Bulletin of Environment, Pharmacology and Life Sciences
Bull. Env. Pharmacol. Life Sci., Vol 14[5] April 2025: 31-39

Online ISSN 2277-1808
Journal's URL: http://www.bepls.com
CODEN: BEPLAD

REVIEW ARTICLE **OPEN ACCESS**

Overview of Analytical Techniques for the Evaluation of Lidocaine Hydrochloride and Diltiazem Hydrochloride

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ABSTRACT
Anal fissure causes severe pain in the anal tract, to relieve pain topical anesthetics like Lidocaine Hydrochloride and Diltiazem Hydrochloride are used. Lidocaine Hydrochloride acts as a tertiary amine which for the temporary condition inhibits the nerve stimulation. Diltiazem Hydrochloride is a calcium channel blocker from the non-dihydropyridine class and acts as a vasodilator by inhibiting L type calcium channels. In the pharmaceutical sector, rapid research and development in drug discovery increase the hand on the development of analytical techniques. Any drug came out of drug research and discovery needs a developed and validated analytical method to prove its identity, safety, purity, and efficacy. Effective method development and validation results in an increase in accuracy and precision. The motive of this review article is to develop an article with well compilation of analytical techniques reported till in the literature for the evaluation of Lidocaine Hydrochloride and Diltiazem Hydrochloride in pharmaceutical or biological formulations. The techniques included are Hyphenated techniques, HPLC, UV Spectroscopy techniques, HPTLC and official pharmaceutical techniques.
Keywords: *Diltiazem Hydrochloride, High Performance liquid Chromatography, High Performance thin layer Chromatography, Lidocaine Hydrochloride, Ultra Violet Spectroscopy, Liquid Chromatography Mass Spectroscopy, Reverse Phase High Performance Liquid Chromatography*

Received 10.12.2024 Revised 07.03.2025 Accepted 17.04.2025

INTRODUCTION

A lesion of the anoderm in the anal tract is known as an anal fissure. A lesion known as an anal fissure (AF) is located in the membrane and the lower-gut anal tract from the anoderm up to the pectinate line. It has a close prevalence in either sex and more frequently affects younger generation. When other linked disorders are taken into account, its incidence becomes less frequent after age 65. 15 percent of women have it after giving delivery, making it especially common [1].

According to the degree of anal muscle contractions, a fissure may be extremely painful or practically unnoticeable. Rectal bleeding is another possibility for people with fissures; typically, this is limited amounts of fresh red blood that can be observed on toilet paper. Anal fissure therapy has generally involved a trial of topical anesthetics, baths, and diet supplement; surgical procedure performed if the pain becomes unbearable or if conservative measures are unsuccessful[1][2].The Lidocaine Hydrochloride and Diltiazem Hydrochloride used as topical anesthetics. They reduce the pain by decreasing the contractions and preventing vascular convulsions in the anal tract.

MECHANISM OF ACTION

Lidocaine Hydrochloride:

A local anesthetic drug made of an amide; lidocaine was formerly known as lignocaine. It is a tertiary amine produced from xylidine and its use quickly spread due to its exceptional safety profile compared to earlier local anesthetic drugs. Lidocaine works by temporarily inhibiting the formation of nerve fiber stimuli, which is how regional anesthesia is achieved. When lidocaine is infused close to a nerve, Sodium channels are then bound by lidocaine, resulting in a structural shift that precludes the temporary input of sodium and causes depolarization. Lidocaine has a rapid progression of effect and, depending on the amount given, the concentration used, the nerves blocked, and the patient's condition, the blockade may persist for up to 5 hours[3].

Diltiazem Hydrochloride:

Diltiazem is a calcium channel blocking agent that is derived from benzothiazepine. It is a member of the non-dihydropyridine calcium channel blocking agent medication class. It works as both a peripheral and coronary vasodilator. By inhibiting slow L type of calcium channels and preventing calcium from enter in to the smooth-muscle, CCB reduces intracellular calcium concentration while also increasing cGMP and cAMP which causes decrease in the level of calcium that is available to join with the messenger protein calmodulin, which therefore hinders the stimulation of the myosin (light-chain) kinase necessary for contraction of the smooth muscles[5]. Therefore, Diltiazem is effective in AF treatment which leads to relaxation of muscle and decrease in pressure over anal tract.

Table 1. PHYSICOCHEMICAL PROPERTIES:

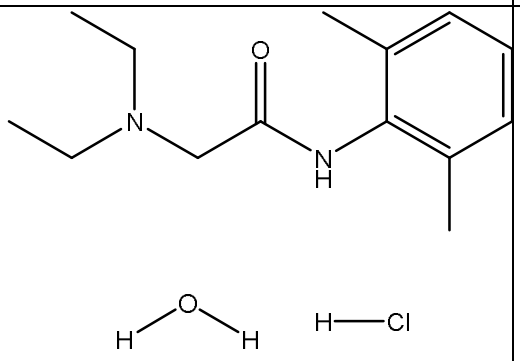
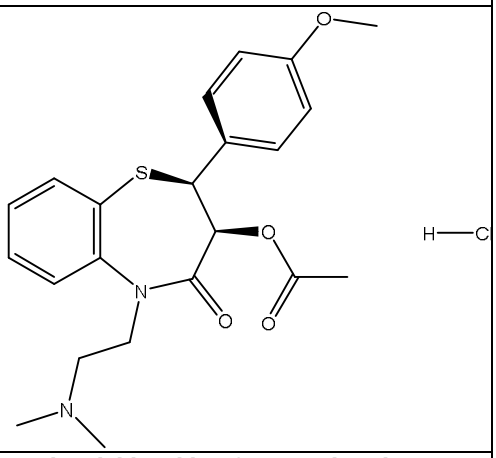
Properties	**Lidocaine Hydrochloride**	**Diltiazem Hydrochloride**
IUPAC name[7]	2-diethylamino-2',6'-xylidide hydrochloride monohydrate.	(2S,3S)-2,3,4,5-tetrahydro-5-(2-dimethyl aminoethyl)-2-(4-methoxyphenyl)-4-oxobenzo[b]thiazepine-3-yl acetate hydrochloride.
Empirical formula[7]	C14H22N2O, HCl, H2O	C22H26N2O4S.HCl
Molecular mass[7]	288.8 g mol-1	451.0 gm mol-1
Appearance[7]	A crystalline, white powder.	A crystalline, white powder or small crystals
Structure		
Solubility profile	Freely-soluble -: 1.Water 2.Chloroform 3.Ethanol 3.Benzene [8]	Freely soluble-:Chloroform, methanol, water. Sparingly soluble-:Dehydrated alcohol. Practically insoluble-:Benzene, Ether[9]

Table 2. OFFICIAL INDIAN PHARMACOPEIAL ANALYTICAL TECHNIQUE FOR LIDOCAINE HYDROCHLORIDE(LIH):

Assay	**Titrate**: 30- ml glacial acetic acid (anhydrous), dissolve 0.5 gm of LIH Add-up 6 ml of mercuric acetate solution. **Indicator:** Crystal violet solution. **Titrant:** Perchloric acid (0.1 M). **Result:** 0.1 M, 1 ml Perchloric acid ≅0.02708 gm C14H22N2O, HCl, H2O [6]

Table 3. REPORTED UV, HPLC, HPTLC AND HYPHENATED ANALYTICAL TECHNIQUES FOR LIDOCAINE HYDROCHLORIDE(LIH):

S No.	Drug name	Analytical-techniques	Description: Analytical techniques	Ref. No.
1.	Lidocaine	U.V spectrophotometry	**Solvent:** 0.1M HCl **Linearity:** 5-30 µ g per mL **Wavelength:** 263 ± 1 nm **Limit of Detection:** 0.1-0.5µg per mL	[9]
2.	Lidocaine Hydrochloride	Extraction-Spectrophotometric technique.	**Solvent:** Distilled water **Linearity:** 0.10 and 10 mg per L **Wavelength:** 508 nm **Limit of Detection:** 0.024 mg per L - 0.100 mg per L	[10]
3.	Lidocaine Hydrochloride	HPLC-UV	**Stationary phase:** Ion Pac ERCUS C18 RP-column **Mobile phase:** Acetonitrile: Water (20:80 V/V) **Flow rate:** per minute 1.0 mL **Wavelength of detection:**254 nm **Time of retention:**19 minutes **Linearity:**0.1 to 0.5µg per mL	[11]
4.	Lidocaine Hydrochloride (LIH) and Tribenoside (TR)	HPLC	**Stationary phase:** Varian C18(5µm,150×4.6 mm) column **Mobile phase:** Acetonitrile: Orthophosphoric acid 0.1% **Flow-rate:**1 mL per minute **Wavelength of detection:** LIH-:230nm, TR-:254 nm **Linearity:** LIH-:100-300µg per mL, TR-:250-750 µg per mL	[12]
5.	Paracetamol (PAR) and Lidocaine Hydrochloride (LIH)	RP-HPLC	**Stationary phase:** HPLC column (5mm, 4.6×150 mm,C18 XDB) **Mobile phase:** Water: Acetonitrile: Tetrahydrofuran (90:5:5 V/V/V) **Flow- rate:**1 mL per min **Wavelength of detection:** excitation-wavelength-:250 nm, emission- wavelength-:410 nm **Time of retention:** PAR-:3.103 minutes, LIH-:3.989 minutes **Linearity**: 20.0 to 100.0 µg per mL	[13]
6.	Dexpanthenol (DTA), Lidocaine Hydrochloride (LIH), Mepyramine Maleate (MPM)	RP-HPLC	**Stationary phase**: Inertsil (ODS-3 V, 5µm, 250 × 4.6 mm) **Mobile phase:** Ammonium acetate (0.01M): Methanol (70:30 V/V) **Flow-rate**: 1.3 mL per min and 1.5 mL per min **Time of retention:** DTA-:3.28, LIH-:11.67, MPM-:12.99 minutes **Linearity:** DTA-:30-180 µg per mL, MPM and LIH-:9-54 µg per mL	[14]
7.	Lidocaine HCl (LIH) and Nifedipine (NIF)	RP-HPLC	**Stationary phase:** Hypersil BDS column (LC-20,C18,5µm,250mm,4.6 mm) **Mobile phase:** (0.05 KH2PO4) Buffer: Methanol (50:50 V/V) **Flow-rate:**1 mL per min **Wavelength of detection:**234 nm. **Time of retention:** LIH-:4.170, NIF-:6.530 minutes **Linearity:** NIF-:1.5-4.5µg per mL, LIH-:7.5-22.5 µg per mL	[8]

8.	Phenazone (PNZ) and Lidocaine Hydrochloride (LIH)	RP-HPLC	**Stationary phase:** Agilent TC (C18,5µm,250×4.6 mm) **Mobile phase:** Phosphate buffer: Acetonitrile: Methanol (70:20:10 V/V/V) **Flow-rate:**1.5 mL per min **Wavelength of detection:**230 nm **Time for retention:** PNZ-:10.1, LIH-:7.2 minutes **Linearity:** LIH-:10-70 µg per mL**,** PNZ-: 50-150 µg per mL	[15]
9.	Lidocaine Hydrochloride (LIH), Hexachlorophene (HC)	HPLC	**Stationary Phase:** YMC-Triart C18 column (150mm × 4.6 mm,5µm) **Mobile phase**: Acetonitrile: (0.05 M) Phosphate buffer **Flow-rate:** 1.0 mL per min **Wavelength of detection:** 220 nm. **Linearity:** HC-:160.0 to 360.0 µg per mL, LIH-:600.0 to 1500.0 µg per mL	[16]
10.	Chlorhexidine Gluconate (CG), Metronidazole (MN), Lidocaine Hydrochloride (LIH) and Triamcinolone Acetonide (TA).	RP-HPLC	**Stationary phase:** Phenomenex Luna (C18, 5µm, 250×4.6 mm) column **Mobile phase:** Acetonitrile: Sodium dihydrogen phosphate buffer (50:50 V/V) **Flow-rate:** 1 mL per min **Wavelength of detection:** 230 nm **Time of retention:** CG-:13.50, MN-:17.98, LIH-:10.52, TA-:16.65 minutes **Linearity**: CG-: 0.05- 0.15 mg per mL, MN-:0.005-0.015 mg per mL, LIH-:0.10-0.30mg per mL, TA-:0.005-0.015 mg per mL	[17]
11.	Lidocaine Hydrochloride (LIH), Ketoprofen (KEP) and Hydrocortisone (HYC)	HPLC	**Stationary phase:** Shimadzu RP-HPLC C8 column **Mobile phase:** Acetonitrile: Phosphate Buffer (50:50 V/V) **Flow-rate:** 1 mL per min **Wavelength of detection**: 254 nm **Time of retention:** LIH-:1.54, HYC-:2.57, KEP-:5.78 minutes **Linearity:** LIH-:0.6-56, HYC-:0.6-56, KEP-: 0.2-100 PPM	[18]
12.	Lidocaine HCl (LIH), Prednisolone acetate (PA) and Dimethyl Sulfoxide (DS)	RP-HPLC	**Stationary phase:** PrincetoneSPHER 100 C18 (5µm, 250 mm× 4.6 mm) column **Mobile phase:** Acetonitrile: (0.01 M)Potassium dihydrogen phosphate buffer (54:46 V/V) **Flow-rate:** 1.0 mL per min **Wavelength of detection:** 261 nm **Linearity:** LIH-: 50.0 µg per mL, PA-:10.5 µg per mL, DS-:5.0 µg per mL	[11]
13.	Benzoxonium Chloride (BZC) and Lidocaine Hydrochloride (LIH)	RP-HPLC	**Stationary phase**: Nucleosil C18 column (5µm, 250 × 4.6 mm) **Mobile phase:** Potassium dihydrogen phosphate(10 mM): Acetonitrile (20:80 V/V) **Flow-rate**: 1 mL per min **Wavelength of detection:** 215 nm	[19]

			Time of retention: LIH-: 5.28± 0.13 min, BZC-: 9.76± 0.36 min. **Linearity:** 20-120 µg per mL	
14.	Choline Salicylate(CHS), Lidocaine Hydrochloride (LIH)	HPLC	**Stationary phase:** ACE C18 (5µm, 250× 4.6 mm) column **Mobile phase:** Acetonitrile: Phosphate buffer solution **Flow-rate:** 1 ml per min **Wavelength of detection:** 260 nm **Time of retention:** LIH-:5.174 minutes, CHS-:8.470 minutes **Linearity:** LIH-:120-180 µg per mL, CHS-: 640-960 µg per mL	[20]
15.	Tetracaine Hydrochloride (TTH),Procaine Hydrochloride (PRH), Mepivacaine Hydrochloride (MPH), Dibucaine (DBC), Ropivacaine Hydrochloride (ROH), Lidocaine Hydrochloride (LIH).	Thin layer chromatography-Raman spectroscopy	**Stationary phase:** Silica gel (thin) layer **Mobile phase:** Cyclohexane: Triethylamine (V: V=7:3) **Wavelength of detection:** 532 nm **RF value:** TTH-:0.17, PRH-:0.09, MPH-:0.50,DBC-:0.41,ROH-:0.73,LIH-:0.62	[21]
16.	Lidocaine Hydrochloride	GC-FID Technique	**Stationary phase:** HP-5 capillary (25µm, 5% phenyl methyl polysilocone 30 m × 0.320 mm) column **Mobile phase:** Nitrogen (carrier gas) **Flow-rate:** 1.6 mL per min **Wavelength of detection:** 356 nm **Time of retention:** 7.53 minute **Linearity:** 0.1-50 µg per mL	[22]
17.	Ceftriaxone Sodium (CFS) and Lidocaine Hydrochloride (LIH)	HPLC-MS/MS Technique	**Stationary phase:** Kinetex C18 column (5µm,50.0 × 4.6 mm) **Mobile phase:** Methanol: Ammonium acetate (0.01M) (70:30 V/V) **Flow-rate:** 0.5 mL per min **Linearity:** LIH-:3-300 ng/mL, CFS-:3-100 µg/mL	[23]

Table 4. OFFICIAL INDIAN PHARMACOPEIAL ANALYTICAL TECHNIQUE FOR DILTIAZEM HYDROCHLORIDE

Drug name	Analytical techniques	Description	Ref. No.
Diltiazem Hydrochloride	Liquid chromatography	**Stationary phase:** Stainless steel column, Octadecylsilane bounded to porous silica (5 µm, 30 cm × 9 mm) **Mobile phase:** Buffer solution of d-10-camphorsulphonic acid (0.116 percent w/v): Acetonitrile: Methanol (50:25:25 V/V/V) **Flow-rate-:**1.6 mL per min **Wavelength detection-:** 240 nm **Time of retention-:** 0.65	[7]

Table 5. REPORTED UV, HPLC, HPTLC AND HYPHENATED ANALYTICAL TECHNIQUES FOR DILTIAZEM HYDROCHLORIDE:

S No.	Drugs Name	Analytical techniques	Description: Analytical techniques	Ref. No.
1.	Diltiazem Hydrochloride	UV spectrophotometry	**Solvent:** Water **Linearity:** 6-16 µg per mL **Wavelength:** 236 nm **Limit of detection:** 0.2756 µg per mL	[24]
2.	Diltiazem Hydrochloride	Zero order derivative UV spectroscopy	**Solvent:** 0.05 N Sulphuric acid **Linearity:** 3-18 µg per mL **Wavelength:** 193 nm **Limit of detection:**0.222 µg per mL	[25]
3.	Diltiazem HCl (DT-HCl) and Levamisole HCl (LM-HCl)	UV Visible spectrophotometry	**Solvent:** Double distilled water **Linearity:** LM-HCl-: 2.41-32.5 and 1.20-16.86 µg per mL, DT-HCl-:2.26-48.48 and 2.26-27.06 µg per mL **Wavelength:** DT-HCl-:399 and 402 nm, LM-HCl-: 405 and 406 nm **Limit of detection:** LM-HCl-: 0.2 and 0.1 µg per mL, DT-HCl-:0.32 and 0.06 µg per mL	[26]
4.	Diltiazem Hydrochloride	Indirect UV spectrophotometric technique	**Solvent:** Water **Linearity:** 3.0-9.0, 3.5-7.0, 3.50-6.3 µg per mL for techniques A, B, C resp. **Wavelength:** Technique A-:521 nm, B-:528 nm, and C-: 525 nm **Limit of detection:** 0.006,0.007,0.024 µg per mL for technique A, B, C resp.	[27]
5.	Diltiazem Hydrochloride	HPLC	**Stationary phase:** Purospher Star C18 (5 µm, 150 × 4.6 nm) column **Mobile phase:** (0.05 percent) Trifluoroacetic acid aqueous solution: (0.05 percent) Trifluoroacetic acid methanolic solution (44:56 V/V) **Flow-rate:** 1.0 mL per min **Wavelength of detection**: 240 nm **Time of retention:** 14 minutes **Linearity:** 15.0-45.0 µg per mL	[28]
6.	Diltiazem Hydrochloride	RP-HPLC	**Stationary Phase:** Zorbax (C8, 4.6 mm × 250,5 µm) **Mobile phase:** Potassium monobasic phosphate buffer: Acetonitrile (60:40 V/V) **Flow-rate:**1.0 mL per min **Wavelength of detection:** 240 nm **Time of retention:** 4.66 minute **Linearity:** 50-150 µg per mL	[29]
7.	Diltiazem Hydrochloride	HPLC	**Stationary phase:** Hypersil BDS (C18,5.0 mm,150 mm, 4.6 mm) column **Mobile phase:** (0.2 percent) Triethylamine (TEA): Acetonitrile (ACN) (3:2 V/V) **Flow-rate:** 1.0 mL per min **Wavelength of detection:** 240 nm **Linearity:** 0.35-1.50 µg per mL	[30]
8.	Diltiazem Hydrochloride (DT-HCl) and Metabolite Desacetyl Diltiazem Hydrochloride (DS-HCl)	HPLC	**Stationary phase:** Microbonapack C18 (5 µm, 4.6× 250 nm) column **Mobile phase:** Acetate buffer: Acetonitrile (650:350 V/V) **Flow-rate:** 1.0 mL per min **Wavelength of detection:** 240 nm **Time of retention:** DT-HCl-:26.4 minutes, DS-HCl-:15.7 minute **Linearity:** 25%-250% of the stated limit i.e. (0.5 percent)	[31]

9.	Lovastatin (LST) and Diltiazem Hydrochloride (DT-HCl)	HPLC	**Stationary phase:** Kromasil (C18,10 µm, 300 mm×4 mm) column **Mobile phase:** Methanol: Water (90:10 V/V) **Flow-rate:** 1 mL per min **Wavelength of detection:** 237 nm **Time of retention:** LST-:4 minutes, DT-HCl-:5.62 minutes **Linearity:** LST-:40-110, DT-HCl-:110-180 µg per mL	[32]
10.	Diltiazem Hydrochloride	RP-HPLC	**Stationary phase:** Ascentis Express (C18 column) **Mobile phase:** (0.1 percent) Triethylamine, (pH 3.0, previously adjusted with Orthophosphoric acid): Acetonitrile (65:35 V/V) **Flow-rate:**1 mL per min **Wavelength of detection:** 236 nm **Time of retention:** 1.5 minutes **Linearity:** 50-150 µg per mL	[33]
11.	Diltiazem Hydrochloride.	HPLC	**Stationary phase:** Zorbax RX C8 (5 µm,150 mm ×4.6 mm) column **Mobile phase:** (0.05 M)Sodium dihydrogen phosphate monohydrate buffer (pH 3.0): Methanol (800:200 V/V) **Flow-rate:** 1.0 mL per min **Wavelength of detection:** 240 nm **Time of retention:** 16.394 minutes **Linearity:** 0.18-5.65 µg per mL	[34]
12.	Diltiazem Hydrochloride	HPLC	**Stationary phase:** Inertsil ODS-3 column (5 µm, 4.6×250 mm) **Mobile phase:** 500 ml Buffer: 250 ml Acetonitrile: 250 ml Methanol **Flow-rate:** 1.6 mL per min **Wavelength of detection:** 240 nm **Linearity:** 840-1560 µg per mL	[35]
13.	Diltiazem Hydrochloride	HPTLC	**Stationary phase:** HPTLC aluminum plates, precoated silica gel 60 F254 (20×10 cm,0.2mm) **Mobile phase:** Ethyl acetate: Methanol: Strong ammonia solution (80:10:10 V/V/V) **Wavelength of detection:** 238nm **RF value:** 0.54	[36]
14.	Diltiazem Hydrochloride	LC-MS	**Stationary phase:** Purospher C18(5 µm, 125 × 4 mm)column **Mobile phase** (100mM) Aqueous ammonium acetate: Acetonitrile (4:1 V/V) **Flow-rate:** 0.6 mL per min **Linearity**: 0.2-10 ppm	[37]

CONCLUSION

The review article described the summary of all analytical techniques from the reported techniques for the evaluation of Lidocaine Hydrochloride and Diltiazem Hydrochloride. The ultimate focus was to compile data on as many analytical techniques as possible and their specifics. There were numerous HPLC and UV analytical techniques available, but relatively little literature was available for hyphenated techniques (GC-FID, HPLC-MS/MS, LC-MS) and HPTLC. This article seeks to provide a thorough evaluation of the literature regarding the instruments used in analytical techniques, with a focus on the function of various analytical components in the assay of pharmaceuticals. It also underlines how the techniques evolved, moving from the more conventional titrimetric approach to the more complex hyphenated techniques.

ACKNOWLEDGMENT
This paper and the efforts behind it would not have been possible without the exceptional support of my guide Ms. Deepanti Gajjar, her enthusiasm, knowledge and exacting attention to detail have been an inspiration and kept my work on track. I want to express my gratitude to Principal, Parul Institute of Pharmacy and Research, Parul University for helping and providing necessary facilities for my work.

CONFLICT OF INTEREST:
Authors listed into the article suggest no conflict of interest.

AUTHOR'S CONTRIBUTION:
Each author contributed in the work is mentioned.

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CITATION OF THIS ARTICLE

Vaishnavi Hyalij and Deepanti Gajjar. Overview of Analytical Techniques for the Evaluation of Lidocaine Hydrochloride and Diltiazem Hydrochloride. Bull. Env. Pharmacol. Life Sci., Vol 14[5] April 2025: 31-39