RESEARCH ARTICLE

Eco-Friendly Quantitative Assessment of Amlodipine Besylate in its Microspheres and Commercial Tablets using ATR-FTIR Spectroscopy

Annapoorani Arjunan[1], Ruby Stanlyraj[1*], Kumar Mohan[2]

[1]*Department of Pharmaceutical Chemistry, Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation, (Deemed to be University), Salem-08, Tamil Nadu, India*
[2]*Principal and Professor, Department of Pharmaceutical Chemistry, Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation, (Deemed to be University), Salem, Tamil Nadu, India*

Received: 19th Aug, 2025; Revised: and; Accepted: 8th Sep, 2025; Available Online: 25th Sep, 2025

ABSTRACT

Aim: Amlodipine Besylate (AMD) is an ACE inhibitor widely used as an antihypertensive drug. The present study was aimed at developing a green, solvent-free, and nondestructive quantification of AMD with the aid of Attenuated Total Reflectance Fourier Transform Infrared (ATR FT-IR) Spectrometry in bulk, tablet, and its polymer-based microsphere formulations.

Background: To develop a nondestructive, eco-friendly analytical technique for the rapid analysis using an ATR FTIR spectrophotometer.

Materials and method: This approach entails gauging the absorbance of the carbonyl group (C=O) peak of AMD at 1676 cm^{-1} as the optimized wavenumber by measuring the absorbance at various concentration levels.

Results and discussion: The Beer's law linearity range was between 0.1% w/w and 10.0% w/w with the R^2 value of 0.9986. The concentrations of lab-made amlodipine microspheres and marketed formulations were found to be 99.96%, 100.69%, and 99.49%, respectively. The method's limit of detection (LOD) and limit of quantification (LOQ) were determined to be 0.0468 mg and 0.1419 mg, respectively. Validation of the proposed method for tablets revealed an RSD of less than 2%, and recovery levels were between 99.18 and 99.50%.

Conclusion: The presented research work was novel, since there was no ATR-FTIR spectroscopy method reported earlier for its quantification. Further, the solvent-free and non-destructive analysis makes it unique in comparison to the conventional spectroscopic and chromatographic methods. Moreover, it is rapid, accurate, and precise for the routine analysis of amlodipine in its bulk, polymer-based, and tablet dosage forms.

Keywords: Analysis, ATR-FTIR, Amlodipine, Microspheres, Quantification, Validation.

How to cite this article: Annapoorani Arjunan, Ruby Stanlyraj, Kumar Mohan. Eco-Friendly Quantitative Assessment of Amlodipine Besylate in its Microspheres and Commercial Tablets using ATR-FTIR Spectroscopy. International Journal of Drug Delivery Technology. 2025;15(3):1094-1100. doi: 10.25258/ijddt.15.3.26

Source of support: Nil.

Conflict of interest: None

INTRODUCTION

In terms of its chemical composition, Amlodipine Besylate (AMB) (Figure 1) is the monobenzenesulphonate of 3-Ethyl-5-methyl (±)-2-[(2-aminoethoxy) methyl] -4-(2-chlorophenyl)-1, 4-dihydro-6-methyl-3, 5-pyridinedicarboxylate, and it serves as an anti-hypertensive and antianginal agent in besylate salt form. Figure 1 depicts the structure of Amlodipine besylate. Literature study revealed that various methods have been documented for determining it using UV-spectrophotometry[1-6], high-performance liquid chromatography (HPLC)[7,8], visible spectroscopy[9], voltammetry[10], and high-performance thin-layer chromatography (HPTLC)[11] techniques, but there is a lack of methods for its determination using infrared spectroscopic technique. Moreover, the literature review reveals that the distinctive feature of the proposed method, in contrast to reported UV, HPLC, and HPTLC analytical techniques, is its ability to direct real-time analysis of samples and require less sample preparation. The solvent-free analysis of this method makes it cost-effective and non-toxic. Therefore, a new analytical method was developed to assay amlodipine besylate in pure, microsphere, and tablet forms, aiming for simplicity, rapidity, and non-destructiveness by employing ATR FT-IR.

MATERIALS AND METHODS

Materials

Instruments used

The ATR FT-IR spectra were obtained using a Brucker ALPHA II Germany ATR-FTIR Spectrometer. The instrument was equipped with OPUS 2.0 software. Schimadzu digital weighing machine of 0.000 sensitivity was used.

Chemicals and Reagents

The reference standard of Amlodipine Besylate was received as a gift sample from M/s Dr. Reddy's Laboratories, Hyderabad. Single-dosage-form tablets of Amdipin (5.0 mg) and Norvasc (5.0 mg) were purchased from the local market in Salem. Potassium bromide of IR

**Author for Correspondence: ruby.s5902@gmail.com*

purity was procured from the E-Merck lab, Mumbai for recording the spectra. Sodium alginate and calcium chloride were of analytical grade.

Methods

Formulation of AMD Microspheres (AMDMS)

Ionotropic gelation techniques were used to formulate AMD-loaded microsphere[12]. Sodium alginate solution was prepared by dissolving 500mg sodium alginate in 50ml deionized water and heating at 60°C. Accurately 100mg of AMD was weighed dissolved uniformly in 50 mL of alginate solution at a temperature below 40°C under continuous stirring until a uniform, homogeneous mixture was formed. The resultant drug alginate mixture was sonicated for 10 minutes using a bath sonicator. The resulting homogeneous, bubble-free slurry dispersion was slowly delivered as drops through a 23-gauge syringe needle into 100 mL of a 2% calcium chloride solution. The mixture was stirred continuously to avoid the formation of aggregates. The AMD microspheres are allowed to cure for five minutes to remove any residual calcium chloride and securely filtered. The filtered microspheres were then washed thrice with 50 ml of distilled water. The purified microspheres were then dried at 40°C for 4 hours in a temperature-regulated oven, and the dried microspheres were stored.

Spectral Characteristics of Amlodipine Besylate

The chemical structure of AMD is based on a 1,4-dihydropyridine ring, with ethyl and methyl esters at positions 3 and 5, an aminoethoxymethyl group at position 2, and a (2-chlorophenyl) group at position 4. The functional group region was chosen to select the ideal wavenumber for analytical assessment of AMD. The spectra of standard AMD were recorded using an ATR-FTIR spectrophotometer by scanning between 4000 cm^{-1} and 400 cm^{-1} wavenumber. The average scanning sequence was 32 scans, and the resolution was 4 cm^{-1}. For calibration, IR pure KBr was used as a diluent to achieve different concentrations of AMD. The spectra were recorded by placing the AMD-KBr physical mixture on the sampling stage of the Brucker Alpha II ATR-FTIR spectrophotometer using the AMD API in the concentration range of 0.01% w/w to 100% w/w.

Selection and Optimization of Amlodipine Besylate Wavenumber

Amlodipine besylate API was scanned in the ATR-FTIR spectrophotometer in the absorbance mode and all the functional group peaks were assessed, and the C=O esters stretching frequency peak of wavenumber 1676 cm^{-1} was selected. The C=O carbonyl stretch at ~1676 cm^{-1} is chosen because it is an intense, diagnostic peak in a unique wavenumber region (~1600-1900 cm^{-1}) that is largely uncrowded by other functional groups, making it a key indicator for identifying carbonyl-containing compounds in a sample. Moreover, the C=O is uncommon in comparison to the common tablet and polymer-based microsphere excipients. Further it reduces the overlapping of absorbances of the formulation excipients. Figure 2 depicts the ATR-FTIR absorbance spectrum of amlodipine besylate. The optimized C=O peak was further fine-tuned by deriving the overlay absorbance spectrum for linear concentration (Figure 3 & Figure 4).

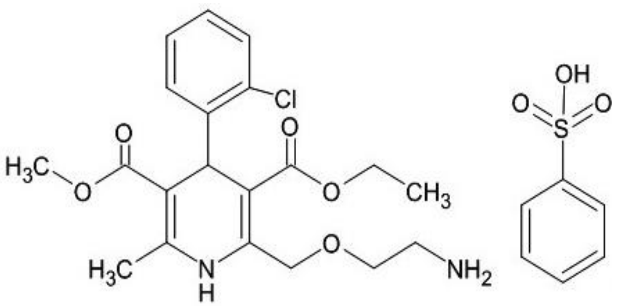


Figure 1: Structure of Amlodipine Besylate

Table 1: Linearity data of AMD

Concen. (% w/w)	Absorbance	Concen. (% w/w)	Absorbance
0.1	0.00321	2	0.09021
0.2	0.00625	3	0.12451
0.3	0.009581	4	0.17301
0.4	0.01254	5	0.21472
0.5	0.01569	6	0.26132
0.6	0.01987	7	0.32140
0.7	0.021987	8	0.36545
0.8	0.024891	9	0.41202
0.9	0.029561	10	0.45293
1	0.03324		

Table 2: Linearity, LOD and LOQ results of Amlodipine besylate

Parameters	Amlodipine Besylate
Linearity	0.1-10 % w/w
	-0.00727
	0.001
	0.032
Correlation coefficient (R^2)	0.9986
LOD	0.0468 % w/w
LOQ	0.1419 % w/w

Preparation of Calibration Curve

A calibration curve was derived to determine the linear relationship between standard drug absorbance and its concentration.

Accurately weighed 100 mg AMD was mixed with a specified quantity of previously dried and finely powdered IR pure potassium bromide to obtain the concentration in the range of 0.01 to 100.0% w/w. Absorbance corresponding to the optimised wavenumber ~1676 cm^{-1} was used for the absorbance measurements. The absorbance of all the AMD standards was measured in the replicates of six. The calibration curve was plotted with all the standard absorbance against concentration to get the linearity and regression equation. The linearity data of AMD and the calibration curve were shown in Table 1 and Figure 4, respectively.

Validation of the ATR-FTIR Spectroscopic Method

Validation of the proposed ATR-FTIR spectroscopic method was performed in accordance with the ICH Q2 (R1) guideline. Specificity, accuracy (% recovery), precision (reproducibility), linearity, range, limit of detection (LOD), and limit of quantification (LOQ) are the parameters subjected to validation[13].

Specificity

Specificity studies in ATR–FTIR spectroscopy were performed to assess the ability to uniquely identify AMD in the presence of formulation excipients and analyze its unique spectral "fingerprint." The selected C=O ester wavenumber of 1676 cm^{-1} was checked for specificity for AMD quantification and interference due to its tablet and microsphere excipients.

Linearity

The linearity evaluation involved preparing a standard sample of AMD within a concentration range of 0.01%–100% w/w.

Peak absorbance of the drug at the specified concentrations was measured to assess the linearity of the investigation. A calibration curve was generated by plotting the AMD concentration on the X axis and the peak absorbance on the Y axis (Figure 5).

The Limit of Detection (LOD) and Limit of Quantification (LOQ)

The limit of detection (LOD) refers to the lowest detectable concentration of an analyte in a sample, while the limit of quantification (LOQ) is the lowest quantifiable concentration of an analyte in a sample. Experimental verification and calculation of both LOD and LOQ were carried out using a specific equation.

$$LOD = 3.3\frac{SD}{Slope}$$

$$LOD = 10\frac{SD}{Slope}$$

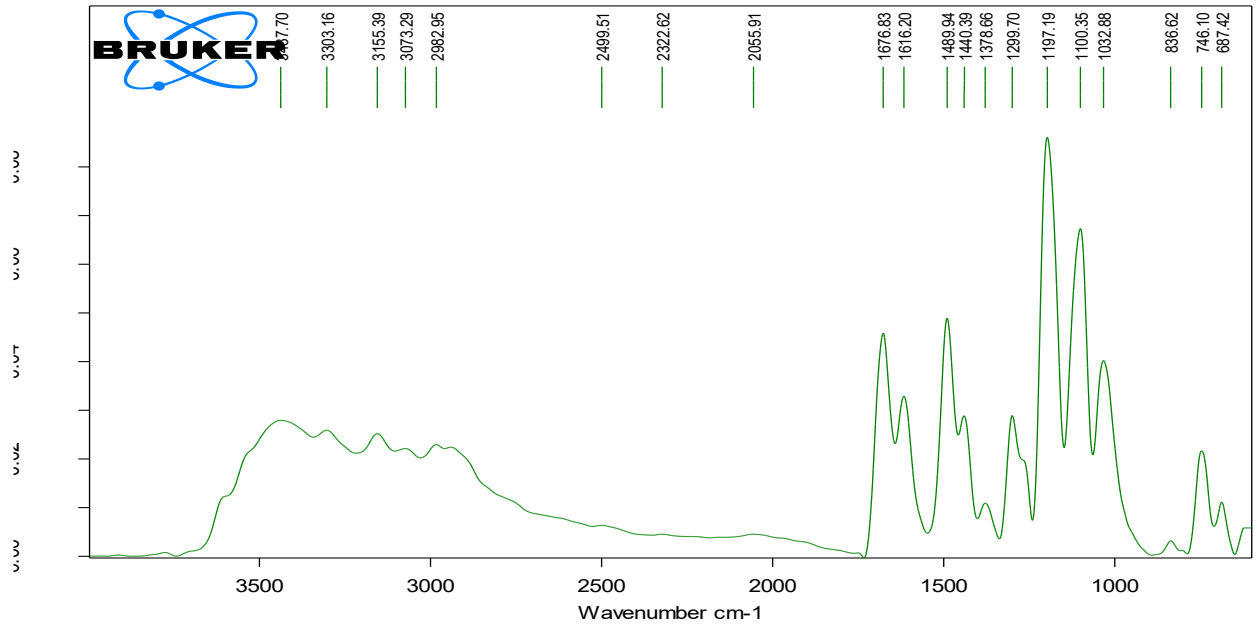


Figure 2: ATR -FTIR absorbance spectra of Amlodipine Besylate API

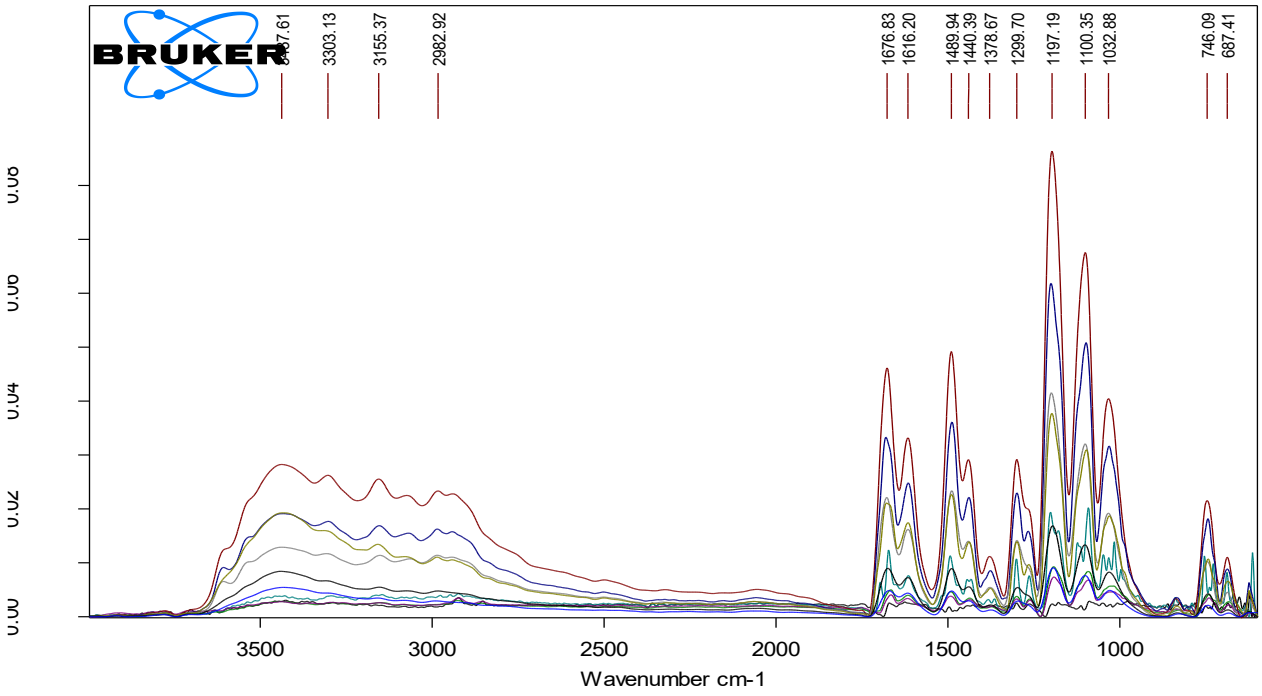


Figure 3: Overlain absorbance ATR-FTIR spectrum of Amlodipine Besylate

Table 3: Recovery study results of Amlodipine Besylate formulations

S. No.	Formulation	% Level	Pre-analysed conc. (mg)	Standard conc. added (mg)	Total amount (mg)	Amount of drug recovered (mg)	Mean % recovery	%RSD
1.	AMDMS	50%	10	5.0	14.98	4.98	99.60	0.0057
		80%	10	8.0	17.96	7.96	99.50	0.0100
		120%	10	12.0	22.01	12.01	100.08	0.0152
2.	AMDT1	50%	5.0	2.5	7.5	2.48	99.20	0.002
	(5.0 mg)	80%	5.0	4.0	9.0	3.96	99.00	0.052
		120%	5.0	6.0	11.0	5.96	99.33	0.054
3.	AMDT2	50%	5.0	2.5	7.5	2.46	98.40	0.043
	(5.0 mg)	80%	5.0	4.0	9.0	3.98	99.50	0.0021
		120%	5.0	6.0	11.0	5.98	99.66	0.007

Table 4: Results obtained from Precision studies

	Sample	Estimated Amount	Assay %	STD	% RSD
Intraday precision	AMD API	5.01 mg	100.20	± 0.0016	0.0015
	AMDMS	4.98 mg	99.96	± 0.020	0.020
	AMDT1	5.03 mg	100.70	± 0.015	0.014
	AMDT2	4.97 mg	99.49	± 0.025	0.0251
Interday precision	AMD API	5.00 mg	100.00	± 0.005	0.005
	AMDMS	4.97 mg	99.40	± 0.026	0.02615
	AMDT1	5.02 mg	100.40	± 0.010	0.0099
	AMDT2	4.98 mg	99.80	± 0.0115	0.0115

Accuracy Studies

Recovery studies were carried out to assess the accuracy and precision and to evaluate the role of interferences from the excipients incorporated in the AMD tablet and microsphere formulations.

The recovery studies were conducted by adding a known quantity of AMD API to its pre-analyzed tablet and microsphere powder at three concentration levels. The scheme for the level of concentration was assigned as 50%, 80%, and 120%, and then analyzed using the proposed method.

Precision Studies

Precision studies were conducted to determine the reproducibility of results. The precision of the method was checked by repeated scanning and measurement of the absorbance of the infrared band at ~1676 cm^{-1} (n = 6). Intraday and interday precision studies are conducted to evaluate the consistency of the method. Intraday precision gauges the consistency of results when testing a sample several times in a single day, whereas interday precision evaluates consistency across days. The reliability and repeatability of the measurement were expressed in terms of relative standard deviation (RSD).

Analysis of AMD Microspheres and Tablets

AMD Sample Preparation

Ground ten selected AMD tablet formations and AMDMS into a very fine powder. It was followed by measuring the required quantity of known tablet powder and microsphere quantity equivalent to the drug's concentration and recording their spectra under the same experimental conditions as the AMD standard.

AMD Sample Preparation and Analysis of AMD Tablets and Microspheres

The analysis of AMD tablets and microspheres was carried out using six samples which were analyzed in six replicates. The sample absorbance of the AMD at 1676 cm^{-1} was compared with the standard absorbance using the calibration curve parameters. The concentration of active drug is calculated by interpolation method. The determined concentration was subjected to statistical analysis to predict the reliability of the method. The relative standard deviation is determined for each determination and it is considered to be acceptable if it falls below 2%. The validated analytical method can be carried forward for the routine analysis of AMD loaded in tablets and polymer based microspheres routine for the quantitative analysis.

Table 5: Analysis results of AMD formulations

S. No.	Formulation	Label claim	% Assay*	± SD	%RSD
1.	AMDMS	5.0mg	99.96	± 0.001	0.1
2.	AMDT1	5.0mg	100.69	± 0.002	0.2
3	AMDT2	5.0mg	99.49	± 0.001	0.1

RESULTS

Green chemistry concepts are fully compatible with the suggested ATR-FTIR spectroscopy technique. In other words, the AMD content can be assessed without the need for any kind of solvents.

Spectral Characteristics and Optimisation of AMD Absorbance Wavenumber

The AMD API when scanned in the ATR FTIR spectrophotometer showed characteristic functional group stretching peaks. Figure 2 displays the AMD ATR- FTIR absorbance spectrum. The spectra illustrates that the hydroxide OH groups are responsible for the band at 3302 cm^{-1}, whilst the N-H groups are responsible for the band at 3474 cm^{-1}.

The stretching of the C=O group is associated with the band detected at 1676 cm^{-1}. The absorbance spectra with the unique C=O carboxylate group with 1676 cm^{-1} was selected for absorbance measurement of AMD standard and samples.

Validation

Specificity

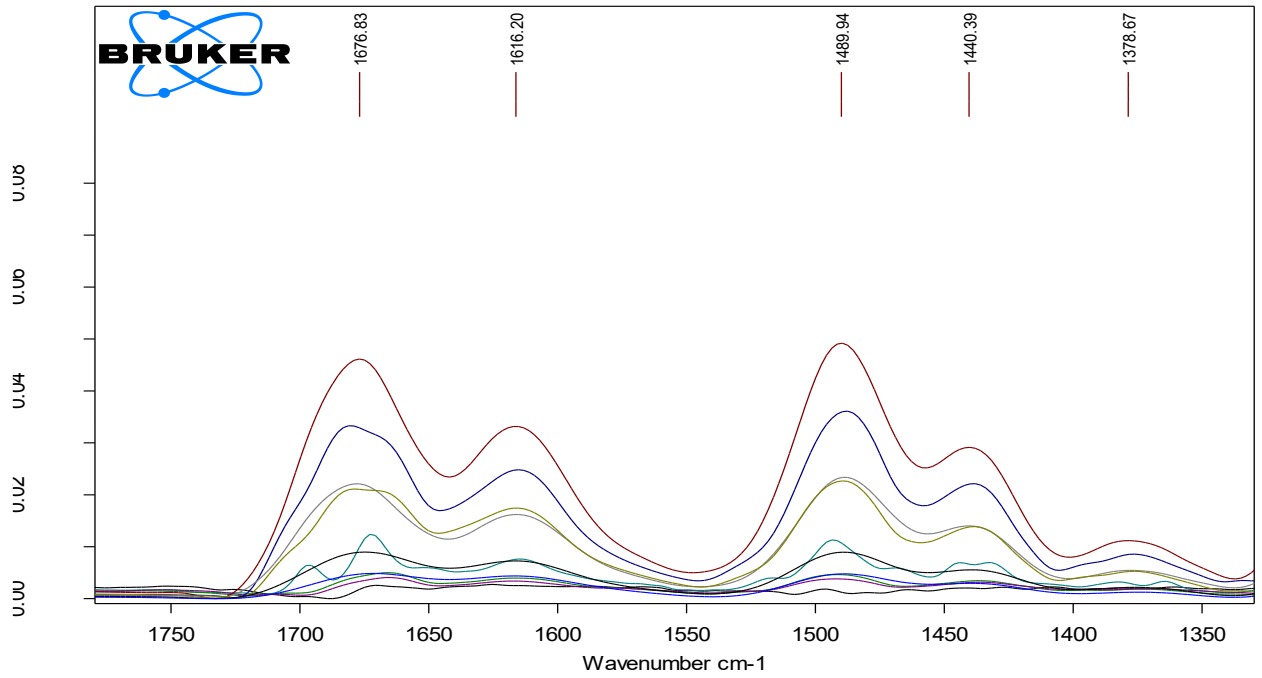
The overlain ATR-FTIR spectra of AMD pure drug, microspheres, and its table are depicted in Figure 4. The spectrum shows no interfering peak of diluent and possible formulation excipient with AMD. Since no interfering peaks were observed during the analysis, the method was specific.

Linearity

The calibration plot of absorbance versus concentration was linear over the range of 0.1% and 10 % w/w. Table 1 presents the linearity findings for the selected optimized concentration range. The linearity was found to be in that order, accompanied by a correlation coefficient (R^2) of 0.9986 (Figure 5). The linearity curve derived was utilized to estimate the amount of drug in the formulations.

Limit of Detection (LOD) and Limit of Quantification (LOQ)

The standard deviation of the response (σ) and slope of the calibration curve (S) were determined by using the calibration curve. The LOD was found to be 0.0468% w/w, and the LOQ was found to be 0.1419% w/w. Results are reported in Table 2. These results indicated the high



Overlain spectra Amlodipine Besylate at 1676 cm^{-1}

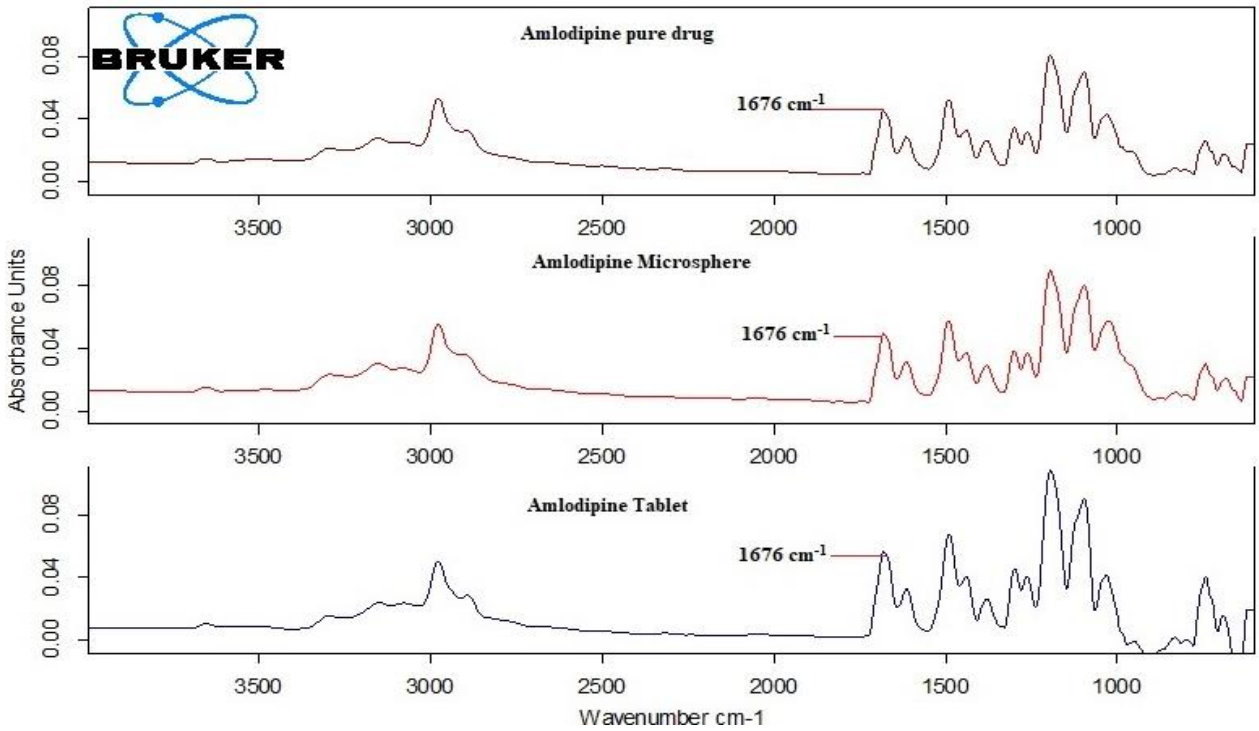


Figure 4: Overlain spectra Amlodipine Besylate pure drug, microspheres and its tablet depicting C=O ester at 1676 cm^{-1}

sensitivity of the proposed ATR-FTIR spectroscopy method.

Accuracy Studies

The accuracy studies indicate a high level of precision for the ATR-FTIR spectroscopic method (Table 3). The percentage recovery was in the range of 98.40 to 99% w/w. The assay and % RSD (n=3) for microspheres and their commercial tablets are displayed in Table 3. The % recovery was well within the acceptable range, and the % RSD was below 2.

Precision Studies

The precision of the method was expressed as the percentage of the relative standard deviation (%RSD). Repeatability was checked with multiple measurements of the AMD spectrum over three days of three distinct types of AMD samples (AMD, AMDMS, and AMDT1&T2) at a concentration of 5 mg, with subsequent calculation of the % RSD. These intra-day and inter-day experiments were performed under the same experimental conditions, in the same laboratory, for the same time period. As shown in table 4, the % RSD values for the intra-day precision studies are in the range of 0.0015-0.0251%, and the inter-day precision was between 0.0050-0.0261 and 1.05-1.92%. Both the intra-day and inter-day precision results were within the accepted limits, in accordance with ICH guidelines.

Analysis of Amlodipine Microspheres and Tablets

The proposed ATR-FTIR technique was used on AMD loaded microspheres and its individual doses of AMD tablets, and the FTIR absorbance spectrums were captured for pure Amlodipine, AMDMS and both AMDIPINE (AMDT1) and NORVASC (AMDT2) marketed tablet dosage forms.

Assay of AMD Formulations

The developed and validated ATR-FTIR spectroscopic method was utilised to estimate the quantity of AMD present in two different marketed tablets and its lab made polymer based microspheres. The results are summarized in table 5. The endurance of the peak around 1676 cm^{-1} in the AMD FTIR spectrum of the marketed tablets and microsphere confirmed that there was no significant interference due to the presence of excepients. The % assay was calculated from standard calibration curve. The assay results for the AMD microspheres (AMDMS) and its tablets formulations (AMDT1 and AMDT2) was 100.69 ± 0.002, 99.49 ± 0.001 and, 99.96 ± 0.001 % w/w, respectively. It presented good agreement within the labeled content (Table 2).

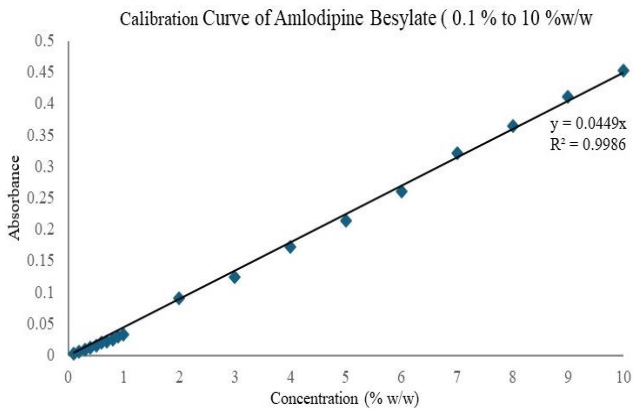


Figure 5: Calibration curve of Amlodipine Besylate

DISCUSSION

The wavenumber of 1676 cm^{-1} of amlodipine besylate was obtained near to the reported range. So, drug analysis was carried out at the optimized wavenumber for the selected C=O carbonyl functional group, which is uncommon in comparison with the polymer-based dosage forms and their tablet excipients. The methods reported earlier were solvent-based destructive analytical methods. Unlike the HPLC method reported earlier, which often requires extensive sample dissolution and extraction, ATR-FTIR needs little to no sample preparation, saving significant time. The assay estimation range was between 99 and 100%, which was equivalent to the reported methods. In ATR-FTIR, a single spectral measurement replaces the hours-long separation and analysis that occurs in HPLC, significantly cutting down on total time. The developed method was linear with the correlation coefficient R^2 near to 1. The accuracy and precision data further supports the accuracy of the proposed method. The specificity, linearity, LOD, LOQ, accuracy, and precision were found to be equal to the ICH guidelines. The method was found to be ideal for polymer-based and tablet formulations that requires less sample preparation steps. The current method stands unique for its nondestructive and nonsolvent analytical screening.

CONCLUSION

In this current study, we examined the potential to quantify amlodipine in a single dosage form using ATR-FTIR spectroscopy. Our findings demonstrate that FT-IR can accurately determine AMD in the mentioned formulations. ATR-FTIR excels over traditional analytical methods in speed, cost-effectiveness, and eco-friendliness by offering real-time, non-destructive measurements with minimal sample preparation and reduced waste. Its ability to perform in situ analysis, even in water, eliminates the need for extractive sampling and complex procedures common in other techniques. This leads to faster data acquisition, lower operating costs, and a smaller environmental footprint, making it a highly sustainable and efficient analytical tool. The FT-IR method we proposed is uncomplicated, quick, consistent, and rapid compared to other quantification techniques retrieved from the existing references. The future scope of our research will be to extend the method for the simultaneous analysis of AMD in combination with other drugs. Extending the technique for the simultaneous study of AMD in combination with different medications is the future focus of our research.

ABBREVIATIONS

AMD: Amlodipine; AMDMS: Amlodipine Microsphere; API: Active Pharmaceutical Ingredient; ATR; Attenuated Total Reflectance; FTIR : Fourier Transform Infra-Red: UV: Ultraviolet; cm^{-1}: Per Centimeter: Milligram; mg; SD: Standard Deviation; RSD: Relative Standard Deviation; LOD: Limit of detection; LOQ: Limit of Quantitation; ICH:

International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use.

Acknowledgements
Authors are thankful to the management of Vinayaka Mission's College of Pharmacy, Vinayaka Mission's Research Foundation (Deemed to be University), Salem for the continuous support and by providing the required facilities to complete this research work successfully

REFERENCES

1. K Swaroopa Rani, Swapna, A, Padma, A, Chaithanya, K, Ramalingam, P. Hari Hara Teja D. Res J Pharm Biol Chem Sci. 2011; 2: 470-479.
2. R Karajgi Santosh, Kulkarni Raghavendra, V. Universal. J Pharmacol. 2013; 1: 92-95.
3. Mehul Kumar P, Ramesh V, Vinay Kumar V, Srinivas R, Prakash V Diwan. Asian J Res Chem. 2009; 2: 127-130.
4. Vijaya Vichare, Tambe, Vrushali, Kashikar, Vrushali, Dhole, SN. Int J Chem Res. 2011; 1: 7-10.
5. Vivek Deshmukh, Chanekar, Pradeep D. Int J Pharm Pharm Sci. 2010; 3: 71-74.
6. L Szabó, Chiş, V, Pirnău, A, Leopold, N, Cozar, O, Orosz, Sz. Spectroscopic and Theoretical Study of Amlodipine Besylate. J Mol Struct. 2009; 924-926: 385-392.
7. Richa Sah. Saahil Arora. J Adv Pharm Edu Res. 2012; 3: 93-100.
8. Pournima S Patil, More, Harinath N, Pishwikar, Sachin A. Int J Pharm Pharm Sci. 2011; 3: 146-149.
9. Aysegül Gölcü, Yücesoy, Cem. KSU J Sci Eng. 2006; 2: 52-54.
10. Mahmoud A Omar, Abdelmageed, Osama H, Abdelgaber, Ahmed A, Saleh, Safaa F. Int Res J. Pure Appl Chem. 2013; 2: 133-146.
11. Aniruddha R Chabukswar, Jagdale, Swati C, Kumbhar, SV, Kadam, Vinayak J, Patil, Vinit D, Kuchkar, Bhanudas S, Lokhande, Pradeep D. Arch Apll. Sci Res. 2010; 3: 94-100.
12. JS Patil, Kamlapur, MV, Marapur, SC, Kadam, DV. Ionotropic Gelation and Polyelectrolyte Complexation: The Novel Technique to Design Hydrogel Particulate Sustained, Modulated Drug Delivery System: A Review. Dig J Nanomater Biostructures. 2010; 1: 241-248.
13. The International Conference on Harmonization. Validation of Analytical Procedure: Text and Methodology. 2005;Q2:R.