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A Reviw on Analytical Methods– Levetiracetam

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ABSTRACT: Levetiracetam is a pyrrolidone-family antiepileptic drug of the second generation with a broad spectrum of activity. The US Food and Drug Administration authorized Levetiracetam as a broad-spectrum antiepileptic drug in 1999. The hydrophilic groups of Levetiracetam differentiate it from other earlier antiepileptic drugs. The following review covers the analytical techniques for the analysis of Levetiracetam.

KEYWORDS: Levetiracetam (LVT); Antiepileptic drug; analytical technique; spectrophotometry; HPLC, LC-MS.

I. INTRODUCTION:

The U.S. Food and Drug Administration has authorised levetiracetam (Keppra), a novel antiepileptic drug, as an adjunctive therapy for partial myoclonic and tonic-clonic seizures as well as a monotherapy treatment for epilepsy in conditions of partial seizures.Other mental and neurological problems such Tourette syndrome, autism, and anxiety disorders may be helped by levetiracetam.LVT has a linear pharmacokinetic profile and is nearly completely absorbed after oral administration.[1]Chemically it is (2S)-2-(2-oxopyrrolidin-1-yl)butanamide (figure.1). It has a molecular formula $C_8H_{14}N_2O_2$ (MW 170.20).Levetiracetam is very soluble in water and methanol, and chloroform. Levetiracetam is practically insoluble in n-hexane[2]. LVT has a linear pharmacokinetic profile and is nearly completely absorbed after oral administration. It is less than 10% protein bound. LVT has no effect on or is influenced by the cytochrome P450 system. It is mostly eliminated via the kidneys. In renal failure, its plasma half-life can be extended.[3]This is a structural analog of piracetam that binds to the synaptic vesicle protein SV2A, obstructing nerve transmission across synapses.[1]

Fig.1 Structure of Levetiracetam

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The present review article summarises the analytical techniques so far developed, such as spectrophotometry[4-10] (Table 1), high performance thin layer chromatography[11] (Table 2), high-performance liquid chromatography[12-26] (Table 3), liquid chromatography-mass spectrometric methods[27-32] (Table 4) for the determination of Levetiracetam (LVT) and some of the analytical parameters were highlighted.

1. Spectrophotometric method:

Table 1. Determination of Levetiracetam by UV Spectrophotometric Method:

Solvent/Reagent	λ_{max}(nm)	Linearity (µg/ml)	Reference
2-Chloro phenyl hydrazine and 2 ml (0.25 %) Anthranilic acid	560 485	-	[4]
Water	209	2-10	[5,6]
Glacial acetic acid	221	30-90	[7]
2,4- Dinitrophenylhydrazine	455	30-130	[8]
Methanol	220	10-50	[9]
Distilled water	265	2-12	[10]

2. High Performance Thin Layer Chromatography (HPTLC):

Table 2. Determination of Levetiracetam by HPTLC Method:

Mobile Phase	λ (nm)	Stationary Phase	Linearity Range (ng/band)	LOD (ng/band)	LOQ (ng/band)	Reference
Toluene: Acetone: Methanol(6:2:2) v/v/)	210	Silica gel 60 F_{254}	500-3000	19.76	65.89	[11]

3. High Performance Liquid Chromatography (HPLC):

Table 3. Determination of Levetiracetam by HPLC Method:

Technique	Mobile Phase	Flow rate	LOD (µg/ml)	LOQ (µg/ml)	References
RP-HPLC	Methanol: Water:TEA 75:25:05 (V/V)	1.0ml/min	0.05	0.15	[12]
RP-HPLC	mixture of 0.1 g/L of triethylamine and acetonitrile(70: 30 v/v)	1.0ml/min	1.66	8.7	[13]
RP-HPLC	MeOH and 25mM KH_2PO_4 buffer pH 3 (38.4:61.6, v/v)	0.8 ml/min	0.20	1.56	[14]
RP-HPLC in Human serum	ammonium acetate buffer (10mM, pH 5) and acetonitrile (50:50v/v)	0.3ml/min	0.8	2.5	[15]
RP-HPLC	diphasic sodiumphosphate buffer and acetonitrile in a ratio of 80:20	1.5 ml/min	0.6-1.2	4	[16]
RP-HPLC	buffer solution (pH 2.8) and acetonitrile in the ratio of 90:10	1.2 ml/min	-	-	[17]
HPLC	Methanol: ACN: Water (60:20:20) pH3	1 ml/min	0.0719	0.218	[9]
HPLC	1 L deionizedwater, one vial of Waters (Milford, MA) D4 mobile phase	0.5 ml/min	-	1	[18]

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	modifier(dibutylammoniumphosphate), and 100 ml of methanol				
HPLC	potassium dihydrogen phosphate buffer (10mM, pH4.6)/acetonitrile(93:7 v/v)	0.5 ml/min	0.19	12-46	[19]
HPLC in Human plasma	methanol:ammonium buffer, 10 mmol/L pH 10 (30:70, v: v)	0.8 ml/min		0.5	[20]
HPLC	potassium phosphate buffer (pH 5.5) and acetonitrile (45:55, v/v)	0.9 ml/min	-	-	[21]
HPLC in Human serum	diluted phosphoric acid/acetonitrile	1 ml/min	0.1	1	[22]
HPLC in Human plasma	a mixture of potassium dihydrogen phosphate buffer (50mM, pH 4.5) and acetonitrile (94:6, v/v)	1.5 ml/min	1.0	2.0	[23]
HPLC in Human serum	a mixture of acetonitrile (A) and water (B)	1.0 ml/min	2.5	29.7Levetiracetam in tablets using monolithic and conventional	[24]
HPLC in Human plasma	a mixture composed of methanol, acetonitrile and a3 mm phosphate buffer, containing 0.5 mLtriethylamine(6:5:89, v/v/v) with an apparentpH of 6.0	1.0 ml/min	150	500	[25]
HPLC	Water: acetonitrile (90:10)	1.0 ml/min	0.8	8.0	[26]

4. Hyphenation Techniques:

Table 4. Determination of Levetiracetam by hyphenated Method:

Matrices	Mobile Phase	Column	Method	Flow Rate	LOQ (ng/ml)	Reference
Human plasma	a mixture of 0.1% formic acid in water and ACN (40:60 v/v)	LC-MS Agilent Zorbax SB-C18	isocratic elution	0.5 ml/min	0.5	[27]
Human plasma	30 mM aq. Orthophosphoric acidsolution/methanol(70:30) (A) and 10 mMaq. ortho-phosphoricacidsolutio	LC GL Sciences IntersilODS-3 (A) and Restek Ultra PFPP column	Gradient elution	1 ml/min	6	[28]

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	n/methanol(55:45)	(B)				
human plasma /serum /saliva	97% methanol in 15 mmol/lammonium acetate (v/v), 0.1% acetic acid	LC-MS/MS C-18 column	isocratic elution	1 ml/min	-	[29]
Humanplasma	5 mM Ammonium acetate (adjusted to pH 3.2 with Glacialacetic acid): Acetonitrile (20: 80) Clonazepam(Internalstandard)	LC-MS/MSThermo Electron Betasil C-18	isocratic elution	0.5 ml/min	0.125	[30]
Humanplasma	0.1% formic acid in 10.0 mM ammonium acetate (A) and100% methanol (B)	LC-MS/MS C18 SPE	isocratic elution	0.4 ml/min	0.50	[31]
Humanplasma	0.1% formic acid–10mMammonium formate in water (pH 3.5) (mobile phase solution A) and 0.1% formic acid in methanol (mobilephase solution B)	UPLC–MS/MS	Gradient elution	0.4 ml/min	0.5	[32]

II. CONCLUSION:

For the estimation of levetiracetam in bulk, pharmaceutical formulations, and biological samples, various analytical methods, including UV, HPLC, HPTLC, and hyphenated techniques such as LC-MS, UPLC-MS/MS methods, were described. This review article will help readers understand the analytical techniques described for quantifying Levetiracetam.

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