Akhil. M.B et al /J. Pharm. Sci. & Res. Vol. 13(8), 2021, 457-468

ISSN:0975-1459
Journal of Pharmaceutical Sciences and Research
www.jpsr.pharmainfo.in

Review on Analytical Techniques for the Estimation of Pregabalin and Etoricoxib in Combined Dosage Form

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Abstract

Fixed dose combinations refer to the products which are combined in one dosage form and contain multiple active ingredients. Due to various benefits, FDCs are justified. These are a) Potential therapeutic effectiveness, b) reduction of adverse effects, c) pharmacokinetic advantages, d) minimizing pill burden, e) minimizing dose of individual drugs and f) avoiding drug resistance. Pregabalin IP (75mg) and Etoricoxib IP (60mg) is a recently approved fixed dose combination for treating chronic neuropathic pain. Effective quality control, pharmacodynamic and pharmacokinetic studies are essential for effective chemical and pharmaceutical analysis. There are several analytical methods for the estimation of these individual drugs, but not for this combination drug. Aim of this survey was to analyse the instrumental methods developed so far for the quantitative estimation of these individual drugs. This survey will be useful to develop a specific method for the analysis of this fixed dose combination of Pregabalin and Etoricoxib. Information's from various articles are collected and coordinated.

Key words: Pregabalin, Etoricoxib, Analytical methods, Fixed dose combination, Review

INTRODUCTION

Neuropathic pain is a class of ongoing agony that originated from the pathology of the sensory system, which is generally normal, crippling, exorbitant, and hard to treat. It is the consequence of different pathways at the fringe, spinal and supra-spinal levels that trigger agony conduction pathway changes. A blend treatment containing drugs with different instruments of activity and targets shows up a normal way to deal with disposing of such condition. A bilayered uncoated FDC tablet containing Pregabalin IP (75 mg) and Etoricoxib (60 mg) has as of late been authorized by Central Medications Standard Control Organization (CDSCO) to consider synergistic consequences for the administration of ongoing back pain related with neuropathic segments.

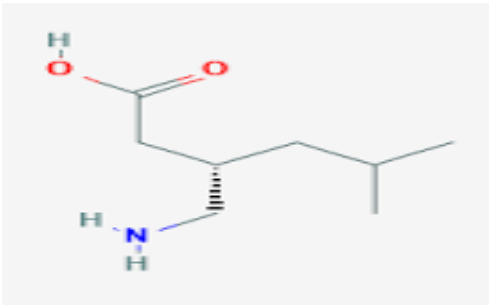


Fig 1. Structure of PGB

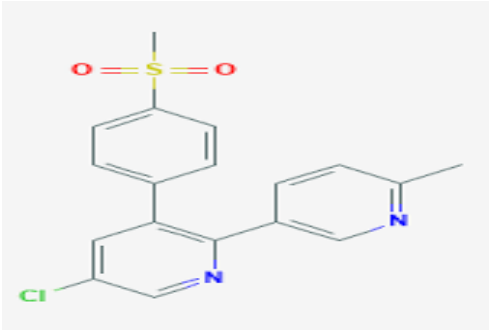


Fig 2. Structure of ETC

Pregabalin, a functioning (S)- enantiomer 3-(aminomethyl)- 5-methylhexanoic corrosive, got endorsement in May 2005 for 40 nations. It has been shown for use as an adjunctive treatment for halfway beginning seizures and in the administration of neuropathic torment associated with difficult diabetic neuropathy and postherpetic neuralgia. It is known as the racemic 3-isobutyl GABA pharmacologically dynamic S-enantiomer. Eight diverse capsular qualities, going from 25 to 300 mg, are accessible for the product. Etoricoxib is a profoundly specific COX-2 inhibitor that was endorsed by EMA in 2002 and is presently accessible in excess of 80 nations because of its mitigating, pain relieving and hostile to pyretic properties. In the therapy of osteoarthritis, rheumatoid joint inflammation, ankylosing spondylitis and gouty arthritis, it is suggested for intense and ongoing therapy. It is likewise valuable in the counteraction and therapy of malignancy, just as in the therapy of essential dysmenorrhea and pain.

Physiochemical properties

Pregabalin

It is white to grey glass like strong powder and officially named as (3S)- 3-(aminomethyl)- 5-methylhexanoic acid. It is a structurally comparative gabapentin compound however is viewed as the most dynamic replacement to it. It has the CAS number 148553-50-8 and Figure 1 shows its atomic structure. Its molecular recipe is C8H17NO2 with a sub-atomic load of 159.23 g/mol. It has a pKa1 (4.2) and a pKa2 (10.6). It is promptly solvent in water and in watery arrangements that are both fundamental and acidic. It has a 186–188 ○C liquefying point and a 274 ○C limit at 760 mm Hg. It is a BCS class I (high permeability, high solubility) drug.

Etoricoxib

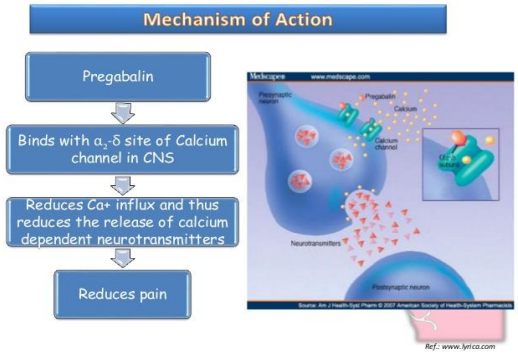
Etoricoxib is a NSAID that comes under the low solubility and high permeability BCS Class II medication classification. It is specific second-age COX-2 inhibitor. It is 5-chloro-2-(6-methylpyr-idin-3-yl)- 3-(4-methylsulfonyl

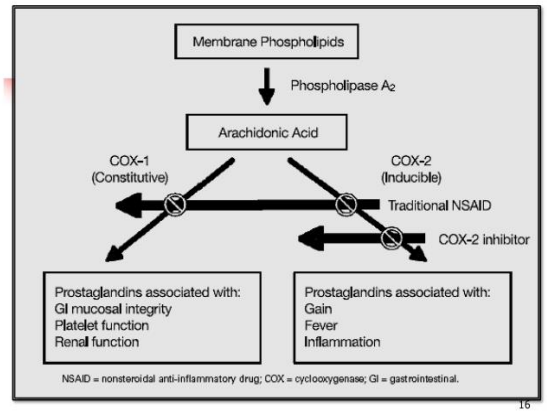
phenyl) pyridine as appeared in Figure 2 and has CAS number 202409-33-4. It has a molecular formula, C18H15ClN2O2S atomic recipe and a sub-atomic load of 358.8419 g/mol. It has an unpleasant taste and accessible as a white to grey powder. It has an immersion dissolvability of 78.48 ± 1.47 mg/mL. It is basically insoluble in water and, in basic fluid arrangements, promptly dissolvable. It shows pH-subordinate dissolvability (high gastric pH solvency, low pH and diminishing pH-expanding dissolvability). It has log P and pKa values 3.9 and 4.6, respectively.

Mechanism of Action

Pregabalin

Pregabalin's system of activity varies from the activity of other antiepileptic and pain-relieving restorative drugs. Pregabalin acts on voltage-gated calcium channel, binds to alpha2-delta subunit at neuronal ending of presynaptic region of spinal cord and brain. It regulates hyperexcited neurons through authoritative to presynaptic neurons of voltage-gated calcium channels which minimize calcium flood into presynaptic terminals. Over the top arrival of excitatory neuro-transmitters (e.g., glutamate, substance P, noradrenaline) diminishes the decreased calcium stream. It accordingly mitigates neuropathic pain and is liable for pregabalin's anxiolytic, anticonvulsant, and pain-relieving capacity.





Etoricoxib

Etoricoxib is a NSAIDs that are COX-2-particular (COXIB) inhibitors. Like some other specific COX-2 inhibitor, cyclooxygenase (COX-2) isoform 2, a compound that is engaged with pain and irritation, is specifically inhibited. It has 106-fold selectivity to COX-2 over COX-1. This diminishes the production of Pgs. In this manner, it has anti-inflammatory, pain relieving and antipyretic impacts.

Pharmacokinetics

Pregabalin

PGB shows rapid absorption on oral administration and has bioavailability around 90 percent within 1.5 hr. By following a repeated administration, steady state concentration is reached within 24-48 hours. C max is directly proportional to administered dose. Administration along with food shows a reduced absorption of about 25-30 percent and increase in Tmax around 3 hours. Absorption of PGB takes place in ascending colon. PGB has a 6-hour half life, and has no plasma protein binding. PGB can rapidly penetrate blood-brain barrier and has impact on activity of CNS. Pregabalin eliminated as N-methylated derivative of PGB through renal excretion. It shows a dose independent elimination and about 98% in unchanged form. Daily dose is been selected based on renal functions of patients. Clinical trials showed that there is no effect of sex or race in pharmacokinetics of the drug.

Etoricoxib

ETX is mainly given by oral route and is rapidly absorbed from GI tract. It has almost 100% bioavailability and elimination by urinary excretion. It has a 22-hour half life. Shows a high rate of protein binding of about 92% and has an apparent V_d of about 120 L. It shows a proportional increase in AUC to Oral administration of 5 and 120 mg. Patients with liver impairment shows increased AUC by 40% when compared with healthy individuals. No significant effects on bioavailability are shown when it is taken along with food. Metabolism of the drug is by liver. Metabolism is aided by the oxidation of ETX by cytochrome P450 enzymes. Upon oxidation it is converted to etoricoxib 6-hydroxymethyl derivatives and further to 6-carboxylic acid derivatives. These two metabolites are inactive and easily eliminated. Even though eliminated mainly through urine, 20% is through faeces.

Quantitative Analytical Methods

Quantitative analytical methods used to quantify each component present in the sample.

HPLC

It can be used for the estimation of active ingredients and related substance using different solvents, columns and detectors. It offers a consistent result with high degree of accuracy and precision. HPLC can be fully automated. It offers an excellent replicability and is applied by careful selection of HPLC column to determine wide variety of compounds. Chiral analysis can also be performed using HPLC and able to isolate the enantiomeric forms. Due to polar nature of most drugs, reverse phase HPLC are commonly used for their quantitative analysis. On the basis of the column stationary phase, the separation aided by absorption, ion-exchange, division or exclusion.

Table 1 : Analytical methods using HPLC

Sample matrix	Drug	Stationary phase, Mobile phase	Linearity(µg/ml)	Wavelength (nm)	Reference
Tablet	Pregabalin, Amitriptyline Hydrochloride	Phenomenex Luna C18 column, buffer (potassium dihydrogen phosphate) pH 4.0 and acetonitrile, (40:60 %v/v)	30-105 µg/mL and 4-14 µg/mL	230	1
Bulk and Dosage form	Pregabalin, Epalrestat	C18 column discovery (250mm × 4.6mm, 5 µ particle size) 0.01M Potassium dihydrogen phosphate buffer: Methanol (25:75% v/v)	15-90 µg/mL and 30-180 µg/mL	226	2
Capsule	Pregabalin	Gemini C18(50 × 4.6mm) column with 3 µm particle size, 0.2% of triethylamine in a mixture of methanol and water (10:90, % v/v)	0.1-25 µg/mL	fluorescent detection (λex 395 nm, λem 476 nm)	3
Bulk and Capsule	Pregabalin, Methylcobalamine	Phenomenex C18 column. acetonitrile: methanol: ammonium acetate(v/v/v)	5-50 µg/mL, 10-60 µg/mL	234	4
Capsule	Pregabalin	C18 column 10 mM NH4OAC in water (pH-6.8) and acetonitrile and methanol (80:20 % v/v)	150µg/ml to 850 µg/ml	210	5
Human plasma	Pregabalin	C8 column acetate buffer (pH 4.6): ACN (50:50% v/v)	2.8–8.2 mg/L	340	6
Bulk and Dosage form	Pregabalin	Nova-Pak C18 column ACN and sodium dihydrogen phosphate (pH 2.5) (60:40, % v/v)	1-100 µg/mL	360	7
Bulk and combined dosage form	Nortriptyline and pregabalin	BDS C18 column. Perchloric acid (0.1%) and ACN (55:45 % v/v).	5-30 µg/mL and 37.5-225 µg/mL	210	8
Tablet	Pregabalin and Nortriptyline	LC- 20 AT C18 column. Phosphate Buffer (pH 5.0): Methanol (70:30, % V/V)	5-15 µg/mL and 37.5-112.5 µg/mL	210	9
Pharmaceutical and Bulk Formulation	Pregabalin	C18 5 µm BDS Hypersil column using phosphate buffer solution (pH 6.9) and acetonitrile in the ratio of 94:6 % v/v	0.60 - 0.89 µg/mL	210	10
Tablet	Pregabalin	Phenomenex C18 column (150 X 4.6 mm Id, ODS 2, 5µm). Methanol & 10mM Ammonium Acetate (pH adjusted to 3.0 with glacial acetic acid) 50:50 % v/v	249.25 – 7976.00 µg/mL	210	11
Capsule	Pregabalin, Methylcobalamine	C18 column ammonium dihydrogen-o-phosphate (buffer 6.0), ACN and methanol in the ratio of 75:15:10 % v/v/v	3200-4800 mcg/ml and 16-24 mcg/ml	210	12
Capsule	Pregabalin	Hypersil BDS, C8 column Phosphate buffer (pH 6.9) and ACN (95:5 % v/v)	50-150 µg/mL	200	13

Sample matrix	Drug	Stationary phase, Mobile phase	Linearity(µg/ml)	Wavelength (nm)	Reference
Bulk Formulation Urine	Pregabalin	C18 5 µm ODS Hypersil column, Methanol: ACN - 0.02 M di - potassium hydrogen orthophosphate (pH - 7.00) (3: 1: 16, v/v/v)	0.75 - 6.00 µg/mL	210	14
Bulk, Tablet	Pregabalin, Celecoxib	Hypersil BDS potassium di hydrogen orthophosphate buffer of pH 6.5 and ACN in the ratio of (70:30 % v/v)	37.5 µg/mL-281.25 µg/mL for Pregabalin, 100 µg/ml -750 µg/ml Celecoxib	238	15
Human Plasma, Bulk	Pregabalin	C18 Column, Disodium hydrogen phosphate (pH 8.0) - methanol (35: 65 % v/v)	1–4500 ng/mL	360	16
Tablet	Etoricoxib, Paracetamol	Kromasil C18 column Buffer: ACN (75: 25 % v/v)	48 to 146 µg/mL of Paracetamol and 6 to 19 µg/mL for Etoricoxib	220	17
Pharmaceutical Dosage forms	Etoricoxib, Paracetamol	Phenomenex® C18, 5µm, 250mm X 4.6mm i.d. column, ACN, methanol and water in the proportion of 60:15:25 (v/v/v)	8.3-41.5 µg/mL for PCT and 1-5 µg/mL for ETX	236	18
Bulk, Tablet	Etoricoxib, Paracetamol	Inertsil ODS, 5µ, C8-3 column, with a mobile phase consisting of methanol: ACN : phosphate buffer pH 3.5 (40:20:40 v/v)	50 to 150 µg/mL for Paracetamol and 6-18µg/mL Etoricoxib	242	19
Bulk	Etoricoxib, Thiocolchicoside	BDS Hypersil C-18 column (250 mm x 4.6 mm, 5-µm particle size) Trifluoroacetic acid buffer (pH 2.6) and ACN (75:25, v/v)	20-160 µg/mL for etoricoxib and 2-16 µg/mL for thiocolchicoside	220	20
Tablet	Etoricoxib	Hypersil ODS C-18 column, ACN and potassium dihydrogen phosphate buffer (46:54 % v/v)	0.5-85 µg/mL	280	21
Tablet, Bulk	Etoricoxib	Hyper ODS 2 C18 Column, HPLC Methanol	20- 55 µg/mL	233	22
Tablet, Bulk	Etoricoxib	C18 column, ACN : Ammonium Acetate buffer	24.6 – 74.5 µg/mL	235	23
Tablet	Etoricoxib	ODS Hypersil C18 column, ACN: water (55:45 % v/v)	10 to 60 µg/mL	269	24
Human Plasma	Etoricoxib	ODS Hypersil C18 column, Buffer (0.3 ml triethylamine and 0.4 ml orthophosphoric acid): ACN (62:38 % v/v)	15-3200 ng/mL	284	25
Tablet	Pregabalin, Aceclofenac	Hiq Sil C18HS Methanol: Phosphate buffer (70:30% v/v)	5-25 µg/mL for PGB and ACE	248	26

HPTLC

High-performance thin-layer chromatography is a modified and advanced form of TLC. Certain enhancements are made on the basic TLC to make automatic and to improve the resolution. By automation, the uncertainty in droplet size and position can be avoided. HPTLC is fast and have a flexible separation process, so can be used for the large sample analysis. It requires a short time for analysis and can be easily managed. It can used to perform both quantitative and qualitative analysis. Multiple samples can be handled at a time. Large theoretical plate count can be achieved in minimum area of plates. HPTLC shows high efficiency due to small particle size less than 5 µm.

Table 2 : Analytical methods using HPTLC

Sample Matrix	Drug	Mobile Phase	Linearity	Wavelength	Reference
Pharmaceutical Dosage Forms	Pregabalin, Gabapentin	Methanol: Ethyl acetate :Ammonia (4:6:1 v/v)	2-12 ng / ml	210	27
Bulk	Pregabalin, Aceclofenac	Toluene: Methanol: Formic acid (7: 3: 0.2 v/v/v)	100-600 ng/band FOR ACE and 75 - 450 ng/band for PGB	210	28
Capsule	Pregabalin, Methylcobalamine	methanol: toluene: ammonia (30%) (8:2:0.4 % v/v/v)	1500-7500 ng/band for PRG, 150-750 ng/ band based for MCA	497	29
Tablet	Etoricoxib	Chloroform: Methanol: Toluene (4:2:4 %v/v/v/)	100-600 ng/spot	289	30
Bulk and Formulation	Etoricoxib	toluene–1,4-dioxane–methanol 8.5:1.0:0.5 (v/v)	100-1500 ng/spot	235	31
Bulk, Tablet	Etoricoxib, Paracetamol	toluene: ethyl acetate: methanol - 6: 4: 1 (v/v/v)	60360 ng/spot for PCT and 50-300 ng/spot for ETX	263	32
Tablet	Etoricoxib, Thiocolchicoside	ethyl acetate–methanol (8:2 v/v)	50–250 for ETX and 100–500 ng/band for TCL	290	33
Human Plasma	Paracetamol, Etoricoxib	Toluene: Dichloromethane: Methanol (6:2.5:1.5 v/v/v)	100 to 600 ng/band	240	34

UPLC

Ultra-performance liquid chromatography is performed for samples having particle size less than 2 µm with better resolution, speed and sensitivity than high performance liquid chromatography. UPLC need a high-pressure system than HPLC. Solvent consumption and time required is very less compared to other techniques. It offers an enhanced product consistency.

Table 3 : Analytical methods using UPLC

Sample Matrix	Drug	Column, Mobile Phase	Linearity	Wavelength	Reference
Bulk, Tablet	Pregabalin	HSS column 0.1% o-phosphoric acid- buffer and CAN (55:45 % v/v)	37.5–225 µg/mL for Epalrestat and Pregabalin	210	35
Blood, Urine	Pregabalin	n HSS T3 column (2.1 mm id × 100 mm, dp = 1.7 m) 0.2% formic acid (A) and methanol (B	Up to 95 mg/L in blood and 1.3g/L in urine	210	36
Tablet	Pregabalin, Aceclofenac	HSS C18 column (100mm x 2.1mm, 1.8µ) 0.05 M phosphate buffer (pH- 6.2): Methanol: ACN (55: 30 :15 v/v)	100–600 µg/mL for ACE and 38–225 µg/mL for PGB	218	37
Human Plasma	Etoricoxib	HSS T3 column (1.8 µm, 50 × 2.1 mm), MP- ACN and water which contained 2 mM ammonium acetate	5-5000 ng/ml	210	38
Bulk	Etoricoxib	BEH C18 column (1.7 Å, 2.1 x 100 mm) 0.01M acetate buffer pH 5.0 - ACN (60: 40, v/v)	0.05 - 120 µg/mL	235	39

UV-Visible Spectroscopy

Ultraviolet–vision spectroscopy (UV–Vis or UV/Vis) means absorption spectroscopy or reflection spectroscopy in the ultraviolet and visible region. Atoms and molecules undergo electronic transitions in this area of the electromagnetic spectrum. Molecules contains bonding and non-bonding electrons which can absorb UV and Visible radiations. These cause excitation to higher anti-bonding orbitals. Absorption spectroscopy complements to fluorescence spectroscopy by handling the transition from excited to ground state.

Table 4 : Analytical methods using UV-Visible Spectroscopy

Sample matrix	drug	Diluent and colouring reagent	Linear Range (µg/ml)	λ_{max} (nm)	Reference
Pharmaceutical dosage form	Pregabalin	potassium iodate (KIO3) in presence of sulphuric acid medium	20-240 µg/mL	600	40
Pharmaceutical dosage form	Pregabalin and Tranexamic Acid	2,4-dinitrophenol and 2,4,6-trinitrophenol	0.02–200 µg/mL	PGB- 418 , TXA- 425	41
Bulk, Capsule	Pregabalin	Water	5.0–50 µg/mL	530	42
Bulk, Capsule	Pregabalin	Methanol	2.5-12.5 µg/mL	223	43
Capsule	Pregabalin	Distilled Water	5- 45 µg/mL	485	44
Bulk, Capsule	Pregabalin	Methanol	5–60 µg/mL	395.8	45
Human Urine, Distilled Water	Pregabalin	Distilled Water	0.5-5 µg/mL	210	46
Bulk, Capsule	Pregabalin	Phosphate buffer pH (7.4)	50-1000 µg/mL	402.6	47
Bulk, Capsule	Pregabalin	NaOH, Deionized Water	2-10 µg/mL	385	48
Capsule	Pregabalin	Distilled Water	2-18 µg/ml	365	49
Capsule	Pregabalin	2,3-dichloro-5,6-dicyano-1,4-benzoquinone and 7,7,8,8-tetracyanoquinodimethane	2.0—30.0 and 1.5—10 µg/mL for DDQ and TCNQ	494 and 841 for DDQ and TCNQ	50
Bulk, Capsule	Pregabalin, Methylcobalamine	Distilled water	2-10µg/mL	351 for MCA, 617 for PGB	51
Tablet, Bulk	Etoricoxib, Thiocolchicoside	0.1N HCl	2.5-30.0 µg/mL for both	240,260	52
Tablet	Etoricoxib	0.1 N HCl.	2-20 µg/mL	416	53
Tablet	Etoricoxib	0.1 N HCl.	2-24 µg/mL	233	54
Tablet	Etoricoxib	0.1 N HCl.	0.1-0.5 µg/mL	233	55
Bulk, Tablet	Drotaverine and Etoricoxib	Methanol	4-20 µg/mL - DRT 4.5-22.5 µg/mL- ETX	274 and 351 nm for ETX and DRT	56

LC-MS Technique

LC-MS is an advanced technique which associates the physical separation property of LC with the mass analysis abilities of mass spectrometry. Individual abilities of these two techniques are enhanced when using synergistically. Liquid chromatography system separate combined drugs and the mass spectrometry system helps for structural identification of individual components. Biochemical, organic, inorganic compounds found in samples of biological and environmental origin can be analysed using LC-MS. It has persistently been used in the development of drugs at several different levels, including detection of metabolite, live drug screening, impurity identification, mapping of peptide and glycoprotein etc.

Table 5 : Analytical methods using LC-MS

Sample Matrix	Drug	Detection System	MP/Reagent	Linearity	Reference
Rat Plasma	Pregabalin, Sildenafil	solid-phase extraction followed by LC–MS–MS	1 mL of methanol, 1 mL of 1 M monochloroacetic acid in 90:10 (v/v) water/methanol	pregabalin (70–10,000 ng/mL, sildenafil (1–2,000 ng/mL)	57
Human Plasma, Tablet	Etoricoxib	positive electrospray ionization in multiple reaction monitoring mode	acetonitrile: water (95:5)/0.1% acetic acid (90:10, v/v).	1–5000 ng/mL	58
Human Plasma	Etoricoxib	Sciex API triple quadrupole mass 18 spectrometers having turbo ion spray source	acetonitrile–water (90:10% v/v)	0.2–200 ng/mL	59
Dried blood & plasma spots	Pregabalin	Positive ion mode by 37 applying two SRM transitions	ACN: formic acid (85: 15, v/v)	0.200-20.0 µg/mL for DBS and 0.400 – µg/mL for DPS	60
Human Plasma	Pregabalin	Triple quadrupole mass spectrometer	Methanol: water with v/v formic acid(98:2, v/v)	0.1–15.0 µg/mL	61
Rat Plasma	Pregabalin	+ ve ionization mode	0.1% formic acid: methanol (40:60, v/v)	0.50–20000.00 ng/mL	62
Blood, Urine	Pregabalin	HILIC technique with a high-resolution mass spectrometer	Methanol: deionized water (50: 50% v: v)	In peripheral blood - 0.4 to 17.0 µg/mL, in central blood - 1.5 to 11.1 µg/mL, in urine 126.6 to 2004.6 µg/mL and in bile -10.5 to 58.3 µg/mL	63
Human Plasma	Pregabalin	high-performance LC coupled with electrospray tandem MS	Methanol and water 60:40 (v/v)	10.0–10000.0 ng/mL	64
Human Plasma	Pregabalin	high-performance LC +ve ion APCI tandem MS method	N, N-dimethylacetamide and methanol (10:90, v/v)	1–10,000 ng/m	65
Blood	Pregabalin, Gabapentin	combination of liquid chromatography time-of-flight mass spectrometry (LC-TOF-MS) and liquid chromatography tandem mass spectrometry (LC-MS-MS)	water and acetonitrile, both with additions of 0.1% formic acid	0.5–50 mg/L	66
Hair	Pregabalin	ultra-high-performance liquid chromatography coupled with tandem mass spectrometry	Solvent A (95: 5 H2O/acetonitrile + 0.5% formic acid) and Solvent B (acetonitrile + 0.5% formic acid)	540 pg/mg	67
Human Plasma	Etoricoxib, Valdecoxib	RP-HPLC coupled to atmospheric pressure chemical ionisation (APCI) mass spectrometry (Finnigan Mat LCQ ion trap)	methanol: water (50:50, v/v) and 1% acetic acid	10–2500 for ETX and 5–1000 µg/L for VAL	68

Capillary Electrophoresis

CE is commonly used to analyse complex sample. Separation is aided by the difference in the charge to size ratio. Since the charge of ions dependent on the pH, a buffer at a certain pH is used for separation. If two ions have same charge, then movement is based on size of ions. It gives quicker results with high resolution separation. Routine capillary electrophoretic analysis and latest advances in metabolomic methods are explored for profiling smaller molecules present in biological samples.

Table 6 : Analytical methods using CE

Sample Matrix	Drug	Buffer	Linearity	Reference
Bulk, Tablet	Etoricoxib	25 mM tris-phosphate solution at pH 2.5	2 – 150 µg/mL	69
Urine	Pregabalin	10 mM ammonium formate- 0.05% acetic acid in methanol	0.1 to 100 µg/mL	70
Pharmaceutical Dosage form	Pregabalin	100 mM sodium phosphate (pH 2.5), 40 mg mL−1 heptakis(2,3,6-tri-O-methyl)-β-cyclodextrin	0.05–1.0%	71
Serum	Pregabalin	5-aminosalicylic acid , cetyl trimethylammonium bromide, 1 mmol L-1) and tri-sodium citrate	1.5-100 µg/mL	72
Pharmaceutical Dosage form	Pregabalin, Gabapentin	ACN	0.1–10 µg/mL for PGB and 0.1-20 µg/mL for GBP	73
Bulk	Pregabalin	80 mM borate, 50 mM phosphate, 50 mM and 15-mM phosphate	-	74

Electrochemical Method

It concerns with the collaboration of chemical and electrical effects. The electric current passage can cause chemical changes and the electrical energy produced by the chemical reactions are studied in this method. To determine the analyte concentration and its chemical reactivity, these analytical techniques measure potential, charge or current. This method will be helpful to gain thermodynamic data about a reaction and to analyze a solution for trace amount of metal ions and organic species. This method is originated from the study of the electron movement in oxidation-reduction reactions.

Table 7 : Analytical methods using Electrochemical methods

Sample Matrix	Drug	Method	Linearity	Reference
Biological samples, Dosage form	Pregabalin	square wave voltametric method	5.0 ×10-8 –1.0 ×10-6 mol/L	75
Plasma, Capsule	Pregabalin	Ion selective electrode	10^{-6} – 10^{-3} M	76
Pharmaceutical formulations	Pregabalin	Potentiometric determination	1.0×10^{-6} to 1.0×10^{-1} M	77

Infrared spectroscopy

The IR radiation interactions with the matter by absorption, reflection and emission is studied using IR spectroscopy. It is related to the vibrational, rotational energy of molecule. It is used in analysis and detection of chemical substance and functional groups in solid, liquid, and gaseous form. Molecules absorbs IR radiation when the IR radiation frequency is equal to natural frequency of vibration. The change in vibrational energy depends on mass of atoms, strength of bonds and atomic arrangement in molecule. No two compounds have similar IR spectrum except the enantiomers. Wave number vs % transmittance is plotted in IR spectrum.

Table 8 : Analytical methods using IR

Sample Matrix	Drug	System	Detection Form	Reference
Bulk	Pregabalin	Ionotropic gelation followed by FTIR analysis	pure drug, polymers and formulations were dispersed in KBr powder	78
Tablet	Pregabalin	Ionotropic gelation followed by FTIR analysis, DSC and XRD	drug/polymer/microspheres with KBr	79
Bulk	Etoricoxib	FTIR spectrophotometer and DSC	applying the KBr disk technique	80
Pharmaceutical dosage form	Etoricoxib and diclofenac	FTIR and NMR Spectroscopic techniques	Drug, KBr (5:95)	81
Pharmaceutical dosage form	Etoricoxib	FTIR	Coupling of folic acid with nanoparticle	82
Dosage form	Etoricoxib	Diffuse reflectance IR Fourier transform spectroscopy, DSC, radiograph powder diffraction, hot stage microscopy and dissolution studies	sample was mixed with dry potassium bromide	83

Sample Matrix	Drug	System	Detection Form	Reference
Dosage form	Etoricoxib	DSC, IR spectroscopy, powder X-ray diffractometry and microscopic study	samples in KBr using FTIR-8400S Software- IR Solution	84
Dosage form	Etoricoxib	FTIR and XRD analysis	samples prepared in KBr using FTIR, IR solution software	85
Dosage form	Etoricoxib	FTIR spectroscopy, DSC, XRD	KBr pellets by using a FTIR spectrometer	86
Dosage form	Pregabalin and Tramadol	analysis using GC-LC with the MS detector and IR spectrophotometry	IR spectral analysis was performed on Agilent Technology FTIR-640 IR spectrometer using an ATR attachment	87
Tablet	Pregabalin	FTIR spectrometry	2 mg sample in 200 mg KBr	88
Tablet	Pregabalin	FTIR, XRPD, DSC & TGA	Nicolet iS10	89

CONCLUSION

In this review article, various analytical techniques used for the study of newly marketed combination of Pregabalin and Etoricoxib are been discussed. For better chemical and pharmaceutical analysis of drugs effective quality control, pharmacokinetic and pharmacodynamic studies are essential. In this work, analysis of these combination using different instruments like HPLC, HPTLC, UV-Visible spectroscopy, IR, Capillary electrophoresis, LC-MS and electrochemical methods are recorded. HPLC is the most used instrument for analysis of pharmaceutical and LC-MS is used for the examination of samples present in the biological matrices. These techniques are applicable for the detection and estimation of active ingredients in bulk, pharmaceutical dosage form, biological matrices and sample analysis in invitro dissolution studies, stability indicating studies, isomerism studies, fragmentation studies, biological activity studies etc.

Due to high separation ability, selectivity, and sensitivity, RP-HPLC with UV-detection is the most used analytical technique for determination of Pregabalin and etoricoxib in combined form. HPLC, however, is still to be identified with other detector systems, including fluorescence, electrochemical, capillary, and LC/MS. The need to develop a stability-specific evaluation system (SIAM) has become clearer with the guidelines of the ICH. The guidelines require specifically that forced decomposition studies be carried out in a variety of stress conditions such as pH, illumination, oxidation, dry heat, etc., and that drugs be isolated from degradation products.

Using IR and Mass spectroscopic techniques, it is helpful to find out structural activity characterization of drug, fragmentation pattern, Co-crystal arrangement, screening of characteristic degradation product, and impurity detection. Finally, the testing of the drugs in pharmaceuticals and biological matrices are possible with a wide range of techniques. The investigation study of the methods showed that for the determination of the drug in both pharmaceutical and biologic matrices, liquid chromatographic system with mass and tandem mass spectrometer detector systems are more extensively used.

Acknowledgment

Authors are very thankful to Grace College of Pharmacy and Bharathiar University to provide necessary library facilities to collect necessary information about Pregabalin and Etoricoxib.

Abbreviations

ACE -Aceclofenac
AUC -Area under curve
BCS -Biopharmaceutical classification system
CNS -Central nervous system
C max -Maximum plasma concentration
DSC -Differential scanning colorimetry
ETX -Etoricoxib
FDC -Fixed dose combination
GABA -Gamma-amino butyric acid
HILIC -Hydrophilic Interaction Liquid Chromatography
HPLC - High Performance Liquid Chromatography
ICH - International Conference on Harmonization
IUPAC - International union of pure and applied chemistry
LC - Liquid chromatography
MCA - Methylcobalamine
MS - Mass spectroscopy
NSAID - Non-Steroidal Anti-inflammatory drug
PCT - Paracetamol
PGB - Pregabalin
PGs - Prostaglandins
TCL - Thiocolchicoside
TGA - Thermogravimetric analysis
TLC - Thin layer chromatography
XRPD - X-Ray powder diffraction
VAL - Valdecoxib
V_d - Volume of distribution

REFERENCES

1. Amit J. Vyas, Grishma P. Nathwani, Ajay I. Patel, Nilesh K. Patel, Ashok B. Patel. Validated Stability Indicating RP-HPLC DAD Method for Simultaneous Determination of Amitriptyline Hydrochloride and Pregabalin in Presence of Stress degradation products in tablet dosage form. Chem. Methodol , 3(2019) 494-508.
2. Swapna Goday, Abdul Rahaman SK, Prameelarani. Development of a New Stability Indicating RP-HPLC Method for Simultaneous Estimation of Epalrestat and Pregabalin and its Validation as per International Conference on Harmonization Guidelines. Asian J Pharm Clin Res, 2018,11(5) 319-324.
3. Bootjan Martinc, Iztok Grabnar, Aleo Mrhar, and Tomaz Vovk. Rapid High-Performance Liquid Chromatography Method for Determination of Pregabalin in a Pharmaceutical Dosage Form Following Derivatization with Fluorescamine. J. AOAC Int, 2010,93(4).
4. Kavitha M P, A. Rajasekhar. A Validated HPLC Method for the Analysis of Pregabalin and Methylcobalamine in Bulk and

Pharmaceutical Formulation. Pharmacie Globale (IJCP) 2013, 07 (4).
5. Jampala Balaji, Bandi Ramachandra, N.V.S.Naidu. Analytical RP-HPLC Method for Development and Validation Of Pregabalin In Bulk And The Determination Of Pregabalin In Capsule Dosage Form. Int. j. innov. Res. Sci. eng. technol, 2014, 3(4).
6. David Berry and Christopher Millington. Analysis of Pregabalin at Therapeutic Concentrations in Human Plasma/Serum by Reversed-Phase HPLC. Ther Drug Monit ,2005, 27, 451–456.
7. E. Souri, M. Eskandari , M. Barazandeh Tehrani, N. Adib And R. Ahmadkhaniha. HPLC Determination of Pregabalin in Bulk and Pharmaceutical Dosage Forms After Derivatization with 1-Fluoro-2,4-dinitrobenzene. Asian J. Chem, 2013, 25(13) 7332-7336.
8. haritha potluri, sreenivasa rao battula, sunandamma yeturu. Validated stability indicating rp-hplc method for simultaneous determination of nortriptyline and pregabalin in bulk and combined dosage formulations. J. Chil. Chem. Soc, 2017, 62(2).
9. Nupur Mewada, Bhumi Patel, Jaymin Patel, Kunjal Vegad .Stability Indicating RP-HPLC Method Development and Validation for Simultaneous Estimation of Pregabalin and Nortriptyline in Tablet. IJPRS, 2017, 6(2).
10. Halima Akther, Md. Masud Morshed. Development of a Method and its Validation for Estimation of Pregabaline in Pharmaceutical and Bulk Formulation. Biomed. Sci. Today, 2015, 2(10).
11. K. Siva Nagaraju , Battula Sreenivasa Rao. Development, validation & stress degradation studies of pregabalin by high performance liquid chromatography. *IJPSR*, 2013,4(7) 2782-2788.
12. N. Kannapan, S.P. Nayak , T.Venkatachalam , V. Prabhakaran. Analytical RP-HPLC Method for Development and Validation of Pregabalin and Methylcobalamine in Combined Capsule Formulation. *J. Appl. Chem. Res*,2010, 13, 85-89.
13. G.B. Kasawar, M.N. Farooqui. Development and Validation of HPLC Method for the Determination of Pregabalin in capsule. Indian J Pharm Sci, 2010, 72(4) 517-519.
14. Rajinder Singh Gujral, Sk Manirul Haque and Sanjeev Kumar. A novel method for the determination of pregabalin in bulk pharmaceutical formulations and human urine samples. Afr. J. Pharm. Pharmacol, 2009, 3(6), 327-334.
15. G. Swapna and Merugu Manasa. RP-HPLC Method Development and Validation for Pregabalin and Celecoxib in Bulk and Dosage Forms. World J. Pharm. Res, 2017, 6(8).
16. Reza Ahmadkhaniha, Siavash Mottaghi, Mohammad Zargarpoor and Effat Souri. Validated HPLC Method for Quantification of Pregabalin in Human Plasma Using 1-Fluoro-2,4-dinitrobenzene as Derivatization Agent. Chromatogr. Res. Int , 2014, 1-6.
17. Krishna R Gupta* , Amruta Likhar and Sudhir G Wadodkar. Application of Stability Indicating HPLC Method for Quantitative Determination of Etoricoxib and Paracetamol in Pharmaceutical Dosage Form. Eurasian J. Anal. Chem, 2010, 5(3), 218-226.
18. Chandrashekhar Narajji and Manohar D. Karvekar. Method development and validation for simultaneous estimation of Paracetamol and Etoricoxib in pharmaceutical dosage form by RP-HPLC method. Der Pharma Chem, 2011, 3 (4) 7-12.
19. S. R. Pattan, S. G. Jamdar , R. K. Godge , N. S. Dighe , A.V. Daithankar , S. A. Nirmal , M.G.Pai. RP- HPLC Method for Simultaneous Estimation of Paracetamol and Etoricoxib from Bulk and Tablets. J. Chem. Pharm. Res, 2009, 1(1) 329-335
20. Sanjiv Kumar, Amit Joshi, Rahul Singh Thakur, Anupam K. Pathak and Kamal Shah. Simultaneous Sstimation of Etoricoxib and Thiocolchicoside by RP-HPLC Method in Combined Dosage Forms. Acta Pol. Pharm, 2011, 68(6), 839-845.
21. Srinivasu topalli, T. G. Chandrasekhar and m. Mathrusri Annapurna. Validated RP- HPLC Method for the Assay of Etoricoxib (A Non-Steroidal Anti-Inflammatory Drug) in Pharmaceutical Dosage Forms. E-Journal of Chemistry, 2012, 9(2), 832-838.
22. Gangane P.S*., S.M.Bagde,S.G.Mujbaile and K.D.Niranjane. Development and Validation of HPLC Assay Method for Etoricoxib in Bulk Drug and Tablet Formulation. IJONS, 2014, 24(4).
23. Mahamudul Haque , Shamema Nasrin , Md. Moniruzzaman Monir , Md. Mahbubur Rahman, Sama-Un-Abed Chowdhury. Method Development and Validation of RP-HPLC Method of Etoricoxib In Bulk and Its Tablet Dosage Forms. Am. J. PharmTech Res, 2012, 2(6).
24. S.B. Bagade, D.B. Meshram and M.R. Tajne. Estimation of Etoricoxib in Tablet Dosage form by RP- HPLC using Internal Standard with Emphasize on Specificity Parameter Method. Orient. J. Chem, 27(2), 697-701.
25. Ashok K. Shakya, Nooman A. Khalaf. High Performance Liquid Chromatographic Determination of Etoricoxib in Plasma. Asian J. Chem, 2017, 19(7), 5241-5250.
26. Shantaram Gajanan Khanagea, Dnyaneshwar Santram Kaleb, Popat Baban Mohitea, Vinayak Kashinath Deshmukhb. Reversed Phase High Performance Liquid Chromatographic Method for Simultaneous Estimation of Pregabalin and Aceclofenac in Tablet Formulation. *JRPS*, 2014, 3(2), 184-192.
27. M. Krishna Chaitanya Prasad, DR. G. Vidhya Sagar, DR. P. Sudhakar. Simultaneous HPTLC Method for Estimation of Gabapentin and Pregabalin. Int J Pharm Pharm Sci, 5(4), 326-333.
28. Rajendra B. Patil, Tushar A. Deshmukh and Vijay R.Patil. Development and Validation of HPTLC Method for Simultaneous Estimation of Aceclofenac and Pregabalin in Bulk Dosage form. Int. j. curr. Innov. res, 2017, 3(6), 691-696.
29. Dimal A Shah, Emanual Michael Patelia and Aswin Mori. Simultaneous Estimation of Pregabalin and Methylcobalamine in Pharmaceutical Formulation by HPTLC-Densitometry Method. J Chromat Separation Techniq, 2013, 4(2).
30. N.J. Shah, S.J. Shah, D.M. Patel. Development and validation of HPTLC method for estimation of etoricoxib. Indian J Pharm Sci, 2006, 68(6), 788-789.
31. Gourab Maheshwari, Ganesa Sundararajan Subramanian, Arumugam Karthik, Averineni Ranjithkumar, Prashant Musmade, Kishore Ginjupalli, and Nayanabhirama Udupa. High-Performance Thin-Layer Chromatographic Determination of Etoricoxib in the Bulk Drug and in Pharmaceutical Dosage Form. J PLANAR CHROMAT, 2007, 5, 335–339.
32. Dhaneshwar, Sunil R.; Raut, Kanchan O.; Bhusari, Vidhya K. Validated HPTLC Method for Simultaneous Estimation of Paracetamol and Etoricoxib in Bulk Drug and Formulation. Asian j. pharm. Boil res, 2011, 1(1), 87-93.
33. Vivek S. Rajmane, Santosh V. Gandhi,1 Upasana P. Patil, And Mahima R. Sengar. High-Performance Thin-Layer Chromatographic Determination of Etoricoxib and Thiocolchicoside in Combined Tablet Dosage Form. J. AOAC Int , 2010, 93(3), 783-786.
34. Poonam H. Chaube, Santosh V. Gandhi, Padmanabh. High Performance Thin Layer Chromatographic Analysis of Paracetamol and Etoricoxib in Spiked Human Plasma. Research J. Pharm. and Tech, 2011, *4(8)*.
35. Sravanthi T., Madhavi N. Stability Indicating UPLC Method for Simultaneous Estimation of Epalrestat and Pregabalin in Tablet Dosage Form. Res J Pharm Technol, 2019, 12(2).
36. Sandra Rinne Dahl , Kirsten Midtbøen Olsen, Dag Helge Strand. Determination of gamma-hydroxybutyrate (GHB), beta-hydroxybutyrate (BHB), pregabalin, 1,4-butane-diol (1,4BD) and gamma-butyrolactone (GBL) in whole blood and urine samples by UPLC–MSMS. J. Chromatogr. B, 2012, 37–42, 885–886.
37. M. Ravisankar, Dr. S. Alexander , M.Shankar , Dr. E. Jeyaseeli Florance , Dr. R. Senthil Kumar. Method Development And Validation of Aceclofenac and pregabalin in Marketed Formulation by UPLC Method. Acta Biomedica Scientia, 2020, 7(1), 25-30
38. Xiaoran Zhang1, Nan Guo, Wei Ji, Qing Wen. Rapid Quantitative Analysis of etoricoxib in human plasma by UPLC-MS/MS and application to a pharmacokinetic study in Chinese healthy volunteers. doi: 10.1002/bmc.4414
39. Dinesh N. Vora and Arun A. Kadav. Separation of Etoricoxib and Its Degradation Products in Drug Substance Using UPLC. Eurasian J. Anal. Chem, 2007, 2(3).
40. Khalaf Omar, Hind Ahmed Mahmoud, Nief Rahman. Indirect Spectrophotometric Method for Estimation of Pregabalin in Pharmaceutical Preparations. Sys Rev Pharm, 2020, 11(4), 32-35.
41. Nawab Sher, Nasreen Fatima Shahnaz Perveen. Pregabalin and Tranexamic Acid Evaluation by Two Simple and Sensitive Spectrophotometric Methods. Int. J. Anal. Chem, Volume 2015, Article ID 241412, 7.
42. Sherin F. Hammad and Ola M. Abdallah. Optimized and Validated Spectrophotometric Methods for the Determination of Pregabalin in Pharmaceutical Formulation Using Ascorbic Acid and Salicylaldehyde. Am. J. Sci , 2012, 8(12).
43. Kaur Navaneet, Mittal Karan, Nagar Rishab. A Sensitive Photometric Method for the Determination of Pregabalin in Pure

Drug and Pharmaceutical Formulations through Benzoylation. IRJP, 2010, 1(1) 175-180.
44. K. Sowjanya, J.C. Thejaswini, B.M. Gurupadayya and M. Indu priya. Spectrophotometric determination of Pregabalin using 1, 2-Napthaquinone-4- sulfonic acid Sodium and 2, 4 dinitrophenyl hydrazine in pharmaceutical dosage form. Der Pharm. Lett, 2011, 3(2) 47-56.
45. Dipak D. Patil, Mukesh S. Patil , Yogita B. Wani. Spectrophotometric method for pregabalin determination: An experimental design approach for method development. J. Assoc. Arab Univ. Basic Appl. Sci, 2015: 1-7
46. Rajinder Singh Gujiral, S.K Manirul Haque and Prem Shankar. Int J Biomed Sci. 2009 jun;5(2): 175-180
47. Alka Bali and Prateek Gaur. A novel method for spectrophotometric determination of pregabalin in pure form and in capsules. Chem. Cent. J ,2011, 5, 59.
48. Mohamed. Rizk,a Mona S. Elshahed,a Ali K. Attiab and Amir S. Farag. Spectrophotometric determination of Pregabalin Using N-(1-Naphthyl) Ethylenediamine, as UV Labeling Reagent. *IJPBS*, 2015, 5(2) 152-162.
49. S. Varik and T. Walke. Spectrophotometric Determination of Pregabalin from the Capsule Dosage Form Based on its Micellar Catalyzed Reaction with Sanger's Reagent. IJRPB, 2013, 4(4).
50. ARMAGAN, Onal. Development and Validation of Selective Spectrophotometric Methods for the Determination of Pregabalin in Pharmaceutical Preparation. Chin. J. Chem, 2009, 27(4).
51. Merin .K. Oommen , Sosamma Cicy Eapen , Sheeja .Velayudhankutty , Dr.Yellina.Haribabu. UV-VIS Spectrophotometric Method for Estimation of Pregabalin and Methylcobalamin in Bulk and Capsule Dosage Form. IJPRLS,2014, 2(1), 6 – 15.
52. Sasmita Kumari Acharjya, Y.Rajesh, Pinakini Panda, Priyambada Mallick and M. Mathrusri Annapurna. Spectrophotometric methods for simultaneous estimation of etoricoxib and thiocolchicoside in bulk and combined pharmaceutical dosage form. J Pharm Educ, 2010, 1(1).
53. Kamal shah, Alka Gupta and Pradeep Mishra. Extractive Spectrophotometric Methods for the Determination of Etoricoxib in Tablets. E-Journal of Chemistry ,2009, 6(1), 207-212.
54. S.R. Shahi, G.R. Agrawal, P.B. Rathi, N.V. Shinde, V.G. Somani , S.B. Mahamuni and A.N. Padalkar. Development and Validation of UV Spectrophotometric Method for the Determination of Etoricoxib in Bulk and Tablet Formulation. Rasayan J. Chem, 2008, 1(2), 390-394
55. Shipra Singh, Amrita Mishra, Anurag Verma, Ashoke K. Ghosh, Arun K. Mishra. A simple Ultraviolet spectrophotometric method for the determination of etoricoxib in dosage formulations. J. Adv. Pharm. Tech. Res. 2012, 3(4), 237-240.
56. Vishnu P. Choudhari, Sanket R. Parekar, Subhash G. Chate, Pradeep D. Bharande, Bhanudas S. Kuchekar. Development and validation of UV-visible spectrophotometric baseline manipulation methodology for simultaneous analysis of drotraverine and etoricoxib in pharmaceutical dosage forms. Pharm. Methods, 2011, 2(4).
57. Paweł Dzygiel , Daniela Fraier. Simultaneous Determination of Pregabalin, Sildenafil and Its Active Metabolite in Rat Plasma Utilising SPE Followed by LC–MS–MS. Chromatographia, 2011, 73, 1177–1182.
58. Liberato Brum Junior, Danieli Cátia Ceni , Marcio Fronza, Paulo Renato de Oliveira & Sérgio Luiz Dalmora. Validation of an LC-Tandem MS/MS Method for the Determination of Etoricoxib in Human Plasma and Pharmaceutical Formulations. J. Liq. Chromatogr. Relat. Technol, 2006, 29, 123–135.
59. Lutz Brautigam, Jens U. Nefflen, Gerd Geisslinger. etermination of etoricoxib in human plasma by liquid chromatography–tandem mass spectrometry with electrospray ionisation. J. Chromatogr. B, 788 (2003), 309–315.
60. Abdel Chahbouni, BSc, Arno Sinjewel, BSc, Jeroen C. G. den Burger, BSc, René M. Vos, BSc, Abraham J. Wilhelm, PharmD, Agnes I. Veldkamp, PharmD, PhD, and Eleanora L. Swart, PharmD, PhD. Rapid Quantification of Gabapentin, Pregabalin, and Vigabatrin in Human Serum by Ultraperformance Liquid Chromatography with Mass-Spectrometric Detection. Ther Drug Monit, 2013, 35(1).
61. Uttam Mandal, Amlan Kanti Sarkar, Kadagi Veeran Gowda, Sangita Agarwal, Anirbandeep Bose, Uttam Bhaumik, Debotri Ghosh, Tapan Kumar Pal. Determination of Pregabalin in Human Plasma Using LC-MS-MS. Chromatographia, 2008, 67, 237–243.
62. Thejo Moorthy Karavadi and B. R. Challa. Bioanalytical Method Development and Validation of Pregabalin in Rat Plasma by Solid Phase Extraction with HPLC-MS/MS: Application to a Pharmacokinetic Study. J. Liq. Chromatogr. Relat. Technol,, 37:130–144, 2014
63. Ce´dric Priez-Barallon, Je´re´my Carlier, Baptiste Boyer, Mounir Benslima3, Laurent Fanton, Ce´dric Mazoyer and Yvan Gaillard. Quantification of Pregabalin Using Hydrophilic Interaction HPLC-High-Resolution MS in Postmortem Human Samples: Eighteen Case Reports. J. Anal. Toxicol, 2014, 38, 143 –148.
64. Vikas V. Vaidya, Santosh M. Yetal, Shikha M. N. Roy, Noel A. Gomes, Santosh S. Joshi. LC-MS–MS Determination of Pregabalin in Human Plasma. Chromatographia 2007, 66, 925–928.
65. Ramakrishna Nirogi , Vishwottam Kandikere, Koteshwara Mudigonda, Prashanth Komarneni, Raghupathi Aleti. Liquid chromatography atmospheric pressure chemical ionization tandem mass spectrometry method for the quantification of pregabalin in human plasma. J. Chromatogr. B, 877 (2009), 3899–3906.
66. Hilda De La Vega, Kim Fox, Justine Pardi, Wendy Santiago-Tirado and Gail Cooper. Validation of a High-throughput Screening and Quantification Method for the Determination of Gabapentinoids in Blood Using a Combination of LC-TOF-MS and LC-MS-MS. J. Anal. Toxicol, 2019, 00, 1–7.
67. Caroline Pauly, Michel Yegles and Serge Schneider. Pregabalin Determination in Hair by Ultra-High-Performance Liquid Chromatography–Tandem Mass Spectrometry. J. Anal. Toxicol, 2013; 37: 676 –679
68. Ulrike Werner,1 * Dierk Werner,2 Burkhard Hinz,1 Christoph Lambrecht1 and Kay Brune. A liquid chromatography–mass spectrometry method for the quantification of both etoricoxib and valdecoxib in human plasma. Biomed. Chromatogr, 2005, 19, 113–118.
69. Sergio Luiz Dalmora Maximiliano da Silva Sangoi Lucelia Magalh¼es da Silva Rui Oliveira Macedo Thiago Barth. Validation of a capillary zone electrophoresis method for the comparative determination of etoricoxib in pharmaceutical formulations. J. Sep. Sci, 2008, 31, 169 – 176.
70. Juana Rodr´ıguez, Gregorio Castaneda, Lorena Munoz. Direct determination of pregabalin in human urine by nonaqueous CE-TOF-MS. Electrophoresis , 2013, 34, 1429–1436.
71. Henrik Harnisch, Yi-han Chien, Gerhard K. E. Scriba. Capillary Electrophoresis Method for the Chiral Purity Determination of Pregabalin Derivatized with Dansyl Chloride. Chromatographia, Feb 2018.
72. Hamid Reza Sargazi , Elnaz Tamizi , Elaheh Rahimpour , Abolghasem Jouyban. Development of a Micellar Electrokinetic Chromatographic Method with Indirect UV Detection for Pregabalin Determination in Serum Samples. Pharm. Sci, 2018, 24, 298-303.
73. Nagendra S. Singha, Rajib K. Paul a, Marc C. Torjmanb, Irving W. Wainer. Gabapentin and (S)-pregabalin decrease intracellular d-serine concentrations in PC-12 cells. Neurosci. Lett, *535*, 2013, 90–94.
74. Szabolcs Béni, Tamás Sohajdaa, Gábor Neumajer, Róbert Iványi, Lajos Szente, Béla Noszál. Separation and characterization of modified pregabalins in terms of cyclodextrin complexation, using capillary electrophoresis and nuclear magnetic resonance. J. Pharm. Biomed. Anal, , 2010, 51, 842–852.
75. Ali F. Alghamdi. Development of Square Wave Voltammetric Method for the Electrochemical Determination of Pregabalin as Pregabalin-Cu (II) Complex Using Graphite Electrode. Mor. J. Chem, 2016, 4(4) 853-861.
76. Hayam M. Lotfy, Adel M. Awad, and Mostafa A. Shehata. Novel Ion Selective Electrode for Determination of Pregabalin in Pharmaceutical Dosage Form and Plasma. Anal. Bioanal. Electrochem, 2012, 4(5) 507 – 517.
77. Eman H El-Naby. Potentiometric sensing platform for selective determination of pregabalin in pharmaceutical formulations. Open J Anal Bioanal Chem, 3(1), 49-56.
78. Brijesh Patel and Nayan Ratnakar. Formulation, Development and Evaluation of Sustained Release Beads of Pregabalin. WJPPS, 2017, 6(6), 1604-1620
79. Shazia Akram Ghumman, Sajid Bashi, Sobia Noreen. Taro-corms mucilage-alginate microspheres for the sustained release of

pregabalin: In vitro & in vivo evaluation. Int. J. Biol. Macromol, 2109, 139, 1191–1202.
80. Alaa H. Salamaa,b , Abdelfattah A. Abdelkhalekc , Nermeen A. Elkasabgy .Etoricoxib-loaded bio-adhesive hybridized polylactic acid-based nanoparticles as an intra-articular injection for the treatment of osteoarthritis. Int. J. Pharm, 2020, 578.
81. M. Kaur Saini and S. Nath Sanyal. Evaluation of chemopreventive response of two cycloxygenase-2 inhibitors, etoricoxib and diclofenac in rat colon cancer using FTIR and NMR spectroscopic techniques. Nutr Hosp. 2010, 25(4), 577-585.
82. Upasana Bilthariya, Nivrati Jain, Vaibhav Rajoriya. Folate-conjugated albumin nanoparticles for rheumatoid arthritis targeted delivery of etoricoxib. Drug Dev Ind Pharm, Early Online: 1–10.
83. Bhaskar Chauhan, Shyam Shimpi and Anant Paradkar. Preparation and Characterization of Etoricoxib Solid Dispersions Using Lipid Carriers by Spray Drying Technique. AAPS PharmSciTech, 2005, 6 (3).
84. Haresh M. Patel, Bhanubhai N. Suhagia Shailesh A. Shah. Preparation and characterization of etoricoxib-b-cyclodextrin complexes prepared by the kneading method. Acta Pharm,2007, 57, 351–359.
85. Inderbir Singh, Banveet Kaur, Pradeep Kumar. Masking the unpleasant taste of Etoricoxib by crosslinked acrylic polyme based ion-exchange resin complexation. Polym. J, 2010, 40(3).
86. Abhisekh Das, Amit Kumar Nayak, Biswaranjan Mohanty. Solubility and Dissolution Enhancement of Etoricoxib by Solid Dispersion Technique Using Sugar Carriers. ISRN Pharm, 2011, Article ID 819765.
87. M.U. Abdullaeva, N.Sh. Khalilova, A.Yu. Tashpulatov, Z.U. Usmanalieva. Comprehensive Method For Forensic Study Of Pregabalin And Tramadol Trace Amounts Using Gas-Liquid Chromatography With Mass Spectrometric Detector And Ir Spectrophotometry. EJMCM, 2020, 7(3).
88. Noorana Tehseen, Vinay Rao, Mohd Abdul Hadi. Design and Characterization Of Twice Daily Mini-Tablets Formulation Of Pregabalin. Int. J. Pharm. Pharm. Sci, 2013, 5(1).
89. Modeling Shrawani Lamichhane, Jun-Bom Park, Dong Hwan Sohn and Sangkil Lee. Customized Novel Design of 3D Printed Pregabalin Tablets for Intra-Gastric Floating and Controlled Release Using Fused Deposition. Pharmaceutics 2019, 11, 564.
90. Blommel M. L, Blommel, A. L. Pregabalin: An Antiepileptic Agent Useful for Neuropathic Pain. Am. J. Health. Syst. Pharm. 2007, 64, 1475–1482.
91. Takemoto J, Reynolds, J, Remsberg, C, Vega-Villa. Clinical Pharmacokinetic and Pharmacodynamic Profile of Etoricoxib. Clin. Pharmacokinet. 2008, 47, 703–720.