WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
Coden USA: WJPRAP Impact Factor 8.63
Volume 15, Issue 6, 376-384. Review Article ISSN 2277–7105

REVIEW ON ANALYTICAL METHODS DEVELOPMENT AND VALIDATION OF LINAGLIPTIN AS AN ANTIDIABETIC AGENT

Nir D. Solanki*, Krupal J. Chaudhary, Manisha M. Parmar, Het R. Soni, Dr. Khushbu K. Patel, Jinal Goswami

Student of Pharmaceutical Quality Assurance, Shri Sarvajanik Pharmacy Collage, Gujarat Technological University, Arvind Baug Mehsana-384001, Gujarat, India.

Article Received on 11 Feb. 2026,
Article Revised on 03 March 2026,
Article Published on 16 March 2026,

https://doi.org/10.5281/zenodo.19044347

***Corresponding Author**

Nir D. Solanki

Student of Pharmaceutical Quality Assurance, Shri Sarvajanik Pharmacy Collage, Gujarat Technological University, Arvind Baug Mehsana-384001, Gujarat, India.

How to cite this Article: Nir D. Solanki*, Krupal J. Chaudhary, Manisha M. Parmar, Het R. Soni , Dr. Khushbu K. Patel, Jinal Goswami. (2026). Review On Analytical Methods Development And Validation of Linagliptin As An Antidiabetic Agent. World Journal of Pharmaceutical Research, 15(6), 376–384.

ABSTRACT

Diabetes mellitus significantly impacts global health, accounting for millions of deaths annually. The development of Dipeptidyl peptidase-4 (DPP-4) inhibitors, particularly linagliptin, offers a promising therapeutic pathway for managing type 2 diabetes. Linagliptin is distinguished by its unique non-renal elimination route, ensuring consistent dosing without adjustment for patients with renal impairment. The aim of this work is to study the pharmacological profile of linagliptin and review the various analytical methods used for its estimation in bulk and pharmaceutical formulations. This paper systematically reviews existing analytical techniques, including UV-Visible Spectroscopy, High-Performance Liquid Chromatography (HPLC), High-Performance Thin-Layer Chromatography (HPTLC), and Liquid Chromatography-Mass Spectrometry (LC-MS). Furthermore, it highlights method development strategies and validation parameters aligned with international regulatory guidelines.

INTRODUCTION

Overview of Diabetes Mellitus

Diabetes mellitus is a widespread endocrine disorder characterized by chronically elevated blood sugar levels. This condition arises either because the pancreas fails to produce sufficient insulin or because the body's cells become unresponsive to the insulin's effects.[1-4]

- **Symptoms:** Classic indicators include polydipsia (excessive thirst), polyuria (excessive urination), polyphagia (excessive hunger), unintended weight loss, blurred vision, fatigue, and dry skin.
- **Global Impact:** Diabetes is responsible for approximately 4.2 million deaths every year, with around 1.5 million deaths directly linked to untreated or poorly managed cases.

Types of Diabetes

- **Type 1 Diabetes:** Results from autoimmune destruction of the pancreatic beta-cells. It typically occurs in children and adolescents and frequently presents with ketoacidosis due to a rapid rate of beta-cell destruction.
- **Type 2 Diabetes:** Occurs when the body cannot use insulin correctly, leading to sugar accumulation in the blood. This can happen because the pancreas doesn't make enough insulin to help sugar enter the cells. Currently, there is no cure for type 2 diabetes.

DPP-4 Inhibitors

Dipeptidyl peptidase-4 (DPP-4) inhibitors, also known as gliptins, represent a relatively new class of oral antidiabetic agents. They are usually prescribed for people with type 2 diabetes who have not responded well to drugs such as metformin and sulphonylureas. Examples include Sitagliptin, Saxagliptin, Linagliptin, Alogliptin, Vildagliptin, and Teneligliptin.

Drug Profile: Linagliptin

Linagliptin is a novel, highly selective, and potent DPP-4 inhibitor that has emerged as a valuable therapeutic option in the management of T2DM. It was first approved by the FDA on May 02, 2011, under the brand name Tradjenta (Generic: Linagliptin).[4-7,12]

Chemical and Physical Properties

- **IUPAC Name:** 8-[(3R)-3-aminopiperidin-1-yl]-7-(but-2yn-1-yl)-3-methyl-1-(4-methyl-quinazolin-2ylmethyl)-3,7-dihydro-purine-2,6-dione.
- **Chemical Formula:** C25H28N8O2.
- **Molecular Weight:** 472.05 gm/mol.
- **Appearance:** White to off-white powder.
- **Solubility:** Freely Soluble in Water and Methanol.
- **Melting Point:** 141 - 146 °C.**2.2 Pharmacokinetics and Dosing**

Linagliptin exhibits a predominantly non-renal route of elimination, with the majority of the drug excreted via the hepatobiliary system. This property eliminates the need for dose adjustments in patients with renal or hepatic impairment.

- **Absorption:** Oral, bioavailability ~30%.
- **Half-Life:** ~12 hours.
- **Dosing:** 5 mg per day (Generally well tolerated).

Analytical Methods Overview

Analytical methods are systematic approaches for examining and interpreting data. These methods work by breaking down complex information into smaller, more manageable components. The majority of analytical techniques fall into instrumental or non-instrumental categories.

Analytical method validation is a systematic process to confirm that an analytical method is suitable for its intended purpose. Validation parameters include Precision, Specificity, Accuracy, Linearity, Limit of Detection, Range, Limit of Quantification, and Robustness[8-11].

Review of Literature on Linagliptin Estimation

Table 1: Official method of estimation of Linagliptin.

SR NO.	OFFICIAL IN	METHOD	DESCRIPTION	REF. NO
1.	Indian Pharmacopoeia (2022)	Liquid Chromatography (API and Tablet)	**Stationary phase:** A stainless-steel column (250 x 4.6 mm, 5 µm) packed with octadecylsilane bonded to porous silica. **Mobile phase:** Acetonitrile: Phosphate buffer pH 3.0 (40:60% v/v) **Flow rate:** 1.0 mL/min **Wavelength:** 225 nm **Injection volume:** 20 µL	13

Table 2: Reported methods for estimation of Linagliptin.

SR NO.	METHOD	DESCRIPTION	REF. NO
UV VISIBLE SPECTROSCOPY METHOD			
1.	First Order Derivative Method for Linagliptin in bulk and pharmaceutical formulation	**Solvents:** Methanol **Wavelength:** 299 nm **Linearity:** 5-25 µg/mL	[14]
2.	Method A (Absorbance), Method B (AUC) for analytical method development and	**Solvent**: Methanol **Wavelength:** 294 nm (Method A), 289-299 nm (Method B**)**	[15]

	validation of Linagliptin	**Linearity:** 2-12 µg/mL	
3.	Assay method for Estimation of linagliptin in bulk and marketed dosage form	**Solvent:** Methanol **Wavelength**: 294 nm **Linearity:** 5-30 µg/mL	[16]
4.	Estimation method for single linagliptin	**Solvent**: Methanol: Water (50:50% v/v) **Wavelength:** 241 nm **Linearity**: 10-35 µg/mL	[17]
5.	Simultaneous Estimation of Linagliptin and Metformin HCl by UV Spectrophotometric Method	**Solvents:** Distilled Water **Wavelength:** Linagliptin: 298 nm, Metformin: 232 nm **Linearity:** Linagliptin: 2.5-12.5 µg/mL, Metformin: 500-2500 µg/mL	[18]
6.	Estimation of Linagliptin with Metformin by Stability Indicating UV- Spectroscopic Method	**Solvents:** Distilled water or Methanol **Wavelength:** Linagliptin:294.4 nm, Metformin:230.4 nm **Linearity:** Linagliptin**:** 10-40 µg/mL, Metformin:2-14 µg/mL	[19]
7.	Estimation of Linagliptin with Empagliflozin by UV-Spectroscopic Method	**Solvents:** Distilled water **Wavelength**: Linagliptin:238 nm, Empagliflozin: 221 nm **Linearity**: Linagliptin: 2.5-30 µg/mL, Empagliflozin: 2.5-30 µg/mL	[20]
8.	Simultaneous Estimation of linagliptin with Empagliflozin by UV- Spectroscopic Method	**Solvents:** water **Wavelength:** Linagliptin:277 nm, Empagliflozin: 233 nm **Linearity**: Linagliptin 2-6 µg/mL Empagliflozin: 5-15 µg/mL	[21]
9.	Different spectroscopy methods for estimation of linagliptin	**Solvents:** 0.1M HCl and 0.1M NaOH **Wavelengths:** 295 nm & 315 nm **Linearity:** 2-12 µg/mL	[22]
HIGH PERFORMANCE LIQUID CHROMATOGRAPHY			
1.	Development and Validation of RP-HPLC Method for the estimation of Linagliptin in Bulk and its Tablet Dosage Form	**Stationary phase**: C_{18} column (250 x 4.6 mm, 5µ) **Mobile phase**: Acetonitrile: Phosphate Buffer pH 3.0 (60:40% v/v**)** **Flow rate:** 1 mL/min **Wavelength:** 293 nm **Linearity:** 5-25 µg/mL	[23]
2.	A validated stability-indicating HPLC assay Method for Linagliptin API and tablets	**Stationary phase**: Inertsil ODS 3V C_{18} column (250 x 4.6mm, 5µ) **Mobile phase**: Acetonitrile: 0.1% Orthophosphoric acid in water (55:45 % v/v) in isocratic elution **Flow rate:** 1.0 mL/min **Wavelength:** 225 nm **Linearity:** 1-7.5 µg/mL	[24]
3.	RP-HPLC Method for the Simultaneous Estimation Linagliptin and Metformin Hydrochloride in Combined tablet dosage forms	**Stationary phase**: Zodiac C_{18} column (250 × 4.6 mm, 5µm) **Mobile phase**: Acetonitrile: Phosphate buffer pH-4.8 (50:50% v/v) **Flow rate:** 1.0 mL/min	[25]

		Wavelength: 272 nm **Linearity:** Linagliptin: 10-50 µg/mL, Metformin: 2-10 µg/mL	
4.	RP-HPLC Method for the estimation of linagliptin as single formulation	**Stationary phase:** C_{18} column (150 x 4.6 mm, 5µm) **Mobile phase:** Mixture of 0.02 M potassium dihydrogen phosphate: Acetonitrile (70:30% v/v) **Flow rate**: 1.2 mL/min **Wavelength:** 226 nm	[26]
5.	HPLC Assay Method Development and Validation of Linagliptin in Tablet Dosage Form.	**Stationary phase:** C_{18} column (150 x 4.6 mm, 5 µm) **Mobile phase:** Mixture of Phosphate buffer: Methanol pH-4.5 (70:30 % v/v) **Wavelength**: 292 nm **Linearity:** 1mL/min	[27]
HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY			
1.	Development and validation of stability indicating High-Performance Thin-Layer Chromatography Method for estimation of Linagliptin in bulk and in pharmaceutical formulation	**Stationary phase**: Aluminium plates pre-coated with Silica gel $60F_{254}$ **Mobile phase**: Toluene: Ethyl Acetate: Methanol (6:3:1 % v/v/v) **Wavelength:** 298 nm **Linearity:** 100-600 ng/band	[28]
2.	Validated HPTLC method for simultaneous estimation of metformin hydrochloride and Linagliptin in bulk drug and formulation	**Stationary phase**: Aluminium plates pre-coated with Silica gel $60F_{254}$ **Mobile phase**: Dichloromethane: Methanol: Glacial Acetic Acid (9:1:0.1 % v/v/v) **Wavelength:** 230 nm **Linearity:** Linagliptin: 100-500 ng/spot, Metformin: 20000-100000 ng/spot	[29]
3.	simultaneous estimation and stability indicating study of metformin HCl and linagliptin in pharmaceutical formulation.	**Stationary phase:** Aluminium plates pre-coated with silica gel $60GF_{254}$ **Mobile phase:** Acetone-methanol-toluene-formic acid 4:3:2:1 % (v/v/v/v) **Wavelength:** 259 nm	[30]
ULTRA PERFORMANCE LIQUID CHROMATOGRAPHY			
1.	Development and Validation of ultraperformance liquid chromatography (UP-LC) method for estimation of a new anti-diabetic drug Linagliptin in bulk and its tablet formulation.	**Stationary phase:** SB-C_{18} columnn (50 × 2.1 mm, 1.8 µm) **Mobile phase:** Mixture of Acetonitrile: 0.01M Potassium phosphate buffer pH-4 (70:30% v/v) **Wavelength:** 292 nm **Flow rate:** 0.3 mL/min	[31]
2.	Estimation of linagliptin by RP-UPLC method	**Stationary phase:** Chiralpak AD-H (250 x 4.6 mm, 5 µm) **Mobile phase:** Ethanol: Methanol: Diethylamine (90:10:0.1 % v/v/v)	[32]

		Wavelength:225 nm	
3.	Estimation of linagliptin by RP-UPLC method	**Stationary phase:** Thermo Scientific® RP-8 (100 mm × 4.6 mm; 5µm) **Mobile phase:** A: 0.1% Formic Acid B: Acetonitrile **Wavelength:** 294 nm	[33]
MASS SPECTROSCOPY LIQUID CHROMATOGRAPHY			
1.	Bioanalytical method development and validation of linagliptin in plasma through LCMS method	**Stationary phase:** Waters, X Bridge, C_{18} column, (50 × 4.6 mm, 5µm) **Mobile phase:** Acetonitrile: 0.1% formic acid (90:10 % v/v)	[34]
2.	Ultrahigh performance liquid chromatography - tandem mass spectrometry method for quantification of linagliptin in human plasma.	**Stationary phase:** Gemini C_{18} column (100 × 4.6 mm, 3µ) **Mobile phase:** 10 mm Ammonium formate: methanol (20:80 % v/v)	[35]
3.	Pharmacokinetic interaction between linagliptin and tadalafil in healthy Egyptian males using a novel LC–MS method.	**Stationary phase:** Zorbax Eclipse XDB C_{18} column (150 × 4.6 mm, 5 µm) **Mobile phase:** Methanol:0.05% aqueous formic acid (50:50 % v/v)	[36]

CONCLUSION

Diabetes mellitus significantly impacts health, causing millions of deaths annually. DPP-4 inhibitors, particularly linagliptin, offer promising management for type 2 diabetes, with unique benefits like non-renal elimination and consistent dosing, thus enhancing treatment adherence in patients, especially those with renal impairment. The literature review details various analytical methods for estimating Linagliptin, highlighting techniques like HPLC, UV spectroscopy, and mass spectrometry. The aim is to understand Linagliptin's pharmacological profile and develop standardized methods for its validation conforming to regulatory and ICH guidelines.

REFERENCES

1. World Health Organization (WHO). Introduction of diabetes [Internet], July 2025.
2. National Institute of Health. Types of diabetes [Internet], July 2025.
3. Overview of diabetes [Internet], July 2025.
4. Diabetes.co.uk. DPP-4 inhibitors [Internet], July 2025.
5. MedlinePlus. Linagliptin introduction [Internet], July 2025.
6. Australian Public Assessment Report (AusPAR). History of linagliptin [Internet], July 2025.
7. Frontiers. Mechanism of action of linagliptin [Internet], July 2025.

8. Riley CM, Rosanske TW. Progress in pharmaceutical and biomedical analysis: Development and validation of analytical methods. *J Pharm Sci.*, 2010.
9. Vidhyasagar G. Instrumental method of drug analysis. PharmaMed Press, 2010.
10. Chatwal GR, Anand SK. Instrumental methods of chemical analysis. HPH Pvt Ltd., 2018.
11. International Conference on Harmonisation (ICH) of Technical Requirements for Registration of Pharmaceuticals for Human Use. Validation of analytical procedures.
12. DrugBank. Linagliptin [Internet], September 2024.
13. Indian Pharmacopoeia. Vol. II. Ghaziabad (India): Indian Pharmacopoeia Commission, 2022.
14. Sruthi PN, Rao DV, Kumar JV. Development and validation of UV spectrophotometric method for the estimation of linagliptin. *Int J Pharm Sci Res.*, 2018.
15. Mounika B, Sravanthi J, Adukondalu D. Development and validation of UV spectrophotometric method for estimation of linagliptin. *Int J Pharm Sci Res.*, 2017.
16. Sujan B, Kaisar MM, Hossain MS. Development and validation of a simple and rapid UV spectrophotometric method for assay of linagliptin in bulk and marketed dosage form. *Indian J Novel Drug Deliv*, 2013; 5: 221–224.
17. Chandra KS, Sudhakar P, Mohan Rao T, Vijaya Babu P, Manikanta KA. A new UV method for determination of linagliptin in bulk and pharmaceutical dosage form. *Int J Pharm Bio Sci.*, 2013; 2: 1–6.
18. El-Bagary RI, Elkady EF, Ayoub BM. Spectrophotometric methods for the determination of linagliptin in binary mixture with metformin hydrochloride and in ternary mixture with metformin hydrochloride and pioglitazone hydrochloride. *Int J Biol Sci.*, 2013; 9(1): 41–49.
19. Sarif N, Niroush K, Jane TJ, Vipin P. Stability indicating UV spectrophotometric method for linagliptin and metformin in pharmaceutical dosage form. *Pharm Methods*, 2017; 8: 121–126.
20. Amrutiya FR, Patel BR, Patel JG, Vegad KL, Patel AS, Darji VC. Development and validation of first order derivative UV spectrophotometric method for determination of empagliflozin and linagliptin. *Int J Pharm Drug Anal.*, 2017; 5: 129–135.
21. Padmaja N, Veerabhadram G. Development and validation of analytical method for simultaneous estimation of empagliflozin and linagliptin in bulk drugs and combined dosage forms using UV-visible spectroscopy. *Der Pharm Lett.*, 2015; 7: 306–312.

22. Kavitha KY, et al. Difference spectrophotometric method for the determination of linagliptin in pharmaceutical formulations. *Asian J Pharm Clin Res.*, 2013; 6(3): 126–128.
23. Shah DA, Patel VB, Chhalotiya UK. Development and validation of a reversed-phase HPLC method for estimation of linagliptin in its tablet formulation. *Chron Young Sci.*, 2017; 8(2): 127–131.
24. Jain PS, Puranik MP, Surana SJ. Development and validation of a stability-indicating HPLC method for the determination of linagliptin in the bulk drug and in a pharmaceutical dosage form. *J Adv Pharm Technol Res.*, 2013; 4(3): 154–159.
25. Achaiah G, Sreeramulu J. RP-HPLC method for simultaneous estimation of metformin and linagliptin in bulk and tablet dosage form. *Int J Pharm Sci Res.*, 2014; 5(11): 4945–4950.
26. Archana V, Sriram N, Gayasuddin M. Method development and validation of RP-HPLC method for determination of new antidiabetic agent linagliptin in bulk and in pharmaceutical formulation. *Int J Med Chem Anal.*, 2013; 3: 1–5.
27. Sneha RB, Anvesha VG, Krishna RG. Quality by design based HPLC assay method development and validation of linagliptin in tablet dosage form. *Eur J Med Res.*, 2017; 4: 486–494.
28. Dewani AP, Bakal RL. Development and validation of a stability-indicating HPTLC method for estimation of linagliptin in the bulk drug and in a pharmaceutical dosage form. *J Planar Chromatogr Mod TLC.*, 2014; 27(3): 200–204.
29. Prajapati RN, Patel CN. Development and validation of HPTLC method for the simultaneous estimation of linagliptin and metformin hydrochloride in their combined dosage form. *J Pharm Sci Bioscientif Res.*, 2016; 6(3): 329–335.
30. Rajasekaran A, Kavitha R, Arivukkarasu R. Development and validation of HPTLC method for simultaneous estimation and stability indicating study of metformin HCl and linagliptin in pharmaceutical formulation. *World J Pharm Sci.*, 2014; 2(4): 317–327.
31. Nidhi D, Singh GN, Anchal T, Rakesh B, Raghav CS. Development and validation of ultra performance liquid chromatography (UPLC) method for estimation of a new anti-diabetic drug linagliptin in bulk and its tablet formulation. *Int J Chem.*, 2014; 5(3): 1136–1139.
32. Cholleti VK, Pasula A, Pavan KVY, Ravindra K, Nitin HD. A new and enantioselective chromatographic method for linagliptin on amylose coated chiral stationary phase. *Am J Anal Chem.*, 2016; 7: 556–567.

33. Raquel BHF, Jonathaline AD, Fla DF, Luis FSO, Michel MM, Marcelo DM. Biological safety studies and simultaneous determination of linagliptin and synthetic impurities by LC-PDA. *J Anal Methods Chem.,* 2019; 1–10.
34. Mahamad SSS, Arifa B, Saradhi NDVR. Bioanalytical method development and validation of linagliptin in plasma through LC-MS/MS. *Int J Bioassays.,* 2014; 3(07): 3146–3151.
35. Nagaraj KN, Sunil SJ, Rajendraprasad M, Sunil KS. An ultra high performance liquid chromatography–tandem mass spectrometry method for quantification of linagliptin in human plasma. *RSC Adv.,* 2016; 6(71): 66756–66766.
36. Sara SM, Eman IEK, Magda AB, Dalia AH. Pharmacokinetic interaction between linagliptin and tadalafil in healthy Egyptian males using a novel LC–MS method. *Bioanalysis*, 2019; 1–10.