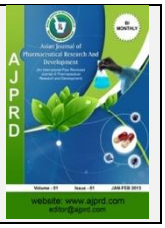
Available online on 15.10.2025 at http://ajprd.com

Asian Journal of Pharmaceutical Research and Development

Open Access to Pharmaceutical and Medical Research



Open Access

Review Article

Review on Analytical Method Validation of Nafithromycin

Nishtha Patel, Ayushi Patel, Zarana Pandya, Helly Shah

A-One Pharmacy College, SNME Campus, Naroda-Dahegam Road, Nr.S.P.Ring Road, Daskroi, Enasan, Ahmedabad, Gujarat.

A B S T R A C T

Background: Nafithromycin (synonym WCK-4873), marketed under the brand name Miqnaf, represents India's first indigenously developed antibiotic targeting drug-resistant infections. Wockhardt Limited developed Nafithromycin with financial and technical support from the Biotechnology Industry Research Assistance Council (BIRAC). The Central Drugs Standard Control Organisation (CDSCO) has approved it for the treatment of community-acquired bacterial pneumonia (CABP).The drug achieves effective lung concentrations and delivers therapeutic out comes with a three-day oral regimen, improving patient compliance while reducing treatment time.

Methods: Researchers validated Nafithromycin analytically using three key methods. They employed the RP-HPLC method with a C18 column, anisocratic mobile phase of buffer and acetonitrile (60:40), and UV detection. They applied the RP-UPLC method with a BEHC18 column, a gradient mobile phase of 0.1%formicacidinwaterandacetonitrile, and PDA detection. They used the UHPLC-MS method with a C18 column, an isocratic mobile phase of 0.1% formic acid and acetonitrile (30:70), and mass spectrometry detection.

Conclusion: Nafithromycin (Miqnaf) emerges as a reliable, indigenous antibiotic for community-acquired bacterial pneumonia. Analytical validation through RP-HPLC, RP-UPLC, and UHPLC-MS confirms accuracy and robustness. These findings ensure quality, safety, and efficacy, highlighting Nafithromycin's promise as a short-course therapy against drug-resistant respiratory pathogens and a milestone in antibiotic innovation.

Key words: Nafithromycin; Miqnaf; WCK-4873; community-acquired bacterial pneumonia (CABP); drug-resistant pathogens; RP-HPLC; RP-UPLC; UHPLC-MS; antibiotic innovation; short-course therapy; Wockhardt Limited; BIRAC.

A R T I C L E I N F O: Received 11 Feb 2025; Review Complete 05 April 2025; Accepted 18 August 2025. ; Available online 15 Oct. 2025

Cite this article as:
Patel N, Patel A, Pandya Z, Shah H, Review on analytical method validation of Nafithromycin, Asian Journal of Pharmaceutical Research and Development. 2025; 13(5):97-101, DOI: http://dx.doi.org/10.22270/ajprd.v13i5.1631

*Address for Correspondence:
Nishtha Patel, A-One Pharmacy College, SNME Campus, Naroda-Dahegam Road, Nr.S.P.Ring Road, Daskroi, Enasan, Ahmedabad, Gujarat

INTRODUCTION

Nafithromycin is India's first indigenous macrolide antibiotic, which belongs to the sub class of ketolides. Wockhardt developed it in India with support from BIRAC. It is a semi synthetic derivative of erythromycin, structurally featuring as tablel actonering and a thiadiazole-pyridine side chain, which enhances lung penetration. This innovation marks a milestone in pharmaceutical development and strengthens India's hand in the global fight against AMR.

The drug targets community-acquired bacterial pneumonia (CABP), including drug-resistant infections, and is a major step in combating global antimicrobial resistance (AMR). Nafithromycin shows strong activity against awide range of gram-positive bacteria,

including Streptococcus pneumoniae, Streptococcus pyogenes, and atypical pathogens such as Mycoplasma pneumoniae and Chlamydophila pneumoniae. In December 2023, Wockhardt completed a Phase 3 clinical trial, demonstrating that a three-day oral regimen of Nafithromycin achieved a 96.7% clinical cure rate, comparable to the traditional seven-day Moxifloxacin treatment at 94.5%. The drug exhibited an eight-fold higher lung exposure and 10- to 100-fold greater potency against respiratory pathogens, including those resistant to azithromycin and levofloxacin. Importantly, the trial reported no severe adverse events; most side effects (like naus or mild GIissues) were mild and largely unrelated to the drug. Nafithromycin offers a Short-course (three-day) oral regimen, which is a unique advantage over traditional antibiotics.

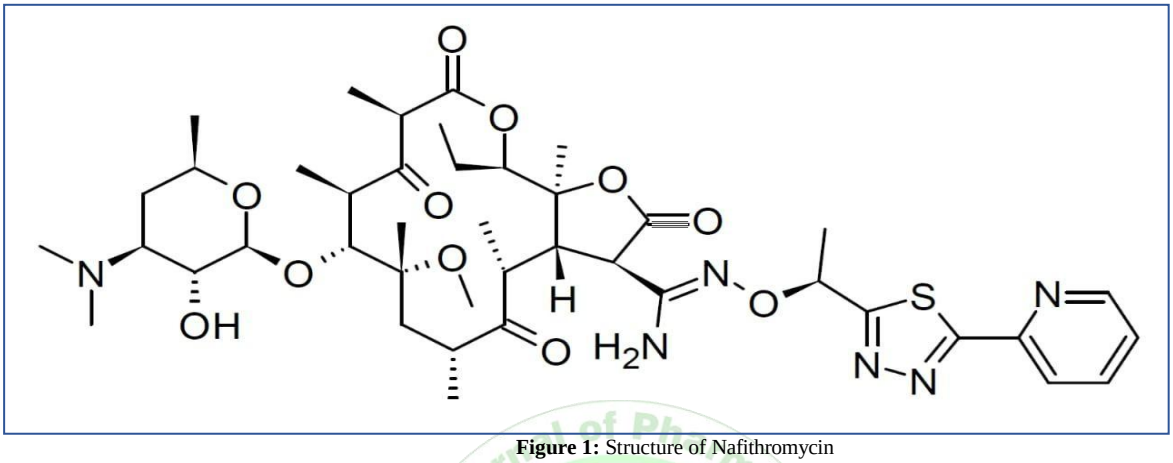


Figure 1: Structure of Nafithromycin

IUPACNAME:-(1S,2R,5R,7R,8R,9R,11R,13R,14S,15R)-8-[(2S,3R,4S,6R)-4(dimethylamino)-3-hydroxy-6-methyloxan-2-yl]oxy-2-ethyl-9-methoxy-1,5,7,9,11,13-hexamethyl-4,6,12,16-tetraoxo-N'-[(1S)-1-(5-pyridin-2-yl-1,3,4-thiadiazol-2-yl)ethoxy]-3,17dioxabicyclo[12.3.0]heptadecane-15-carboximidamide.

Nafithromycin, with a Molecular formula of C42H62N6O11Sanda molecular weight of ~859.05 g/mol, is a semisynthetic ketolide antibiotic featuring a 15-membered macrolide ring at its core. It features a key structural modification: a keto group replaces the cladinose sugar at the C3 position, which helps it overcome the resistance that traditional macrolides often face. The C3 keto function group prevents the Erm enzyme from binding and methylating the ribosomal binding site, which would otherwise block macrolide activity. Additionally, adistinctive thiadiazole- pyridine side chain enhances its potency, lung penetration, and pharmacokinetics profile. Various covalent bonds (single, double, and aromatic), along with polar covalent and hydrogen bonds, stabilize the molecule and support its biological activity. The side chain contains anoxime functional group (-C=NOH), which enhances activity against resistant bacteria. A unique side chain, which consists of a thiazolidine ring linked to a pyridine ring, attaches to the C11-C12 positions. This side chain is a key innovation. It enhances the drug's binding to the bacterial ribosomes by providing additional hydrogen bonding and hydrophobic interactions.

OVERVIEW

- Nafithromycin marks amilestonein public health in India."
- This medicine is ten times more effective than current treatments like Azithromycin and offers a three-day treatment regimen, significantly shortening the recovery time while improving patient outcomes.

· Nafithromycin marks a key milestone against AMR, offering hope for treating multidrug- resistant infections like sinusitis and strep throat.

Drug profile Nafithromycin:

Parameters	Details
Drug Name	Nafithromycin
Class	Lactone ketolides
Therapeutic indication	Treatment of community-acquired bacterial pneumonia (CABP) in adults caused by multi drug- resistant pathogens, including Streptococcus pneumoniae.
Dosage form	Oral tablets (400mg)
Pharmacokinetics	**Absorption:** - well-absorbs orally. **Distribution:**-Reaches high, sustained concentration in lung tissue. **Metabolism:** - metabolized by the liver. **Excretion:**-Mainly viabile/faeces; minimal renal elimination.
Half-Life	Long half-life (9.16to14.4hours) Supports once-daily dosing

Pharmacological action	Bactericidal, it is also highly potent against respiratory pathogens.
Advantages	Short treatment duration. Combats Antimicrobial Resistance (AMR). High lung penetration, effective in respiratory infections.
Adverse effect	Well-tolerated with minimal gastrointestinal side effects.
Contraindication	Allergy to Nafithromycin QT interval prolongation Myasthenia gravis

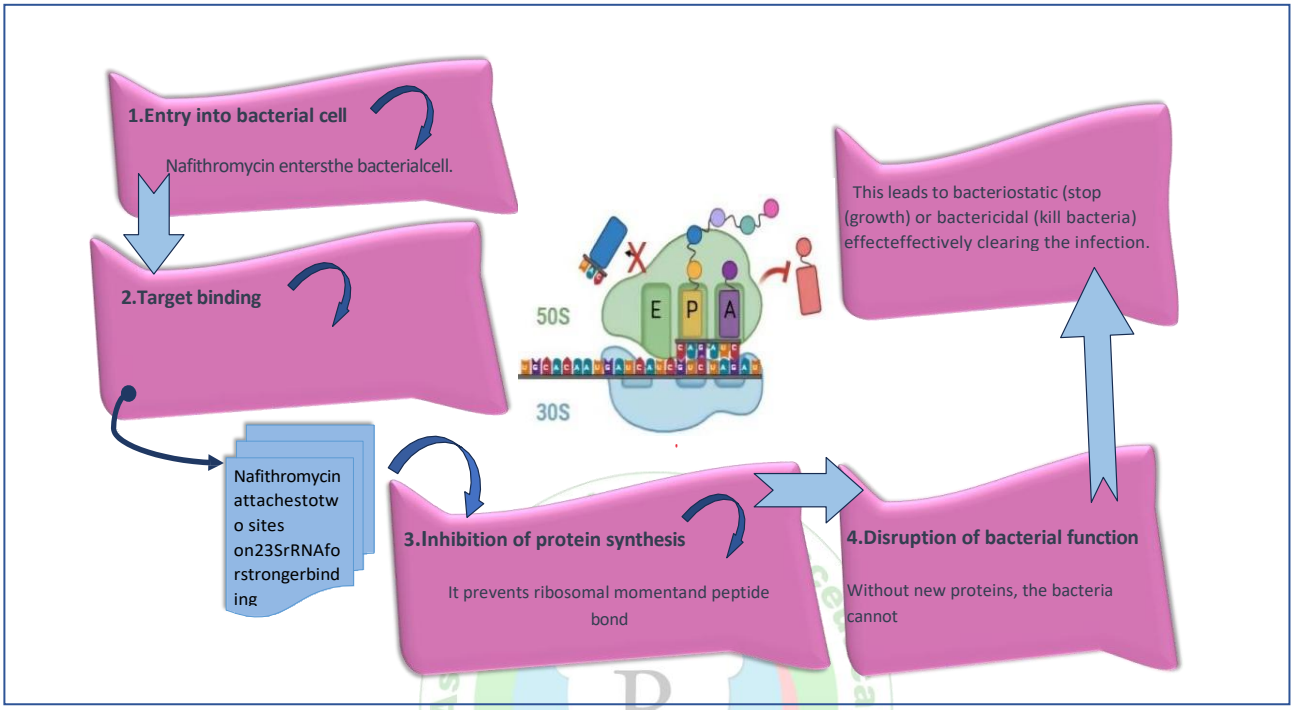


Figure 1: Mechanism of Action of Nafithromycin

Figure 2: Nafithromycin Tablet

BRAND NAME	Miqnaf
GENERIC NAME	Nafithromycin
DOSAGE STRENGTH	400 mg
ROUTE OF ADMINISTRATION	Oral

Researchers studied IV or suspension formsintrials, but companies have not commercialized them because routine treatment does not require them.

So, companies market only tablets because theygive the best balance of effectiveness, safety, stability, cost, and patient compliance.

Analytical validation method:

Sr. No.	Drugs	Method	Description	Reference no
1.	Nafithromycin	RP-HPLC	**Mobile phase:-**Buffer(KH_2PO_4): Acetonitrile (60:40 v/v) **Column:-**C18(250×4.6mm*5µm) **Flow rate:** - 1.0ml /min **Temperature:** - Between 25-40˚ **Detection:** - UV at 230-3-5 nm	4
2.	Nafithromycin	RP-UPLC	**Mobile phase:** - Gradient (A) 0.1 % formic acid in water (B) Acetonitrile10-90 % B in 15 min. **Column:-**BEHC18 (100×2.1mm*1.7µm) **Flow rate:** -0.3mL/min **Detection:-**PDA230nm	9
3.	Nafithromycin	UHPLC- MS	**Mobile phase:-**(A) 0.1%formicacidin water, (B) Acetonitrile: isocratic 30:70 **Column:** - C18 (50×2.1mm*1.7µm) **Flow rate:** - 0.25 mL/min **Detection:-**MS (depends on analyte)	6

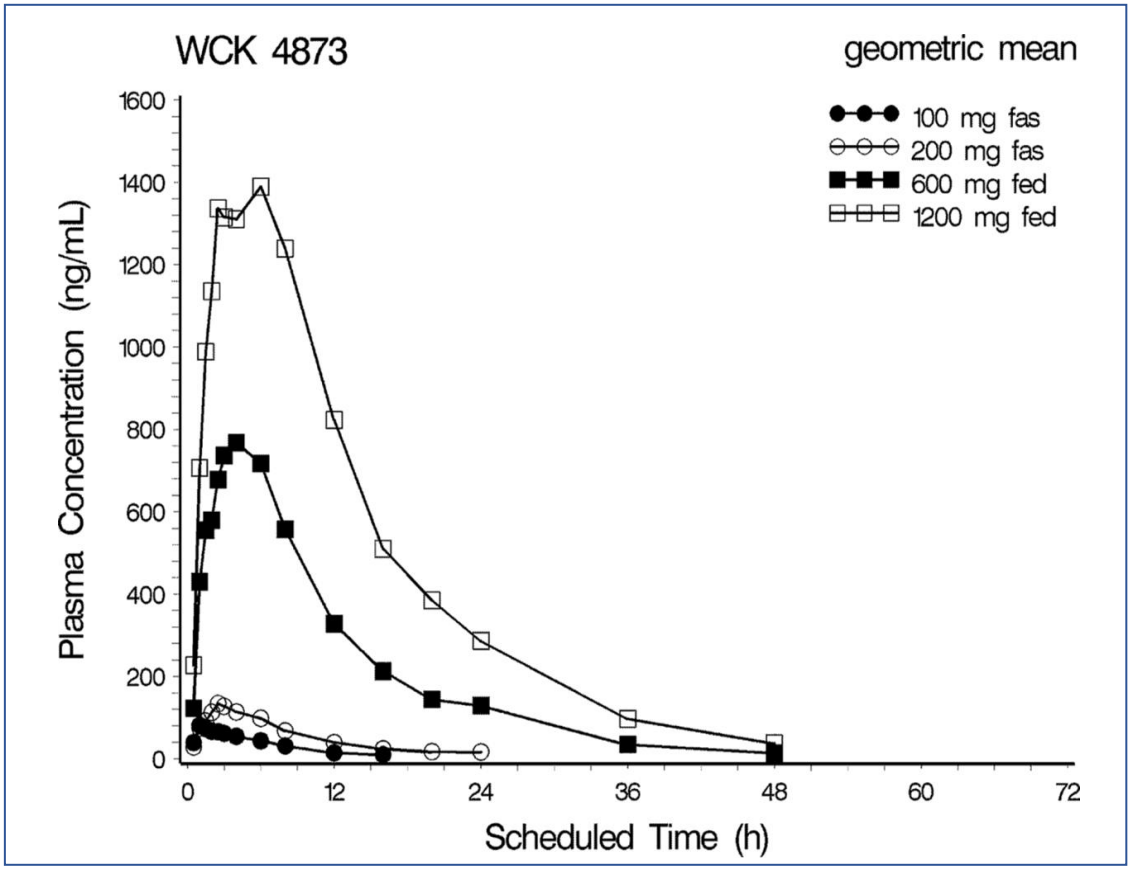


Figure: Chromatogram of Nafithromycin in human plasma using HPLC method.

CONCLUSION:

The presented review on the analytical method validation of various methods reported on Nafithromycin. Some Introduction on Nafithromycin (WCK 4873), a novel ketolide antibiotic, represents a significant advancement in the fight against antimicrobial resistance (AMR), a growing global health crisis. Developed to address multidrug-resistant (MDR) respiratory pathogens, such as Streptococcus pneumoniae and Haemophilus influenzae, Nafithromycin exhibits enhanced activity compared to older macrolides. The researcher mainly used LC-MS and then employed RP-HPLC and RP-UPLC to analyse Nafithromycin. Particularly, using LC-MS played a fundamental role in its drug development. These methods proved to be sensitive, accurate and specific for the reliable quantification of Nafithromycin. These methods established regulatory standards, ensuring the High-quality data necessary to support Nafithromycin as a new treatment for combating antibiotic-resistant bacterial infection.

ACKNOWLEDGEMENT:-

The authors sincerely express their gratitude to the A-One Pharmacy College, Gujarat Technological University, Gujarat, Ahmedabad, INDIA, for providing the necessary facilities and support to carry out this review work.

REFERENCE:

1. Bhawsar, S.,Tadiparthi, R., Kayastha, A.K. et al. Nafithromycin (MIQNAF®): ultramodern lactone ketolide designed to treat community-acquired bacterial pneumonia (CABP). Med Chem Res 33, 1715–1733(2024)
2. Paliwal, S., Chaudhary, K., Agarwal, U. et al. Novel Macrolide Antibiotic Nafithromycin for Tackling Emerging Resistance in Respiratory Pathogens Encountered in India. Curr Microbiol 82, 465 (2025)
3. Dinesh Kumar, Debayan Sil, Rashmi Ghosh, Kumari Komal, Prachi Pandey, Ghanshyam Das Gupta, Manish Kumar, Nafithromycin: a potential ketolide antibiotic for combating antimicrobial resistance in community-acquired bacterial pneumonia, Next Research, Volume 2, Issue3, 2025,100592,ISSN3050-4759,
4. Patil, K. R., Yeole, R. D., de Zwart, M., & Pruim, P. (2019). Simultaneous Determination of Novel Ketolide Antibiotic Nafithromycin and its Major Metabolite in Human Plasma Using Liquid Chromatography Tandem Mass Spectrometry. Bioanalysis,11(19),1767-1776.
5. Iwanowski P, Bhatia A, Gupta M, Patel A, Chavan R, Yeole R, Friedland D.2019.Safety, Tolerability, and Pharmacokinetics of Oral Nafithromycin (WCK4873) after Singleor Multiple Doses and Effects of Food on Single-Dose Bioavailability in Healthy Adult Subjects. AntimicrobAgents Chemother 63:10.1128/aac. 01253 19
6. Mustafa, A.M., Bastawesy,G.A., Hatem,S. et al. Polyphenolic protection: the role of mangiferinin mitigating neuro degeneration and neuro inflammation. Inflammo pharmacol 33,4535–4552(2025)
7. Wockhardt Limited. (2025, January 2). Indian Drug Regulator Approves Wockhardt's New Generation Oral Antibiotic Miqnaf® (Nafithromycin) forthe Treatment of Community-Acquired Bacterial Pneumonia (CABP).
8. Miqnaf (Nafithromycin) package insert. Wockhardt Limited; 2025.Ahirrao, V.K., Rane, V.P., Patil, K.R.et al. Identification and Quantification of Related Impurities of a Novel Ketolide Antibiotic Nafithromycin. Chromatographia 82,1059–1068 (2019) .