



Determination of Chemical Stability of the diabetes drug, Metformin Using UV-VIS and FT-IR Methods

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تحديد الثبات الكيميائي لدواء الميتفورمين المستخدم لعلاج داء السكري باستخدام طريقتي الأشعة فوق
البنفسجية والمرئية ومطيافية تحويل فورييه بالأشعة تحت الحمراء

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Received: June 15, 2026

Accepted: August 21, 2026

Published: September 17, 2026

Abstract:

Metformin hydrochloride is taken as a medicine for diabetes. Its chemical form needs to stay stable while it sits in storage. If it changes, the final tablet may lose quality. So, this work checked how stable metformin was after short heat periods. Two lab tools were used. These were ultraviolet-visible spectroscopy and Fourier-transform infrared spectroscopy. For the test, twenty Metformin tablets were ground into powder. Then a weighed amount of 110.92 mg was mixed with deionized water. The mixture was brought up to 100 mL to form a stock solution at a nominal strength of 1000 ppm. After that, a 100-ppm working standard was made for the spectral runs. UV-Visible data were collected at the maximum analytical wavelength of 233 nm for concentrations from 0 to 60 ppm. Absorbance rose as concentration increased. This made it possible to build a Beer-Lambert type calibration line. The same 233 nm wavelength was identified for the unheated sample and for the sample exposed to 70°C. For the heat stress, the solid tablets were placed at 30°C for 30 days, at 50°C for 14 days, and at 70°C for 24 hours. Samples were then prepared and checked. FT-IR spectra were also recorded. The goal was to compare the main functional group peaks from the unheated material with the peaks from the heat-treated samples. By combining the UV-Visible concentration check with the FT-IR structural scan, the study could track changes linked to concentration and also look for shifts in the molecular vibration pattern. Overall, the results give a workable stability check for metformin tablets under controlled intermittent heating. It also supports the idea of using paired spectroscopic methods in routine pharmaceutical quality testing.

Keywords: Metformin hydrochloride, chemical stability, UV-Visible spectroscopy, FT-IR spectroscopy, Beer-Lambert law, thermal exposure, pharmaceutical analysis.

الملخص

هدفت هذه الدراسة إلى تقييم الاستقرار الكيميائي لأقراص ميتفورمين هيدروكلوريد (metformin hydrochloride) تحت ظروف التعرض الحراري المتقطع، وذلك باستخدام تقنيتي مطيافية الأشعة فوق البنفسجية والمرئية (UV-Vis) ومطيافية الأشعة تحت الحمراء بتحويل فورييه (FTIR). تم تحضير محاليل قياسية ضمن نطاق تركيز يتراوح بين 0 و 60 جزءاً في المليون (ppm)، وسُجّلت القياسات عند طول موجي قدره 233 نانومتر، وهو الطول الموجي الذي يُظهر أقصى امتصاصية لكل من العينة غير المعرضة للحرارة والعينة التي عُرضت لدرجة حرارة 70 درجة مئوية. وأظهرت معايرة (UV-Vis) وجود علاقة خطية بين التركيز والامتصاصية، وصفت بالمعادلة: $A = 0.0403C + 0.0614$ (حيث $R^2 = 0.9973$)؛ كما قُدرت حدود الكشف وحدود التقدير الكمي بـ 4.09 جزء في المليون و 12.39 جزء في المليون على التوالي. وأشارت المقارنة الحرارية إلى أن التقييم الكمي للعينة المعرضة لدرجة حرارة 70 درجة مئوية اعتمد على مقارنة امتصاصية التركيزات المتماثلة (تحديداً 10 و 20 و 30 و 40 جزءاً في المليون) عند الطول الموجي 233 نانومتر. وكشفت أطيف (FTIR) عن بقاء الحزم المميزة للميتفورمين -وإن لوحظ انخفاض أكبر في النفاذية عند درجة حرارة 70 درجة مئوية- دون وجود أي دليل على حدوث تغيير كيميائي جوهري في ظل ظروف التعرض المطبقة.

الكلمات المفتاحية: ميتفورمين هيدروكلوريد، الاستقرار الكيميائي، مطيافية UV-Vis، مطيافية FTIR، قانون بير-لامبرت، التعرض الحراري.

1. Introduction

Metformin hydrochloride is a biguanide that can be taken by mouth and is widely used to treat type 2 diabetes mellitus. The pharmaceutical dosage forms are important for quality control, starting from manufacture, during storage and distribution, and at the time of patient use, given the extensive usage. The active substance(s) should remain unchanged in terms of identity and assay until the end of the shelf life as stated on the label. Heat, moisture, light, interaction with excipients or any alteration caused by these could alter the reliability of the dosage form with the potential to create impurities or degradation products. The methods for metformin bulk and finished tablet determination hence are focuses of current analytically research less or more selective (Pandian et al., 2025; Kurdi et al., 2024).

Metformin hydrochloride has the molecular formula $C_4H_{11}N_5 \cdot HCl$ and a biguanide structural formula that can be represented as $H_2N-C(=NH)-NH-C(=NH)-N(CH_3)_2 \cdot HCl$. This structure contains the nitrogen-rich biguanide group that is relevant to the characteristic N-H, C=N, and C-N bands examined by FT-IR spectroscopy.

ThiERA 101 is one of the theses that may affect the stability of drugs. A medicine is likely to come into contact with high temperature during some parts of its practical transportation, storage near a heating or cooling source or not maintaining the proper temperature for storage. This is exposure as opposed to continual heating as the material is reheating and cooling continually. For this reason, reading the tablet samples after intermittent thermal treatment may be more helpful to learn about their ability to withstand an actual temperature change. In the case of pharmaceutical preparations containing metformin, stability assessment has taken on great importance, as evidenced by several studies employed stability indicating methods for the detection of degradation products and serve as methods for identification of the active substance and its related impurities in metformin tablets (Marie et al., 2025; Demirović et al., 2025).

In case an analyte absorbs the UV-Vis region it is a convenient method to reach the quantitative determination of a pharmaceutical compound. The approach is based upon Beer-Lambert's law

stating a linear ($< c$) relationship between absorbance and concentration. Standard solutions of known concentrations are drawn up to make a calibration curve and an unknown sample is then estimated using the regression equation. Spectrophotometric methods are found to be acceptable parameters for the analysis of drug metformin, due to the fact that they are desirable for drug analysis, such as those used in routine pharmaceutical laboratories, with respect to preparing standard solutions, using minimum volumes of solvents, and speed of the analysis (Alqarni et al., 2024; El-Masry et al., 2026).

The Beer-Lambert relationship is given by:

$$A = \epsilon bC$$

Here, A is absorbance, ϵ is molar absorptivity, b is the path length of the cell in cm and C is the concentration. The relationship is often shown using the linear regression equation during a calibration.

$$A = mC + b$$

This equation has the form $y = mx + b$, where m is the slope of the “calibration line” and b is the “intercept. The coefficient of determination, R^2 , is used to determine the quality of the fit of the experimental calibration points to the linear calibration model. The closer the value is to 1.000 the closer the linearity in the investigated concentration range. After successful validation, recently good linear response and use in pharmaceutical dosages have been reported using this method (El-Abassy et al., 2024; Attimarad et al., 2021).

UV-Visible analysis can provide information that will suggest a change in the amount of detectable metformin concentration in the solution but cannot independently determine if the structure and/or functional group of the molecule has changed during the exposure period. FT-IR spectroscopy is a complementary technique for the UV method as it is based on the measurement of vibrations of bonds. Typical peaks in the IR spectrum of metformin hydrochloride correspond to nitrogen-hydrogen (N-H) stretching, C=N vibrations, C-N vibrations and other absorptions in the fingerprint region. Spectral shifts (change in wavenumber), changes in transmittance in the bands, loss or gain of a band, or an overall shift may be observed before and after exposure. Thus, observations can be a sign of chemical changes, either to the product and its excipients, or of a potential thermal modification in the product itself (Elnadi et al., 2022; Rajeshwari et al., 2022).

When the two types of evidence, UV-Visible spectroscopy and FT-IR, are used in combination they give two complementary pieces. UV-Visible measurement provides a quantitative result based on the calibration equation and absorbance values while FT-IR examination provides qualitative results and functional groups identification and comparison. A combined design is particularly appropriate for metformin tablets in order to allow the concentration response and molecular spectral behavior of a compound to be assessed independently, without having to use one analysis method. Similar analytical studies show that spectroscopic techniques can be used for the determination of metformin, monitoring of impurities, and evaluation of drug formulation (Castro et al., 2023; Moussa et al., 2021).

Consequently, in this research the chemical stability of metformin hydrochloride tablets under intermittent heating at 30°C, 50°C and 70°C was determined. The study will determine the relationship between UV-Visible measurements and the quantity to be measured, evaluate whether the Beer-Lambert law is followed, use the UV-Visible measurements to determine the regression parameters from the available set of calibration measurements and compare the spectra of FT-IR of untreated sample with the heat-exposed sample. The work will also include presenting the equations for the limit of detection and the limit of quantification, and calculating these only as many times as necessary based upon the number of experimental replicates or the blank response that is available.

2. Materials and Methods

2.1 Materials and sample identification

The tested product was the metformal film coated tablet-containing 500mg of metformin hydrochloride. The commercial carton states that it contains the product Metformal which is produced by Berlin-Chemie AG, Berlin, Germany. Dose form was chosen because the standard tablets contain multiple combinations of recessed tablet features and the active ingredient needs to be chemically identifiable following environmental exposure; hence multiple variable storage conditions are likely to be encountered prior to tablet administration. The study was not a pure reference standard and thus any effects due to the formulation matrix could be taken into account when interpreting the spectra (Pandian et al., 2025; Kurdi et al., 2024).

Twenty tablets were mixed together and crushed to get a uniform powder. 110.92 mg was transferred to a 100mL volumetric flask, dissolved in deionized water, and, made up to the volume with deionized water. In the lab record, this was prepared as the 1000 ppm stock solution. The stock solution was then diluted to make a working solution of 100 ppm, which was used for the UV-Visible measurement of the sample of metformin. The lower the content of the drug in the tablet, the more probable that the obtained weighed quantity will be from tablet powder, not a certified pure standard, and if extracted, the result should be reported as a percentage drug content and the average mass of the tablet should be taken into account when calculating the final assay (Alqarni et al., 2024; Sha'at et al., 2022).



Figure 1: Commercial package of the Metformal tablet sample used in the study.

The picture records the tested medicinal product's commercial identification. Each film-coated tablet contains 500 mg of metformin hydrochloride, according to the label.

2.2 Exposure to intermittent heat – design considerations

The samples were exposed to intermittent, not continuous, heat. These exposure periods were spread throughout the day for about 2–3 hours and then returned to ambient conditions. This design is selected to exhibit handling and storage temperature changes and is not representative of continual heating for the entire listed time. These were heated with the tablet being in the solid state, and the heated tablets were then ground to powder and measured by either the UV-Visible or FT-IR.

Table 1: Intermittent thermal exposure schedule for Metformal tablet samples.

Sample code	Exposure temperature	Exposure duration	Exposure pattern	Analytical techniques
Control	Room temperature	No intentional heat treatment	Untreated reference material	UV-Visible and FT-IR

Sample 30	30°C	30 days	Intermittent daily exposure, approximately 2–3 h per day	FT-IR
Sample 50	50°C	14 days	Intermittent daily exposure	FT-IR
Sample 70	70°C	24 h	Intermittent exposure over the stated period	FT-IR and UV-Visible

The table shows the difference between the reference sample (no exposure) and the three temperature-exposed samples. The reported durations refer to the "total" experimental duration, and not the duration of thermal exposure throughout the day.

This temperature study design allows one to compare the features in the spectra as the temperature of the material being exposed is raised from 30°C (plus or minus a few degrees) to 70°C. It also serves an excellent basis for selection of whether absorbance or FT-IR band shift is related to treatment temperature. Stress testing and/or analytical separation have also been studies to detect changes in drug response and/or related drug degradation products to ensure formulations are stable (Marie et al., 2025; Demirović et al., 2025).

2.3 UV-Visible calibration procedure

Standard solutions at concentrations of 0, 10, 20, 30, 40, 50 and 60 ppm were used to prepare the calibration data. Deionized water was used as the blank. Absorbance was measured at 233 nm, the maximum analytical wavelength identified for metformin in both the unheated sample and the 70°C-exposed sample. The same instrument settings and wavelength were used for all UV measurements to provide a consistent comparison of the concentration responses.

A calibration graph was made showing absorbance against the concentration in ppm. The slope, intercept and coefficient of determination were obtained by linear regression. A calibration equation was given as:

$$A = mC + b$$

The absorbance A that is measured when the current concentration of metformin in ppm is used in the regression equation. The current concentration of metformin in ppm used in the regression equation is the absorbance A that is measured. The validity of Beer-Lambert law within the chosen range was verified by checking the linear trend in this relationship. Typical UV methods with metabolites turn mostly to calibration curve and regression statistics to show the proportionality between the concentration of an analyte and its detector response (Attimarad et al., 2021; El-Masry et al., 2026).

2.4 Calculation of LOD and LOQ

The limit of detection and limit of quantification were tested by the standard calibration method. The following equations were used:

$$LOD = \frac{3.3\sigma}{m}$$

$$LOQ = \frac{10\sigma}{m}$$

We used the slope of the calibration curve, m , and the standard deviation of the analytical response, σ . A best estimate of σ is made using repeated blank measurements, or replicate measurements at low concentration. When just one calibration response is found at each concentration, the LOD and LOQ may be estimated by the residual standard deviation for the regression, but they should be cited as estimated LOD and LOQ, not as independent experimental LOD and LOQ. This differentiation is vital to avoid exaggerated method sensitivity (El-Abassy et al., 2024; Mohamed et al., 2026).

2.5 FT-IR spectral measurement

The thermally exposed samples were characterized by FT-IR spectra of the samples taken with a range from 4000 to 650 cm^{-1} . For samples 30°C, 50°C and 70°C, the following 8 sample scans, 8 background scans, and a spectral resolution of 8 cm^{-1} are reported, along with acceptable system status and Happ-Genzel apodization. The three reports display the instrument software, which is Agilent MicroLab. The exported treated Metformal spectrum from the SW version 7.5.18 of Bruker OPUS was used as the available treated Metformal reference spectrum for comparison.

For FT-IR analysis, the band positions and intensities of the N-H stretching regions, C=N stretching/out-of-plane bending regions, as well as the fingerprint region frequencies of the N-H and C-X bonds of the metformin hydrochloride have been taken into consideration. The untreated material was compared to each material exposed. Change in the position of a peak or the transmittance does not necessarily mean chemical degradation but could mean a change that needs to be interpreted based on UV-Visible and considering the overall pattern of the spectrum (Elnadi et al., 2022; Rajeshwari et al., 2022).

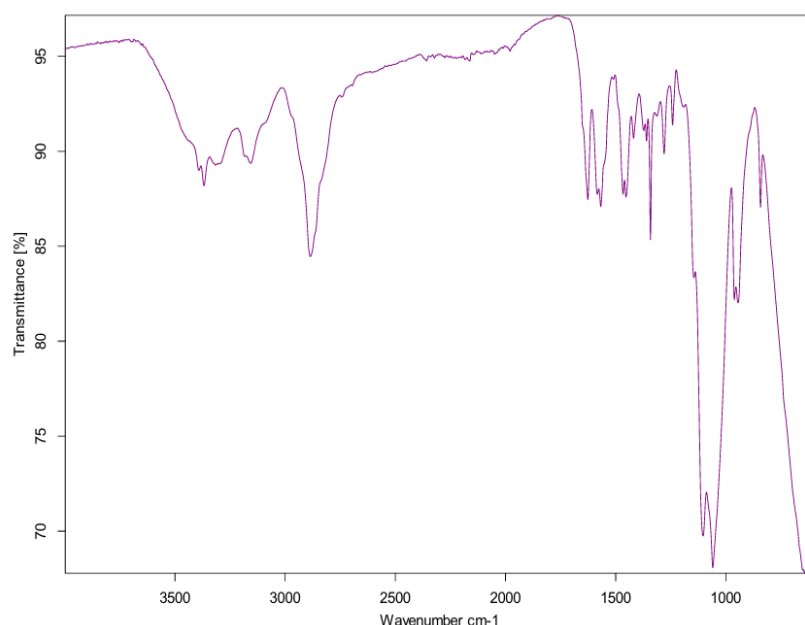


Figure 2:FT-IR spectrum of the untreated Metformal reference material.

The spectrum represents the Metformal material's accessible reference FT-IR record before to comparison with samples that have been subjected to heat. It is employed to determine the metformin tablet formulation's general spectral pattern.

3. Results and Discussion

3.1 UV-Visible calibration curve and Beer-Lambert relationship

The UV-Visible results rose as the metformin level went from 0 to 60 ppm. Absorbance increased in a smooth way across that set of points. The numbers match a direct link between concentration and response for the range that was tested. This pattern fits the Beer-Lambert law. It also shows that the chosen range worked well for making a quantitative calibration curve for metformin.

Similar straight-line UV-Vis trends have been described before for metformin, including in tablet samples and in mixed antidiabetic formulations (Alqarni et al., 2024; Attimarad et al., 2021; El-Abassy et al., 2024).

Table 2: UV-Visible calibration data for metformin standard solutions.

Concentration, ppm	Absorbance
0	0.000
10	0.480
20	0.897
30	1.293
40	1.739
50	2.050
60	2.434

Source: Prepared based on the results compiled by the researcher.

Absorbance rose from 0.000 at 0 ppm up to 2.434 at 60 ppm. These readings were taken as is, then used for the regression setup and for the reported performance metrics.

A straight line was fit to the calibration set. The fit gave this relation.

$$A = 0.0403C + 0.0614$$

Here, A means absorbance. C is the metformin level in ppm. The fit had $R^2 = 0.9973$. In other words, about 99.73% of the absorbance changes matched what the concentration would predict over the tested range. This points to clear linear behavior. It also supports using the calibration line to estimate metformin in diluted unknown samples.

The intercept is slightly above zero. That can happen for several practical reasons, such as instrument drift in the baseline, cuvette-to-cuvette differences, solvent response, or small mix-up during sample prep and reading. (El-Masry et al., 2026; Kurdi et al., 2024; Mohamed et al., 2026)

Table 3:Regression and sensitivity results for the UV-Visible calibration curve.

Parameter	Result
Linear range	0–60 ppm
Regression equation	$A = 0.0403C + 0.0614$
Slope	0.0403 absorbance unit ppm ⁻¹
Intercept	0.0614 absorbance unit
Coefficient of determination, R^2	0.9973
Residual standard deviation, σ	0.0499
Estimated LOD	4.09 ppm
Estimated LOQ	12.39 ppm

Source: Calculated from the calibration data.

High R^2 value is indicative of the suitability of the data for Beer-Lambert calibration within the chosen concentration range. Due to the fact that the chosen file includes one response for each concentration and no blank readings for independent replicates, the calculated settings (sensitivity) should be considered as estimated LOD and LOQ.

The residual standard deviation of the regression was used to estimate the detection limit at 4.09 ppm and the estimated quantification limit at the value of 12.39 ppm. The LOQ is contained within the calibration range, so it is suitable for making quantitative measurements of compounds at concentrations higher than the LOQ. But several blank and low concentration replicate readings should be taken in a different experiment for a complete method validation process to report final values for validation of LOD and LOQ. This method is in agreement with recent analytical validation for metformin, in which replicates of experimental responses are used for support of linearity and sensitivity, instead of just a single one for each concentration (Ahmad & Deore, 2025; Sha'at et al., 2022; Sattar et al., 2026).

The absorbance value at the 60ppm concentration was 2.434, which is relatively high for a UV-Visible absorbance measurement. To obtain further dilutions for future routine analysis it is useful to have measurements within a lower range of absorbance and to minimise any potential for deviation from linearity at high absorbance. However, the present data showed a good linear relationship throughout the entire range tested. This calibration model can therefore be used in this work as the quantitative reference of the prepared solutions of Metformin (Alqarni et al., 2024; El-Abassy et al., 2024; Kurdi et al., 2024).

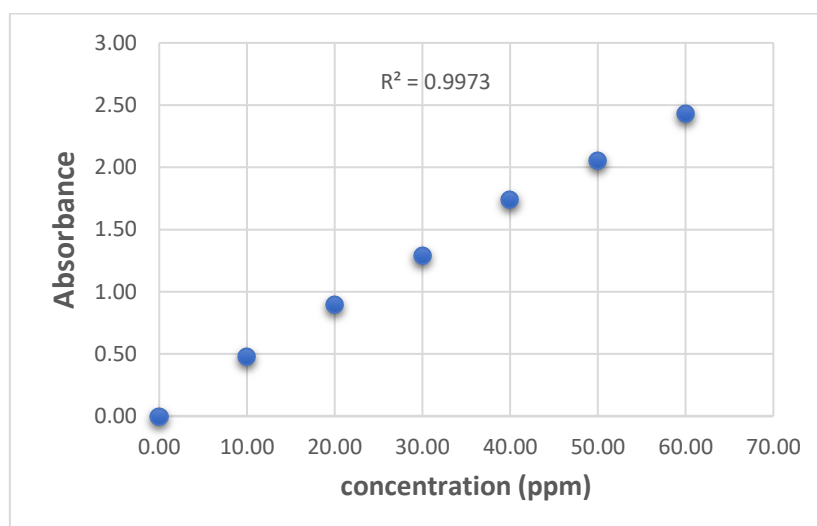


Figure 3: UV-Visible calibration curve for metformin standard solutions.

The concentrations of 0–60 ppm and the related absorbance measurements are used to create the figure. The Beer-Lambert connection, which is utilized in quantitative analysis, is illustrated by the fitted regression line.

3.2 FT-IR spectral comparison after intermittent thermal exposure

FT-IR measurements were performed with 30°C, 50°C and 70°C-exposure samples and were performed in the range of 4000–650 cm^{-1} . Eight scans and eight background scans at 8 cm^{-1} resolution are reported in each scan. Comparisons were made on the characteristic regions corresponding to the metformin structure such as N-H stretching in the range 3400-3000 cm^{-1} , C=N vibrational region near 1625 cm^{-1} and C-N vibrational region in fingerprint region. FT-IR can be useful here to check if the absorption peak changes significantly in wavelength, new absorption peak emerges, or an expected absorption peak has been lost in the presence of increasing thermal exposure (Elnadi et al., 2022; Marin et al., 2026; Rajeshwari et al., 2022).

The positions of the spectra were not significantly affected by the three treatment temperatures and the main absorption regions corresponding to a tablet sample of metformin were retained in the spectra. The peak lists available for the test did not report any new dominating peaks. This indicates that after periods of heating up to 70°C, the dominant molecular vibration mode was maintained. Several of the bands, however, exhibited a decrease in percentage transmittance, and small wavenumber shifts as the temperature increased. The interpretation of these changes must be carried out with great care, as these may also be influenced by the physical state of the tablet matrix, sample thickness, powder packing and contact pressure. Thus, the results of observation are consistent with the variation in the spectra due to thermal treatment, but not to extensive chemical degradation (Castro et al., 2023; Elnadi et al., 2022; Rajeshwari et al., 2022).

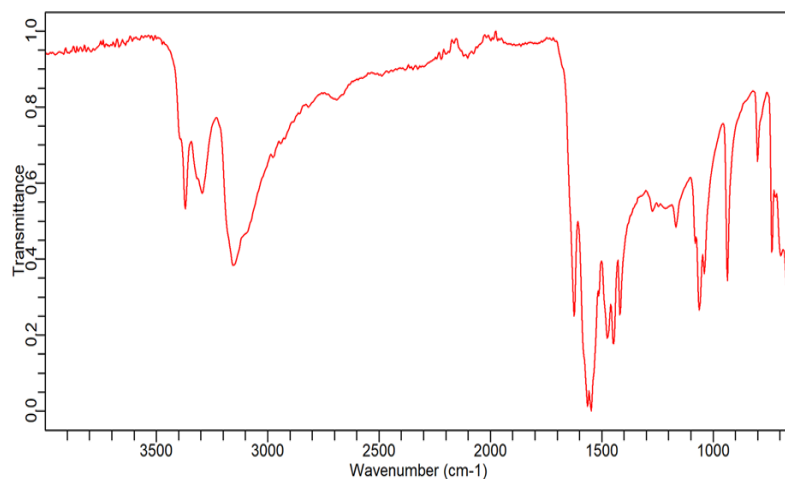


Figure 4:FT-IR spectrum of the Metformin tablet sample exposed intermittently to 30°C.

The FT-IR spectra and peak table for the sample exposed at 30°C are displayed in the figure. For spectrum comparison, it stands for the lowest temperature treatment condition.

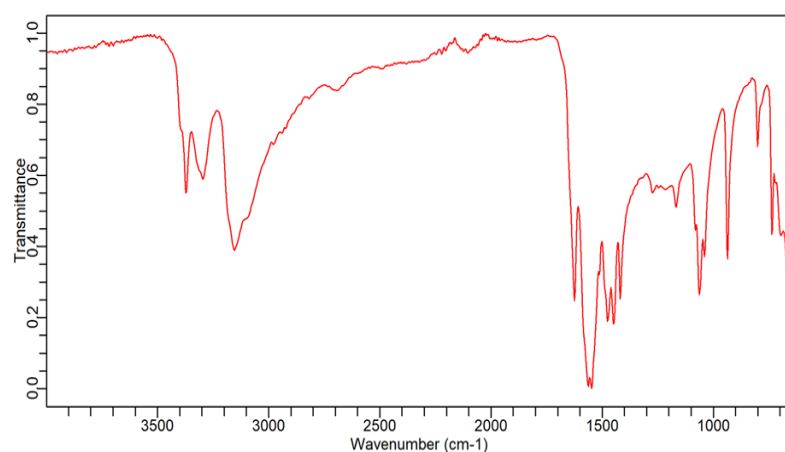


Figure 5:FT-IR spectrum of the Metformin tablet sample exposed intermittently to 50°C.

The spectral record for the sample subjected to intermittent daily heating at 50°C for 14 days is shown in the figure. Peak intensity and location can be compared with samples at 30°C and 70°C.

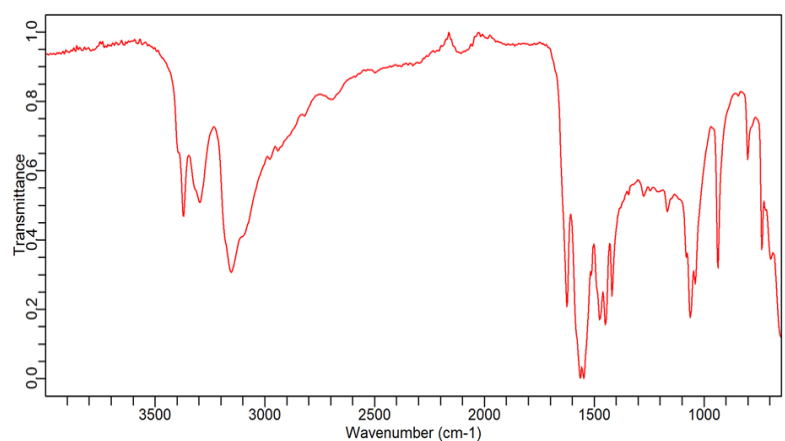


Figure 6:FT-IR spectrum of the Metformin tablet sample exposed intermittently to 70°C.

The spectrum of the treatment sample with the greatest temperature is shown in the picture. The numerical comparison shown in Table 4 is visually consistent with the decrease in transmittance at many bands.

Table 4: Selected FT-IR peaks of metformin tablet samples after intermittent heat exposure.

Approximate band assignment	30°C, cm ⁻¹ / transmittance %	50°C, cm ⁻¹ / transmittance %	70°C, cm ⁻¹ / transmittance %
Fingerprint-region vibration	704.47 / 79.01	704.47 / 72.94	697.01 / 58.22
C-N-related fingerprint vibration	1036.20 / 76.74	1036.20 / 69.94	1036.20 / 55.56
C-N-related fingerprint vibration	1062.29 / 72.59	1062.29 / 63.82	1062.29 / 47.87
C-N and N-H deformation region	1416.39 / 72.39	1420.12 / 63.26	1420.12 / 51.67
N-H deformation and C=N-related region	1550.57 / 63.64	1546.85 / 51.78	1546.85 / 37.08
C=N-related vibration	1625.12 / 72.04	1625.12 / 63.06	1625.12 / 49.80
N-H stretching region	3294.97 / 83.47	3294.97 / 78.60	3294.97 / 68.25
N-H stretching region	3369.51 / 82.03	3373.24 / 76.81	3369.51 / 65.79

Source: Prepared from FT-IR reports

The above peaks show the same key spectral peaks were present in all samples. The most distinct numerical application presents a gradual decrease of percentage transmittance as the treatment temperature increased from 30°C to 70°C.

The band at approximately 1625 cm⁻¹ remained present in all samples, with a recorded position of 1625.12 cm⁻¹ at 30°C and 50°C and 1625.12 cm⁻¹ at 70°C. The band did not change in position, although its intensity decreased from 72.04% at 30°C to 49.80% at 70°C, which indicates that the structures of C-N bond environment of metformin have been preserved and their intermolecular or tablet powder had undergone changes after the heat treatment. It can be also observed from the previous spectroscopic studies, that the nitrogen containing vibrations are still detectable in metformin containing systems regardless of the alterations in the chemical environment resulting from formulation, complexation, or in analytical purposes (El-Masry et al., 2026; Elnadi et al., 2022; Rajeshwari et al., 2022).

This was also observed in the fingerprint region. The peak at 1062.29 cm⁻¹ in the 30°C sample decreased from 72.59% transmittance to 63.82% at 50°C and 47.87% at 70°C. The transmittance of the peak in wavenumbers decreased from 76.74% to 55.56% but the wavenumber remained constant at 1036.20 cm⁻¹ in all three samples. The results of retention of peak position within a decrease of transmittance suggest the existence of a vibration at this position, while the increase of heat exposure could have resulted in some effect on the local physical or chemical environment. These are changes that can be seen in the spectrum as a reaction to the therapy, not as a proven degradation, unless they are backed up by a validated assay, impurity profile, or structural determination of a degradation product (Demirović et al., 2025; Emam & Emam, 2023; Poncarová et al., 2025).

In the region of N-H stretching, the peak did not change even for all three samples and remained at 3294.97 cm⁻¹. Its transmittance declined from 83.47% in sample 30°C to 78.60% in sample 50°C and 68.25% in sample 70°C. The band near 3370 cm⁻¹ showed a small displacement from 3369.51 cm⁻¹ at 30°C to 3373.24 cm⁻¹ at 50°C, then returned to 3369.51 cm⁻¹ at 70°C. This is not a loss of the N-H functionality, but it could suggest a change in hydrogen bonding or association between the molecules in the heated tablet material. This is an area of great interest,

and rich in groups containing nitrogen that allow the monitoring of changes in the characterization of the vibrational profile (Marin et al., 2026; Rajeshwari et al., 2022; Sattar et al., 2026).

3.3 Overall view of thermal stability

The FT-IR data suggest that short, stop-and-go heating at 30°C, 50°C, and 70°C did not remove the main absorption peaks linked to metformin in the tablet samples. Peaks still appear in the N-H stretching area, the C=N area, and the fingerprint zone. This means the spectral pattern for the metformin-containing material was still there. Even so, the changes in transmittance with temperature show that the 70°C condition caused the biggest shift at several wavelengths compared with the other tested settings.

The UV-Visible calibration data confirm a linear metformin response over the 0–60 ppm range at 233 nm. The maximum wavelength was the same for the unheated sample and the 70°C-exposed sample, so the thermal treatment did not shift the selected analytical wavelength. A quantitative assessment of the effect of 70°C should therefore compare the absorbance readings of matched metformin concentrations, particularly 10, 20, 30, and 40 ppm, measured at 233 nm under the same conditions. These paired readings can be entered into the calibration equation to compare the concentration response before and after thermal exposure. The FT-IR findings remain suitable for a relative comparison of spectral response between the exposure conditions (Alqarni et al., 2024; El-Abassy et al., 2024; Kurdi et al., 2024).

3.4 Comparison of Spectral Response under the Exposure Conditions

The comparison of the FT-IR peak list indicated that all the samples (30°C, 50°C, and 70°C) had the same broad spectral regions related to the metformin tablet formulation. Bands in the N-H stretching region, the C=N related region and the fingerprint region were still observed in all three samples. This means that no major identifiable spectral features of the material containing metformin were eliminated by exposure to heat in intermittent fashion.

The most obvious difference between samples was a gradual decrease in transmittance due to an increase in the exposure temperature. For example, the peak at 1062.29 cm⁻¹ decreased from 72.59% transmittance at 30°C to 63.82% at 50°C and 47.87% at 70°C. Similarly, the band near 1625 cm⁻¹ decreased from 72.04% at 30°C to 63.06% at 50°C and 49.80% at 70°C. The transmittance of this peak decreased from 83.47% at 30°C to 68.25% at 70°C, maintaining its position in all samples.

The transmittance of the 70°C sample decreased the most. This trend suggests that for maximum thermal condition the maximum change in the FT-IR spectral response was observed. This observation does not necessarily indicate full chemical degradation however, as the main bands remained and intensity of the FT-IR is dependent of packing of the powder, sample thickness, measurement pressure, interaction with tablet excipients or metformin.

A few features—particularly those associated with strong bands—did not shift significantly from sample to sample. The band corresponding to the C=N was kept at about 1625 cm⁻¹ of the three samples and the band corresponding to the 1036.20 cm⁻¹ was preserved in the three samples. This helps maintain a preferred molecular pattern of metformin following a temporary mild temperature. In some areas, minor changes were observed, which could be due to modification of the local molecular environment, or changes in hydrogen bonding in the solid-acting ingredient matrix of the tablet, rather than to a complete change in the active ingredient. It is observed that the spectral pattern matches with the usage of FT-IR, as a qualitative method for monitoring change in formulation, retention of functionalities, and possible stress-related variation. If the degradation pathway is confirmed then the peak or peaks in the degradation product chromatogram have to be chromatographically separated from the parent peak or peaks and structurally identified, and new (dominant) bands should not appear in the chromatogram for any form of degradation (Elnadi et al., 2022; Marie et al., 2025; Demirović et al., 2025).

4. Discussion

The calibration of the UV-Visible showed a good linear relationship between the concentrations of metformin and the absorbance range tested was between 0–60 ppm. Based on the values of the regression equation ($A = 0.0403C + 0.0614$) and coefficient of determination ($R^2=0.9973$), it can be seen that the prepared standard solutions obeyed the Beer-Lambert relationship within the concentration range investigated. This shows appropriate dilution of tablet samples and confirmed that a calibration curve was appropriate for the measurement of metformin concentration following the dilution.

The estimated LOD and LOQ values obtained was 4.09 ppm and 12.39 ppm, respectively. These values are signs that the method is able to detect metformin in the selected calibration range at low concentrations. The values were calculated for an absorbance of one which was recorded at each concentration of data but no replicate blank data were available, so the values were determined from the residual standard deviation of the regression. For these reasons, the estimated values of their sensitivity should be reported as such until confirmed by independent replicate measurements. This interpretation is in accordance with the validation procedures of the metformin analytical methods carried out in the recent years where the linearity, sensitivity and precision are be evaluated based on repeated experimental responses (Ahmad & Deore, 2025; Alqarni et al., 2024; El-Abassy et al., 2024).

FT-IR data provided complimentary qualitative evidence on the effect of intermittently heat exposed metformin tablet samples. The major spectral regions were still visible at 30°C, 50°C and 70°C after the treatments. The N-H stretching band at 3295-3400 cm^{-1} , the C=N stretching band at 1625 cm^{-1} and some bands in the fingerprint region were left in all the samples. The broadness of these peaks suggests that the broad pattern of the general molecular spectrum would still exist under the heat conditions applied to the material containing metformin.

This transmittance decrease due to temperature was most noticeable for sample 70°C. For example, the transmittance of the band near 1062 cm^{-1} decreased from 72.59% at 30°C to 47.87% at 70°C. The band near 1625 cm^{-1} decreased from 72.04% to 49.80%, while the N-H stretching band near 3295 cm^{-1} decreased from 83.47% to 68.25%. From these changes, it is seen that the most noticeable change in the observed spectral response was observed with intermittent exposures to 70°C.

This reduced transmittance could occur due to changes in hydrogen bonding and/or molecular association, drug-excipient interaction, or physical properties of the tablet powder upon heating. But the transmittance values obtained by FT-IR also depend on the sample thickness, packing density of the powder sample and pressure applied to the sample when being measured. Thus, it may be thought of as representing spectral variation, but not necessarily a chemical degradation, and changes in the observations must, therefore, be treated as such. It has been demonstrated with similar analysis that the alterations in the spectral intensity of metformin necessitate the confirmation of phenomenon via particular quantitative method prior to determination of degradation mechanism (Castro et al., 2023; El-Masry et al., 2026; Rajeshwari et al., 2022).

In consequence of no presence of a new dominant absorption band the major bands related to the presence of metformin in the tablets indicate that the tested tablet material is not so thermolabile when exposed to the intermittent exposure conditions. However, the sample at 70°C should also be characterized further as it showed the highest drop in transmittance over a number of spectral regions. If the stability indicating chromatographic procedure is available, then it will separate the metformin from the possible degradation products and impurities and it will give better indications of how many times and for how long the product was exposed to temperature (Kurdi et al., 2024; Pawar et al., 2024; Shah & Kotadiya, 2025).

5. Study Limitations

The calibration dataset contains one recorded absorbance value at each concentration. Accordingly, the LOD and LOQ reported in this work were estimated from the residual standard deviation of the regression equation and are presented as estimated sensitivity values for the available calibration dataset.

The UV-Visible analysis identified 233 nm as the common maximum wavelength for the unheated sample and the 70°C-exposed sample. Accordingly, any temperature-related UV comparison should be based on the absorbance values for equivalent metformin concentrations at 233 nm rather than on a change in wavelength. Matched measurements at 10, 20, 30, and 40 ppm provide the appropriate basis for comparing the concentration response of the two conditions.

FT-IR spectroscopy provides qualitative information through the retention, displacement, or intensity variation of functional-group bands. Changes in transmittance should be interpreted together with the spectral pattern because they can also reflect powder packing, sample thickness, tablet excipients, moisture content, or measurement pressure.

The thermal exposure schedule was intermittent, with approximately 2–3 hours of heating per day. The reported findings therefore describe the stated intermittent exposure conditions.

6. Conclusion

In this study, periodic severe heating of 30°C, 50°C and 70°C for a duration of 21 days on Metformin hydrochloride tablets was evaluated for chemical stability using UV-Visible and FTIR spectroscopy. The calibration curve was made from the data obtained for 0 to 60 ppm of UV-visible and had a good linear range with regression equation ($A = 0.0403C + 0.0614$) and correlation coefficient, ($R^2 = 0.9973$). The Limit of Detection (LOD) and the Limit of Quantification (LOQ) respectively was estimated at 4.09 ppm and 12.39 ppm.

The major bands related to metformin were unaltered in the FT-IR spectra following each of the exposure. The 70°C experienced sample had the least transmittance, especially at the N-H and C=N related regions and the fingerprint region. The general character of the spectrum was the same, and no new dominant absorption band could be found, except for the more prominent positions of the main peaks. The results have shown that a measurable spectral variation was observed as a result of repeated heat exposure, particularly at 70°C, but no extensive chemical alteration of the metformin was confirmed in the samples of tablets tested.

7. Recommendations

- Measure the unheated and 70°C-exposed metformin solutions at the common maximum wavelength of 233 nm for matched concentrations, including 10, 20, 30, and 40 ppm. Compare the paired absorbance values and apply the calibration equation to quantify any temperature-related difference in concentration response.
- Record the replicate FT-IR spectra of the various samples, and compare them with a digitized spectrum of certified metformin hydrochloride standard.
- An HPLC method for separation of metformin and separation/identification of any thermally produced degradation products should be used in future work that involves separating the drug from impurities by this method.
- Bring humidity under control and document more accurately the daily amount of thermal exposure in future stability testing experiments.

Compliance with ethical standards

Disclosure of conflict of interest

The authors declare that they have no conflict of interest.

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