ESI), Separation of analytes and internal standard (pravastatin) was achieved on a Luna C18(2) (150 mm×4.6 mm, 3 m) column using a gradient elution mode with a run time of 15 min, and the mobile phase composition mixture of acetonitrile and water containing 0.01% formic acid and 10 mM ammonium formate at pH 4.1 and Sample extracted by protein precipitation by using acetonitrile [81]

Mikaël Lev et al. (2009) studied assay method for direct analysis of VAL in human plasma and urine by a direct on-line solid-phase extraction coupled to tandem-mass spectrometry [82].

Hao Li et al. (2007) established a rapid and sensitive liquid-chromatography/tandem-mass spectrometry (LC/MS/MS) method was developed and validated for simultaneous quantification of VAL and HCTZ in human plasma. After a simple protein precipitation using acetonitrile, the analytes were separated on a Zorbax SB-Aq C18 column using acetonitrile −10 mM ammonium acetate (60:40 *v/v)* (pH 4.5) as a mobile phase with flow rate 1.2 mL/min [83].

Chi-Yu Lu et al. (2009) reported a simple and sensitive method for analysis of clinical drug and biomarkers in human plasma using LC connected to tandem mass spectrometry (LC–MS/MS) with a nanospray ion source. Drug and proteins were separated on a 5 and 10

cm RP C18 nano-flow column. Undesired polar substances in human plasma were washed out by using ACN-1% FA= (20: 80 *v/v*) as the loading mobile phase for drug analysis [84].

P. Senthamil Selvan et al. (2007) reported a rapid, sensitive and accurate liquid chromatographic-tandem mass spectrometry method for the simultaneous determination of NEB and VAL in human plasma. NEB and VAL were extracted from plasma using acetonitrile and separated on a C18 Column; the mobile phase consisting of a mixture of acetonitrile and 0.05mM formic acid (50:50 *v/v*, pH 3.5) was delivered at a flow rate of 0.25 mL/min [85].

Oskar Gonzalez et al. (2011) studied LC–MS/MS method with positive electro-spray ion ization for the screening of commonly prescribed cardiovascular drugs (VAL) in human plasma, including compounds with antihypertensive (57), antidiabetic (12), hypolipemiant (5), anticoagulant (2) and platelet anti-aggregation (2) effects. Sample treatment consisted of a simple protein precipitation with MeOH/0.1M ZnSO4 (4:1 *v/v*) solution after the addition of internal standard, followed by evaporation and reconstitution. Analytes separation was achieved on a Polar-RP column (150 mm×2 mm, 4 m) using a gradient elution of 15 min [86].

Surbhi Mehta et al. (2010) demonstrated the applicability of a strategy involving use of liquid chromatography (LC) and liquid-chromatography mass spectrometry (LC-MS) techniques for identification and characterization of minute quantities of degradation products, without their isolation from the reaction matrix in pure form. In that, they used a VAL as a model drug, three small degradation products were formed, which were separated on a C-18 column using a gradient method [87].

Capillary Electrophoresis (CE) Method

S. Hillaert et al. (2003) implemented the capability of the capillary zone electrophoretic (CZE) and micellar electro kinetic capillary chromatographic (MEKC) methods to simultaneously separate hydrochlorothiazide and six angiotensin-II-receptor antagonists (ARA-IIs): candesartan, eprosartan, mesylate, irbesartan, losartan potassium, telmisartan, and VAL. Experiment were performed on thermo capillary electrophoresis, a fused silica capillary was used 85 cm in length and 50 mm. Absorbance was detected at 214 nm, two different internal standard were involved sulfanilamide and eprosartan mesylate for study [88].

Potentiometric Methods

Nazife Aslan et al. (2010) developed and validated a potentiometrically titration method for determination of VAL in pharmaceutical dosage forms. From the titration data, stochiometric protonation constants are calculated and these constants are found to be 4.57 and 5.47, titration were carried out in ethanol solutions using NaOH as titrant, at constant temperature of 25 ± 0.1°C and ionic strength of 0.10 M NaCl [89].

Shrikant H. Patil et al. (2012) developed and validated a novel and simple titrimetric method for determination of commonly used angiotensin-II-receptor antagonists (ARA-IIs). The direct acid base titration of four ARA-IIs, namely eprosartan mesylate, irbesartan,

telmisartan and VAL, was carried out in the mixture of ethanol: water (1:1 *v/v*) as solvent using standardized sodium hydroxide aqueous solution as titrant, either visually using phenolphthalein as an indicator or potentiometrically using combined pH electrode [90].

Nesrin K. Ramadanv et al. (2012) reported a potentiometrically method for VAL and AML in that they concluded two poly (vinyl chloride) matrix membrane electrodes responsive to some drugs affecting the cardiovascular system [91]. Potentiometric method offers a simple system and cost effective method than that of other methods which are having high cost, multiple steps and time consuming.

Voltammetric Methods

I. H. I. Habib et al. (2008) reported stripping voltammetry determination of VAL using a Hanging Mercury Drop Electrode (HMDE), was based on adsorptive accumulation of the species at HMDE followed by first harmonic alternating current AC stripping sweep at pH 6, the response was linear over the concentration range of 0.08–0.64 mg/mL with regression coefficient 0.999 [92].

Pinar Esra Erden et al. (2014) reported anodic behavior of binary mixture of AML and VAL on glassy carbon electrode based on the irreversible oxidation signal of AML at 0.95 and that of VAL at 1.15 V versus Ag/AgCl at pH 5.0 in Britton-Robinson buffer. Differential pulse voltammetry method was proposed to direct determination of AML and VAL in pharmaceuticals and spiked human serum. Linearity for AML was in the range from 1.0 µM to 35.0 µM and that for VAL was in the range from 1.5 µM to 32.0 µM [93].

Jinlong Yan et al. (2008) reported an electrochemical behavior of VAL in Britton–Robinson buffer solution at pH 7.0 at the Mercury Film Electrode (MFE) by cyclic, linear sweep, differential-pulse and square-wave voltammetry. The property of VAL adsorption at the MFE using accumulation potential of (+0.10V) was observed [94]. Voltammetry method is simple and there is no need of expensive grade solutions, which are needed for other analytical methods such as HPLC. The voltammetry method may possibly a good substitute for simultaneous estimation of bulk drugs.

CONCLUSION

The present review illustrates various analytical approaches exercised for the estimation of VAL. A numerous investigation had perform including, Bio-analytical, HPLC, HPTLC, UV/Vis-Spectroscopy, Spectrofluorimetry, capillary electrophoresis, electrochemical method, LC-MS, LC-ESI-MS etc. for estimation of VAL in bulk and in its combined pharmaceutical formulations and in plasma. Liquid chromatography with UV detection has been found to be most studied for estimation of VAL in bulk as well as pharmaceutical dosage forms, while hyphenated LS-MS, LS-MS/MS methods are reported for determination of VAL and its metabolite in plasma and other biological fluids. Further, methods were reported for its pharmacokinetic as well as bioequivalence studies. Few chromatography approaches like

HPTLC and Stability-indicating HPLC and HPTLC are also reported in literature. Certain Spectrophometric methods in UV-Visible along with fluorimetric are most often used for assessment for VAL.

ACKNOWLEDGEMENTS

Authors are thankful to R.C. Patel Institute of Pharmaceutical Education and Research Shirpur, Dist: Dhule (MS) 425 405 for providing necessary library facilities.

CONFLICT OF INTEREST

Authors do not have conflict of interest for this manuscript.

ABBREVIATIONS

- VAL- Valsartan
- AML- Amlodipine besylate
- HCTZ- Hydrochlorothiazide
- NIF- Nifidipine
- CIL- Cilnidipine
- EZT- Ezetimibe
- NEB- Nebivolol
- ALK- Aliskiren
- LST- Losartan
- IRB- Irbesartan
- PROP- Propranolol
- RMP-Ramipril
- NBH-Nebivolol hydrochloride
- ATV- Atorvastatin
- FLUV- Fluvastatin
- TMT-Telmisartan
- SA- Simvastatin
- KTP- Ketoprofen
- PTP-Pentaprazole
- CLTD- Chlorthalidone
- ACE- Angiotensin Converting Enzyme
- RAAS- Renin Angiotensin Aldosterone System
- LC-ES/MS/MS- Liquid chromatography electrospray-mass spectroscopy-mass spectroscopy
- GC-MS-MS- Gas chromatography- mass spectroscopy-mass spectroscopy
- LC-MS- Liquid chromatography-mass spectroscopy
- SEM- Simultaneous equation method
- RF- Retention factor

- RT- Retention time
- ESI- Electro-spray ionization
- nm-Nanometer
- M.P.- Melting point
- ACM-Absorption correction method
- ACN- Acetonitrile
- FA- Formic acid
- MFE- Mercury film electrode
- HMDE- Hanging mercury drop electrode
- CZE- Capillary zone electrophoretic
- MEKC- Micellar electro kinetic capillary chromatographic

REFERENCES

1. The Merck Index an Encyclopaedia of chemicals. (2001). *Drugs, and biological*. Merck Research Laboratories, Whitehouse station. 13th Edition, New Jersey, pp. 1705.
2. Martindale. (2005). *The Complete Drug Reference*, 34th Edition, Pharmaceutical Press, pp. 1018 & 933.
3. Wilson, & Gisvolds. (2004). *Text Book of Organic Medicinal and Pharmaceutical Chemistry*, 11th Edition. Philadelphia, PA: Lippincott-Williams & Wilkins, pp. 610- 649.
4. Sharma, H. L., & Sharma, K. K. (2012). *Principles of Pharmacology*, 2nd Edition. Delhi, India: Paras's Medical Publishers, pp. 253 & 229.
5. Saydam, M., & Takka, S. (2007). Bioavailability File, Valsartan. *Fabad J Pharm Sci., 32*, 185-96.
6. Burnier, M. & Brunner, H. R., (2000). Angiotensin II receptor antagonist. *The lancet, 355*, 637-645.
7. Siddiqui, N., Husain, A., Chaudhry, L., Alam, M. S., Mitra, M., & Bhasin, P. S. (2011). Pharmacological & Pharmaceutical Profile of Valsartan, A Review. *Journal of Applied Pharmaceutical Science, 1*, 12-19.
8. Novartis Pharmaceuticals Corp, East Hanover, NJ, pp. 1-19.
9. Ternes, T. A. (2001). Analytical Methods for the Determination of Pharmaceuticals in Aqueous Environmental Samples. *Trac Trends in Analytical Chemistry, 20*, 419-434.
10. United States Pharmacopeial Convention. Committee of Revision. (1984). The United States Pharmacopeia. United States Pharmacopeial Convention, Incorporated, 17.
11. Pharmacopoeia-Volume II, I. (2010). Published by Indian Pharmacopoeial Commission for Ministry of Health and Family Welfare, Noida, New Delhi, 1681, pp. 2226.
12. British Pharmacopoeia Commission, General Medical Council (Great Britain) and Great Britain. Medicines Commission. (2001). *British pharmacopoeia (Vol. 1). Her Majesty's Stationery Office*, pp. 6490.
13. Spooner, N. (2010). Dried blood spot sampling for quantitative bioanalysis, time for a revolution. *Bioanalysis, 2*, 1781.
14. Piao, Z. Z., Lee, E. S., Tran, H. T. T., & Lee, B. J. (2008). Improved Analytical Validation and Pharmacokinetics of Valsartan using HPLC with UV Detection. *Archives of Pharmacal Research, 31*, 1055-1059.
15. Gonzalez, O., Iriarte, G., Ferreirós, N., Maguregui, M. I., Alonso, R. M., & Jiménez, R.M. (2009). Optimization and Validation of a Spe-Hplc-Pda-Fluorescence Method for the Simultaneous Determination of Drugs Used in Combined Cardiovascular Therapy in Human Plasma. *Journal of Pharmaceutical and Biomedical Analysis, 50*, 630-639.

16. Del Rosario Brunetto, M., Contreras, Y., Clavijo, S., Torres, D., Delgado, Y., Ovalles, F., Ayala, C., Gallignani, M., Estela, J. M., & Martin, V. C. (2009). Determination of Losartan, Telmisartan, and Valsartan by Direct Injection of Human Urine into a Column-Switching Liquid Chromatographic System with Fluorescence Detection. *Journal of Pharmaceutical and Biomedical Analysis, 50*, 194-199.
17. Iriarte, G., Ferreirós, N., Ibarrondo, I., Alonso, R. M., Itxaso Maguregui, M., & Jiménez, R. M. (2007). Biovalidation of a Spe-Hplc-Uv-Fluorescence Method for the Determination of Valsartan and Its Metabolite Valeryl-4-Hydroxy-Valsartan in Human Plasma. *Journal of Separation Science, 30*, 2231-2240.
18. Koseki, N., Kawashita, H., Hara, H., Niina, M., Tanaka, M., Kawai, R., Nagae, Y., & Masuda, N. (2007). Development and Validation of a Method for Quantitative Determination of Valsartan in Human Plasma by Liquid Chromatography-Tandem Mass Spectrometry. *Journal of Pharmaceutical and Biomedical Analysis, 43*, 1769-1774.
19. Macek, J., Klima, J. & Ptáček, P. (2006). Rapid Determination of Valsartan in Human Plasma by Protein Precipitation and High-Performance Liquid Chromatography. *Journal of Chromatography B, 832*, 169-172.
20. Ramani, A. V., Sengupta, P., & Mullangi, R., (2009). Development and Validation of a Highly Sensitive and Robust LC-ESI-MS/MS Method for Simultaneous Quantitation of Simvastatin Acid, AmLodipine and Valsartan in Human Plasma, Application to a Clinical Pharmacokinetic Study. *Biomedical Chromatography, 23*, 615-622.
21. Koçyiğit-Kaymakçoğlu, B., Ünsalan, S., & Rollas, S. (2006). Determination and Validation of Ketoprofen, Pantoprazole and Valsartan Together in Human Plasma by High Performance Liquid Chromatography. *Die Pharmazie-An International Journal of Pharmaceutical Sciences, 61*, 586-589.
22. Venkata, S. P., Rama Rao, N., & Challa, B. R. (2013). Bio-Analytical Method Development and Validation of Valsartan by Precipitation Method with HPLC-MS/MS, Application to a Pharmacokinetic Study. *Journal of Chemical and Pharmaceutical Research, 5*, 7-20.
23. Kumar, P. S., Sahu, M., Prasad, K. D., & Shekhar, M. C. (2011). Development and Validation of Analytical Method for the Estimation of Valsartan in pure and Tablet Dosage Form by RP-HPLC Method. *Int. J. Res. Pharm. Chem, 1*, 945-949.
24. Vinzuda, D. U., Sailor, G. U., & Sheth, N. R. (2010). RP-HPLC Method for Determination of Valsartan in Tablet Dosage Form. *International Journal of Chemtech Research, 2*, 1461-1467.
25. Thanusha, G., Jose, C., Babu, G., Channa Basavaraj, K., Reddy, V. P., & Sharadha, C. (2010). Validated RP-HPLC Method for the Quantitative Estimation of Valsartan in Bulk and Pharmaceutical Dosage Forms. *International Journal of Chemtech Research, 2*, 1194-1198.
26. Nagakanyaka Devi Paladugu., G., Devala R., Bonthu S., & Deepthi P. (2013). Development and Validation of RP-HPLC Method for Quantification of Valsartan and its Pharmaceutical Formulations. *Int. J. Drug Dev. & Res, 5*, 199-205.
27. Nissankararao, S., Kumar, A., Sravanthi. S. L., & Naga Silpa, J. (2013). Method Development and Validation for the Estimation of Valsartan in Bulk and Tablet Dosage Forms by RP-HPLC. *Der Pharma Chemica, 5*, 206-211.
28. Kendre, M. D., & Banerjee, S. K. (2012). Precise and Accurate RP-HPLC Method Development for Quantification of Valsartan in Tablet Dosage Form. *Int. J. Pharm. Sci. Drug Res., 4*, 137-139.
29. Raju, V. B., & Rao, A. L. (2011). Reversed Phase HPLC Analysis of Valsartan in Pharmaceutical Dosage Forms. *International Journal of Chemical, Environmental and Pharmaceutical Research, 2*, 56-60.
30. Ghanty, S., Das, R., Maiti, S., & Sen, K. K. (2014). RP-HPLC Method for Estimation of Valsartan in Solid Oral Dosage Forms. *Journal of Pharma Scitech, 3*, 88-91.

31. Akiful Haque, M., Amrohi, H. S., Prashanth Kumar, K., Nivedita, G., Pradeep Kumar, T. D., Mohanty, D., & Diwan, P. V. (2012). Stability Indicating RP-HPLC Method for the Estimation of Valsartan in Pharmaceutical Dosage Form. *Iosr J Pharm., 2*, 8-12.
32. Gandla, K., Kumar, J. M. R., Rao, J. V. L. N., & Surekha, M. L. (2012). Validated RP-HPLC Method for Simultaneous Estimation of Aliskiren and Valsartan In Tablet Dosage Form. *Journal of Drug Delivery and Therapeutics, 2*, 162-166.
33. Sharma, M., Kothari, C., Sherikar, O., & Mehta, P. (2014). Concurrent Estimation of AmLodipine Besylate, Hydrochlorothiazide and Valsartan by RP-HPLC HPT-LC and UV–Spectrophotometry. *Journal of Chromatographic Science, 52*, 27-35.
34. Sheik, R. J., Puranik, M. P., Bavadkar, D. N., Mali, P. R., & Yeole, P. G. (2011). RP-HPLC Method for Simulateous Estimation of Valsartan and Hydrochlorothiazide in Solid Dosage Form. *International Research Journal of Pharmacy, 2*, 162-164.
35. Youssef, R., Hbash, A., & Hassan, A. (2014). Development and Validation of RP-HPLC Method for the Estimation and Separation of Valsartan, Losartan and Irbesartan In Bulk and Pharmaceutical Formulation. *Int J Pharm Sci Rev Res., 24*, 311-314.
36. Imam, S. S., Ahad, A., Aqil, M., Sultana, Y., & Ali, A. (2013). A Validated RP-HPLC Method for Simultaneous Determination of Propranolol and Valsartan in Bulk Drug and Gel Formulation. *Journal of Pharmacy & Bioallied Sciences, 5*, 61.
37. Rao, B. U., & Nikalije, A. P. (2014). Simultaneous Determination of Valsartan and Hydrochlorothiazide in Tablet Dosage Form by Liquid Chromatography. *African Journal of Pharmacy and Pharmacology, 8*, 953-961.
38. Chokshi, P. V., Trivedi, K. J., & Patel, N. S. (2012). Development and Validation of RP-HPLC Method for Analysis of Aliskiren Hemifumarate and Valsartan in Their Combination Tablet Dosage Form. *Int. J. Chemtech Res., 4*, 1623-1627.
39. Vignaduzzo, S. E., Castellano, P. M., & Kaufman, T. S. (2011). Development and Validation of an HPLC Method for the Simultaneous Determination of Amlodipine, Hydrochlorothiazide, and Valsartan in Tablets of Their Novel Triple Combination and Binary Pharmaceutical Associations. *Journal of Liquid Chromatography & Related Technologies, 34*, 2383-2395.
40. El-Gizawy, S. M., Abdelmageed, O. H., Omar, M. A., Deryea, S. M., & Abdel-Megied, A. M. (2012). Development and Validation of HPLC Method for Simultaneous Determination of Amlodipine, Valsartan, Hydrochlorothiazide in Dosage Form and Spiked Human Plasma. *American Journal of Analytical Chemistry, 3*, 422.
41. Kandikattu, K., Bharath Rathna Kumar, P., Devanna, N., Rama, K., & Ravi, M. (2014). Analytical Method Development and Validation of Simultaneous Determination of Amlodipine Besylate, Valsartan and Hydrochlorothiazide in Oral Dosage Form (Tablets) by RP-HPLC Technique. *Pelagia Research Library, 5*, 74-81.
42. Çelebier, M., Kaynak, M. S., Altinöz, S., & Sahin, S. (2010). HPLC Method Development for the Simultaneous Analysis of Amlodipine and Valsartan in Combined Dosage Forms and in Vitro Dissolution Studies. *Brazilian Journal of Pharmaceutical Sciences, 46*, 761-768.
43. Bhole, R. P., Pawara, V. C., Chitlange, S. S., & Wankhede, S. B. (2015). Development and Validation of HPTLC Method for Simultaneous Estimation of Cilnidipine and Valsartan in Bulk and Tablet Dosage Form. *International Journal of Pharmaceutical Chemistry and Analysis,* 2, 102-107.
44. Mathew, M. (2011). Quantitative Analysis of Valsartan in Tablets Formulations by High Performance Thin-Layer Chromatography. *Journal of Applied Pharmaceutical Science, 1*, 76.
45. Gaikwad, A. V., Rajurkar, V. G., Shivakumar, T., Dama, G. Y., & Tare, H. L. (2011). Simultaneous Estimation of Ramipril & Valsartan in Tablets by HPTLC. *Indo-Global Journal of Pharmaceutical Sciences, 1*, 99-112.

46. Singh, S. U. N. I. L., Patel, K. U. L. D. E. E. P., Agarwal, V. K., & Chaturvedi, S. H. A. S. H. A. N. K. (2012). Stability Indicating HPTLC Method for Simultaneous Determination of Valsartan and Hydrochlorothiazide in Tablets. *Int J Pharm Pharm Sci., 4*, 468-71.
47. Mrinalini, C. D., Kailash, G. B., & Kirti, S. T. (2009). Stability-Indicating HPTLC Method for Determination of Nebivolol Hydrochloride and Valsartan. *Journal of Pharmaceutical Research, 8*, 198-201.
48. Jadhav, M. L., Girase, M. V., & Tidme, S. K. (2015). Development and Validation of HPTLC Method for Simultaneous Estimation of Valsartan and Hydrochlorothiazide in Tablet Dosage Form. *Int J Pharm & Biosci., 2*, 20-25.
49. Rao, K. S., Jena, N., & Rao, M. E. B. (2010). Development and Validation of A Specific Stability Indicating High Performance Liquid Chromatographic Method for Valsartan. *Journal of Young Pharmacists, 2*, 183-189.
50. Ramachandran, S., Mandal, B. K., & Navalgund, S. G. (2014). Stability-Indicating HPLC Method for the Simultaneous Determination of Valsartan and Ezetimibe in Pharmaceuticals. *Tropical Journal of Pharmaceutical Research, 13*, 809-817.
51. Lakshmi, K. S., & Sivasubramanian, L. (2010). A Stability Indicating HPLC Method for the Simultaneous Determination of Valsartan and Ramipril in Binary Combination. *Journal of the Chilean Chemical Society, 55*, 223-226.
52. Kharoaf, M. A. H. E. R., Malkieh, N. U. M. A. N., Abualhasan, M. U. R. A. D., Shubitah, R. A. Q. I., Jaradat, N. I. D. A. L., & Zaid, A. N. (2012). Tablet Formulation and Development of a Validated Stability Indicating HPLC Method for Quantification of Valsartan and Hydrochlorothiazide Combination. *International Journal of Pharmacy and Pharmaceutical Sciences, 4*, 284-290.
53. Prasad, C. V. N., Kumari, C. S., & Sriramulu, J. (2011). A Stability Indicating RP-HPLC Method for Simultaneous Estimation of Valsartan and Atorvastatin from Their Combination Drug Product. *Int J of Pharm Res & Analy, 1*, 26-31.
54. Shaalan, R. A., Belal, T. S., El Yazbi, F. A., & Elonsy, S. M. (2013). Validated Stability-Indicating HPLC-Dad Method of Analysis for The Antihypertensive Triple Mixture of Amlodipine Besylate, Valsartan and Hydrochlorothiazide in Their Tablets. *Arabian Journal of Chemistry*.
55. Sudesh, B. M., & Uttamrao, K. S. (2009). Determination and Validation of Valsartan and its Degradation Products by Isocratic HPLC. *J. Chem. Metrl, 3*, 1-12.
56. Gupta, K. R., Wadodkar, A. R., & Wadodkar, S. G. (2010). UV-Spectrophotometric Methods for Estimation of Valsartan in Bulk and Tablet Dosage Form. *International Journal of Chemtech Research, 2*, 985-989.
57. Tarkase Kailash, N., Tajane Sachin, R1, & Jadhav Manisha B. (2012). Development and Validation of UV-Spectrophotometric Methods for Estimation of Valsartan in Bulk and Tablet Dosage Form. *Journal of Pharmacy Research, 5*, 2344-2346.
58. Patel Satish, A., & Patel Chiragkumar, B. (2016). Development and Validation of Dual Wavelength Spectrophotometric Method for Simultaneous Estimation of Valsartan and Nifedipine in Synthetic Mixture. *Human Journals Research Article, 5*, 58-67.
59. Şatana, E., Altinay, Ş., Göğer, N. G., Özkan, S. A., & Şentürk, Z. (2001). Simultaneous Determination of Valsartan and Hydrochlorothiazide in Tablets by First-Derivative Ultraviolet Spectrophotometry and LC. *Journal of Pharmaceutical and Biomedical Analysis, 25*, 1009-1013.
60. Nataraj, K. S., Ramakrishnama, C., Goud, E. S., & Saigeethika, S., & Ramanjineyulu, K. (2011). Simple Quantitative Method Development and Validation of Valsartan in Pure form and Pharmaceutical Dosage Forms by UV–Spectroscopy. *International Journal of Pharmacy and Biological Sciences, 1*.

61. Tatar, S., & Sağlik, S. (2002). Comparison of UV and Second Derivative Spectrophotometric and LC Methods for the Determination of Valsartan in Pharmaceutical Formulation. *Journal of Pharmaceutical and Biomedical Analysis, 30*, 371-375.
62. Gupta, K. R., Mahapatra, A. D., Wadodkar, A. R., & Wadodkar, S. G. (2010). Simultaneous UV Spectrophotometric Determination of Valsartan and Amlodipine in Tablet. *International Journal of Chemtech Research. 2*, 551-556.
63. Chaudhary, A. B., Patel, R. K., Chaudhary, S. A., & Gadhvi, K. V. (2010). Estimation of Valsartan and Hydrochlorothiazide in Pharmaceutical Dosage Forms by Absorption Ratio Method. *International Journal of Applied Biology and Pharmaceutical Technology, 1*, 455-464.
64. Patel, N. R., & Patel, S. K. (2012). First Derivative Spectrophotometric Method for the Simultaneous estimation of Valsartan and Hydrochlorothiazide in their Combined Dosage Form. *International Journal of Pharmacy & Life Sciences, 3*, 1828-1832.
65. Abdallah, O. M., & Badawey, A. M. (2011). Determination of Amlodipine and Valsartan in Binary Mixture using Derivative-Ratio Spectrophotometric, Chemometric and High Performance Liquid Chromatographic-UV Methods. *International Journal of Industrial Chemistry*.
66. Nikam, M. B., Dhamane, H., Aligave, A., & Kondawar, M. S. (2010). Simultaneous Estimation of Valsartan, AmLodipine Besylate and Hydrochlorothiazide by First Order Derivative UV-Spectrophotometric Method. *Int J Pharm Technol., 2*, 642-50.
67. Jadhav, M. L., Girase, M. V., Tidme, S. K., & Junagade, M. S. (2014). Development and Validation of Spectrophotometric Methods for Simultaneous Estimation of Valsartan and Hydrochlorothiazide in Tablet Dosage Form. *International Journal of Spectroscopy*.
68. Singh, S., Yadav, A. K., & Gautam, H. (2011). Simultaneous Estimation of Valsartan and Hydrochlorothiazide in Solid Dosage Form using UV-Spectroscopy. *Bulletin of Pharmaceutical Research, 1*, 10-2.
69. Mohamed, N. G. (2011). Simultaneous Determination of Amlodipine and Valsartan. *Analytical Chemistry Insights, 6*, 53.
70. Buchiya, F. V., Bhim, A. I., Raj, H. A., & Jain, V. C. (2015). Simultaneous Determination of Cilnidipine and Valsartan in Synthetic Mixture using Spectrophotometric Technique (Simultaneous Equation Method). *Asian Journal of Pharmaceutical Analysis, 5*, 21-25.
71. Ramachandran, S., Mandal, B. K., & Navalgund, S. G. (2011). Simultaneous Spectrophotometric Determination of Valsartan and Ezetimibe in Pharmaceuticals. *Tropical Journal of Pharmaceutical Research, 10*, 809-815.
72. Chitlange, S. S., Bagri, K. I. R. A. N., Wankhede, S. B., & Sakarkar, D. N. (2008). Simultaneous Spectrophotometric Estimation of Amlodipine and Valsartan in Capsule Formulation. *Oriental Journal of Chemistry, 24*, 689-692.
73. Rajesh, V., Praveen, P. S., Ramesh, D., Jagathi, V., Manohar Babu, C. H., & Devalarao, G. (2010). Simultaneous Spectrophotometric estimation of Valsartan and Ramipril by derivative method. *Int.J.Pharm. & Health Science, 1*, 132-135
74. Redasani, V. K., Kothawade, A. R., Mali, B. J., & Surana, S. J. Pelagia Research Library.
75. Kalaimagal, A., Jerad Suresh, A., & Niraimathi. V. (2012). Spectrophotometric Methods for the estimation of Valsartan in Bulk and Oral Dosage Form. *International Journal of Pharmacy and Pharmaceutical Sciences, 4*, 481-483.
76. Meyyanathan, S. N., Birajdar, A. S., & Suresh, B. (2010). Simultaneous Estimation of Nebivolol Hydrochloride and Valsartan and Nebivolol Hydrochloride and Hydrochlorothiazide in Pharmaceutical Formulations by UV- Spectrophotometric Methods. *Hypertension, 1*, 4.
77. Jothieswari, D., Anandakumar, K., Vijay Santhi, D., Vijayakumar, B., Priya, D., & Stephen Rathinaraj, B. (2010). A Validated UV-Spectrophotometric Method for Simultaneous Estimation of Amlodipine Besylate, Valsartan and Hydrochlorothiazide in Bulk and in Combined Tablet Dosage Form. *Journal of Pharmaceutical and Biomedical Science, 5*, 1-5.

78. Mohammed, T. A. E. F. (2015). Native and Synchronous Spectrofluorimetric Methods for Simultaneous Determination of Amlodipine Besylate and Valsartan Combination in Tablets. *Asian Journal of Science and Technology, 6*, 1690-1698.
79. Shaalan, R. A., & Belal, T. S. (2010). Simultaneous Spectrofluorimetric Determination of Amlodipine Besylate and Valsartan in their Combined Tablets. *Drug Testing and Analysis, 2*, 489-493.
80. Ferreirós, N., Dresen, S., Alonso, R. M., & Weinmann, W. (2007). Validated Quantitation of Angiotensin Ii Receptor Antagonists (Ara-Ii) In Human Plasma by Liquid-Chromatography-Tandem Mass Spectrometry, Using Minimum Sample Clean-Up and Investigation of Ion Suppression. *Therapeutic Drug Monitoring, 29*, 824-834.
81. Gonzalez, O., Iriarte, G., Rico, E., Ferreirós, N., Maguregui, M. I., Alonso, R. M., & Jiménez, R. M. (2010). LC–MS/MS Method for the Determination of Several Drugs Used In Combined Cardiovascular Therapy in Human Plasma. *Journal of Chromatography B., 878*, 2685-2692.
82. Levi, M., Wuerzner, G., Ezan, E., & Pruvost, A. (2009). Direct Analysis of Valsartan or Candesartan in Human Plasma and Urines by On-Line Solid Phase Extraction Coupled to Electro-spray Tandem Mass Spectrometry. *Journal of Chromatography B, 877*, 919-926.
83. Li, H., Wang, Y., Jiang, Y., Tang, Y., Wang, J., Zhao, L., & Gu, J. (2007). A Liquid Chromatography/Tandem Mass Spectrometry Method for the Simultaneous Quantification of Valsartan and Hydrochlorothiazide in Human Plasma. *Journal of Chromatography B, 852*, 436-442.
84. Lu, C. Y., Chang, Y. M., Tseng, W. L., Feng, C. H., & Lu, C. Y. (2009). Analysis of Angiotensin-II Receptor Antagonist and Protein Markers at Microliter Level Plasma by LC–MS/MS. *Journal of Pharmaceutical and Biomedical Analysis, 49*, 123-128.
85. Selvan, P. S., Gowda, K. V., Mandal, U., Solomon, W. S., & Pal, T. K. (2007). Simultaneous Determination of fixed Dose Combination of Nebivolol and Valsartan in Human Plasma by Liquid Chromatographic-Tandem Mass Spectrometry and its Application to Pharmacokinetic Study. *Journal of Chromatography B, 858*, 143-150.
86. Gonzalez, O., Alonso, R. M., Ferreirós, N., Weinmann, W., Zimmermann, R., & Dresen, S. (2011). Development of an LC–MS/MS Method for The Quantitation of 55 Compounds Prescribed in Combined Cardiovascular Therapy. *Journal of Chromatography, 879*, 243-252.
87. Mehta, S., Shah, R. P., & Singh, S. (2010). Strategy for Identification and Characterization of Small Quantities of Drug Degradation Products using LC and LC-MS Application to Valsartan, a Model Drug. *Drug Testing and Analysis, 2*, 82-90.
88. Hillaert, S., & Van den Bossche, W. (2003). Simultaneous determination of hydrochlorothiazide and several angiotensin-II-receptor antagonists by capillary electrophoresis. *Journal of pharmaceutical and biomedical analysis, 31*, 329-339.
89. Aslan, N., Erden, P. E., Canel, E., Zeybek, B., & Kiliç, E. (2010). Potentiometric determination of valsartan in a pharmaceutical preparation and its protonation constants. *Asian Journal of Chemistry, 22*, 4010.
90. Patil, S. H., & Janjale, M. V. (2012). Novel and validated titrimetric method for determination of selected angiotensin-II-receptor antagonists in pharmaceutical preparations and its comparison with UV spectrophotometric determination. *Journal of Pharmaceutical Analysis, 2*, 470-477.
91. Ramadan, N. K., Mohamed, H. M., & Mostafa, A. A. (2012). Potentiometric Determination of Amlodipine Besilate and Valsartan Using Microsized and Polymeric Matrix Membrane Sensors. *Portugaliae Electrochimica Acta, 30*, 15-29.
92. Habib, I. H. I., Weshahy, S. A., Toubar, S. S., & El-Alamin, M. M. A. (2008). Stripping voltammetric determination of valsartan in bulk and pharmaceutical products. Die Pharmazie-*An International Journal of Pharmaceutical Sciences, 63*, 337-341.

93. Erden, P. E., Taşdemir, İ. H., Kaçar, C., & Kılıç, E. (2014). Simultaneous Determination of Valsartan and Amlodipine Besylate in Human Serum and Pharmaceutical Dosage Forms by Voltammetry. *Int. J. Electrochem. Sci., 9*, 2208-2220.
94. Yan, J., Wang, W., Chen, L., & Chen, S. (2008). Electrochemical behavior of valsartan and its determination in capsules. *Colloids and Surfaces B, Biointerfaces, 67*, 205-209.

http://iserjournals.com/journals/ejac