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UV Spectroscopy in the Analysis of Metformin: A Review of Simultaneous Estimation Technique

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ABSTRACT
This review explores advancements in UV spectroscopy for the simultaneous estimation of metformin in pharmaceutical formulations. UV spectroscopy, known for its simplicity, cost-effectiveness, and precision, is crucial in quality control and bioavailability studies of metformin. The article discusses various analytical techniques, including calibration curve methods, derivative spectroscopy, and chemometric approaches, highlighting their applications and benefits. Recent technological innovations and research trends are examined to provide a comprehensive understanding of current methodologies. By addressing challenges and future directions, this review aims to enhance the analytical capabilities of UV spectroscopy in pharmaceutical analysis, ensuring accurate and efficient estimation of metformin in complex drug formulations.

KEYWORDS: *metformin, uv spectroscopy, method development, solubility enhancement, simultaneous estimation*

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INTRODUCTION
Nowadays, most people agree that the best oral treatment for reducing blood glucose levels in people with type 2 diabetes is metformin. Clinical trials were prompted by the rediscovery of its efficacy in the 1940s while antidiabetic medicines were being evaluated. Metformin primarily acts by reducing the amount of glucose produced by the liver, increasing insulin sensitivity in peripheral tissues, and promoting glucose absorption and utilization. This is why it was first thought to be less effective than other glucose-lowering biguanides and was not used extensively. Metformin can cut HbA1c by about 1% to 2% and is beneficial in lowering blood glucose levels, according to studies.Metformin side effects that are most commonly mentioned include gastrointestinal problems like nausea, diarrhea, and abdominal pain. There is a new tablet formulation of a fixed-dose combination that contains clomid on the market (1). A wavelength of 233 nm and spectra were selected to create the calibration curve. Six independent assessments were averaged to establish the computed values, which were then validated by checks on factors including quantification accuracy, specificity, and system fit. As part of the method development process, a precision sample was also created .(2)It makes a substantial difference in poorly soluble medication solubility. The concentration of a solute in a saturated solution at a specific temperature is one way to quantify solubility. Another way to define it is qualitatively. A crucial factor in the development of oral medications with poor water solubility is solubility enhancement.(3) The study of the interactions between matter and electromagnetic radiation is known as spectroscopy. These interactions, in which molecules receive or emit energy in definite amounts known as quanta,(4) are important because The two-stage least squares method is a simple method for calculating structural equations. The two stages of this procedure are as follows: the first estimates the moment matrix of the reduced form, which takes disturbances into account; the second estimates the coefficients of a single

structural equation, which is regarded as "purified."There are two steps in the two-stage least squares approach for structural equation estimation. The reduced form's moment matrix, which takes disturbances into account, is estimated in the first stage. Estimating a single structural equation's coefficients is the second stage. This approach offers comprehensive information features up to the point where the structural disturbance's moment matrix is non-diagonal.

Metformin:
The historic usage of Galega officinalis as medicine in medieval Europe is where metformin first appeared. Culpeper's Complete Herbal states that G. officinalis was thought to be useful against a number of illnesses, such as fever, worms, epilepsy, and the plague. Due to this historical use, galegine and later the more effective synthalin were shown to be useful in the treatment of diabetes. Eventually, Hesse, Taubmann, Slotta, and Tschesche's animal research revealed that metformin and other biguanides lower blood glucose levels. In several nations, metformin and phenformin, a similar medication, were extensively used to treat diabetes. However, because of the possibility of serious adverse effects, especially lactic acidosis, their usage has been restricted in most places.(1) In several nations, metformin and phenformin, a similar medication, were extensively used to treat diabetes. However, because of the possibility of serious side effects, especially lactic acidotic disease, their usage has been prohibited in most places.

Metformin has a major impact on cellular metabolism because it builds up in mitochondria at concentrations up to 1000 times greater than in the extracellular environment. Because of the membrane potential that exists between the inner mitochondrial membrane and the plasma membrane, metformin can enter the mitochondria because of its positive charge. Metformin decreases the generation of ATP in Complex I of the respiratory chain, which is one of its main effects. ADP/ATP ratios are changed as a result of this inhibition, especially following Metformin treatment, albeit these effects are not limited to one level. Certain transporters aid in the uptake of metformin into hepatocytes, which causes the medication to build up inside the cells and concentrate even more in the mitochondria because of variations in membrane potential(5). Consequently, ATP production We investigated the connection between prostate cancer in Denmark and the diabetes drug metformin. It was discovered that whereas other oral antidiabetic drugs did not lower the chance of prostate cancer detection, metformin did(16). For the most of this time, metformin has been prescribed for pregnancy; nevertheless, usage has been restricted both geographically and in terms of quantity(15)

For the treatment of type-2 diabetes, a recently released fixed-dose tablet containing metformin hydrochloride (MET) has been made available. This long-term illness is caused by the body's decreased sensitivity to the hormone insulin, which is crucial for regulating the absorption of sugar in the body. (2)

UV spectroscopy:
The study of the interactions between light and electromagnetic radiation and matter is known as spectroscopy. The energy absorbed or emitted in discrete amounts known as quanta indicates this interaction. From radio waves to gamma rays, these absorption and emission processes reveal important details regarding the characteristics of the materials under study throughout the electromagnetic spectrum.

UV spectroscopy is an analytical technique that utilizes light with wavelengths ranging from 200 to 800 nm, which falls within the ultraviolet (UV) or visible spectrum. Due to the technique's versatility, colorless chemicals in the UV range (200-400 nm) and colored substances in the visible range (400-800 nm) can both be analyzed. In essence, UV spectroscopy counts the precise UV or visible light wavelengths that a sample absorbs or transmits in comparison to a reference or blank. This measurement can provide information on the components and their concentrations within the sample because it is directly related to the composition of the sample. Uv principle:The transition of electrons within a molecule or an ion from a lower to a higher energy level produces the UV absorption spectrum, while the opposite type of transition produces the UV emission spectrum. A molecule's or an ion's valence electrons can be stimulated or promoted by UV radiation to move from their ground state orbital to an excited state orbital, or anti bonding, at a higher energy level. orbital, which exhibits absorption upon detection (5).

Chromophores: The presence of specific chromophores allows many organic compounds to absorb ultraviolet and visible light. operational unit. Chromophores are the real groupings that absorb the radiation. Statistically speaking, some electronic transitions are.

Auxochromes: The color of a molecule can be enhanced by certain groups known as auxochromes. These groups typically do not absorb light strongly within the 200-800 nm range but can influence the absorption spectrum of the chromophore they are attached to.

Solvents: The absorption spectrum of a compound can change depending on its chemical structure when it is dissolved in a solvent. Typically, non-polar solvents and molecules have minimal impact on the absorption spectrum. In contrast, polar molecules can experience significant changes when interacting with a polar solvent. This interaction often results in broadening of the absorption bands and a decrease in both structural resolution and the maximum molar absorptivity (ε max).

Potentially: there are three ground state orbitals involved. They are: σ (bonding) molecular orbital, π (bonding) molecular orbital and n (non bonding) atomic orbital

Instrumentation:
There are two type of absorbance instruments to collect uv absorption spectra

- Single beam UV spectrophotometer
- Double beam UV spectrophotometer

A monochromator or filter is positioned between the light source and the sample in a single beam UV spectrophotometer. The device can measure one wavelength at a time with this configuration. UV Spectrophotometer with Dual Beam A double beam UV spectrophotometer, on the other hand, features a beam splitter and a number of mirrors in addition to a monochromator and light source that are similar. These elements aim the laser beam at the sample under analysis as well as at a reference sample. The measurements are more accurate in this configuration. In general, the double beam instrument is quicker and more effective than the single beam model.(4)

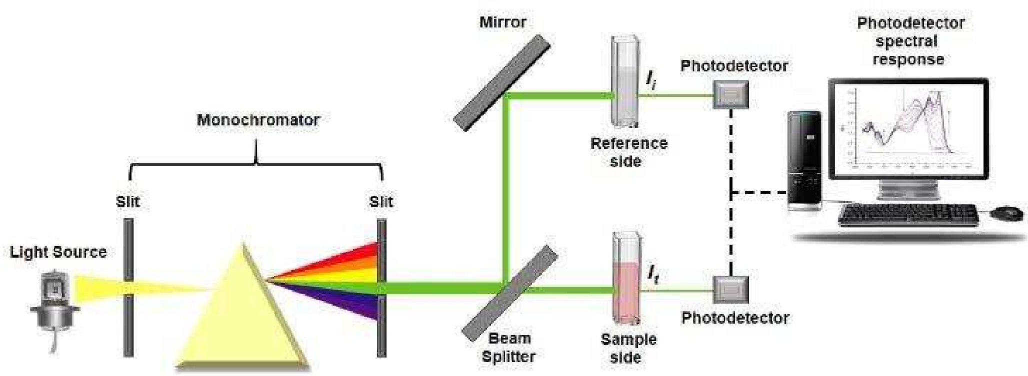


Figure: Double beam uv spectrophotometer

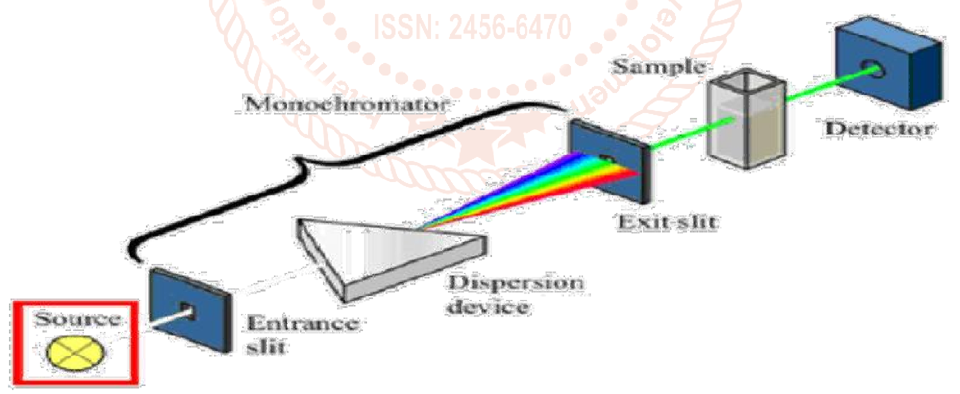


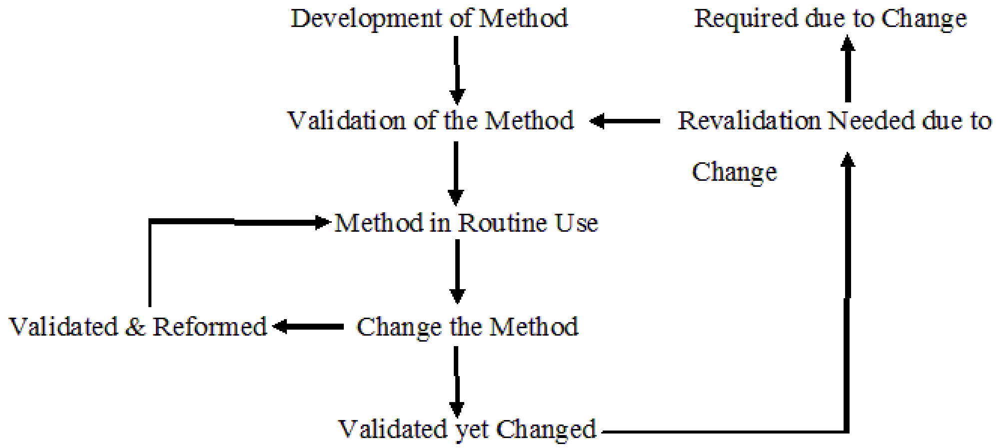
Figure: single beam uv spectrophotometer

For people with type 2 diabetes mellitus, vildagliptin and metformin hydrochloride are administered in addition to diet and exercise to improve blood sugar management. Patients who have not achieved adequate glycemic control with metformin hydrochloride or vildagliptin alone, or who are currently taking vildagliptin and metformin hydrochloride as separate tablets, are the target audience for this medication.(13)

For the treatment of type 2 diabetes, a new fixed-dose combination tablet containing metformin hydrochloride (MET) is now available. The hormone insulin, which controls how much sugar is absorbed by the body, is resistant to the body in this long-term metabolic illness. MET levels have been estimated using a variety of methods, such as capillary electrophoresis (CE) with UV detection and high-performance liquid chromatography (HPLC) with ultraviolet (UV) or fluorescence detection(2). UV spectrophotometric methods are mostly used for multi-component simultaneous analysis. This method shortens the time and expense of analysis by eliminating the requirement to separate interference-causing materials and allowing the simultaneous detection of many analytes (6).

Method development:

Novel approaches are developed to evaluate pharmacopoeia or non-pharmacopoeia items with the goal of maximizing strength and accuracy while reducing expense and time. Initial trials are used to refine and validate these procedures. Using comparative laboratory data, alternative methodologies are created and deployed to replace the current procedures, taking into account all relevant advantages and disadvantages(17).



Necessity of method development

Drug evaluation entails the identification, characterization, and resolution of medications in combination, including biological fluids and dosage forms. The primary objective of analytical methods used in drug development and manufacture is to offer information about the medicine's efficacy, which is directly related to the dosage that is needed. Additionally, it evaluates stability, which denotes degradation products, impurities, which pertain to the safety of the medicine, and bioavailability, which covers essential pharmacological features including crystal shape, uniformity, and drug release. It also assesses the effect of manufacturing parameters to guarantee reliable drug production.(17). Numerous analytical methods, including spectrophotometry, chromatography, and electrophoresis, can be employed to analyze various components. The use of UV spectrophotometric techniques for the simultaneous determination of pharmaceuticals is highlighted in this review(6).

Solubility:

The term "solubility" describes the degree to which a substance, known as the "solute," may dissolve in another substance, known as the "solvent," as well as the solute's and the solvent's respective solid, liquid, and gaseous states.(10)

Solubility, as defined by IUPAC, is the percentage of a certain solute in a particular solvent as shown by the composition of a saturated solution. Several units, including concentration, molality, mole fraction, mole ratio, and others, can be used to indicate this measurement. In the watery fluids of the gastrointestinal system, drugs that dissolve poorly in water typically have low rates of dissolution as well, which frequently results in insufficient absorption and decreased bioavailability. (10)When creating formulations for medications to be taken orally that have low water solubility, improving solubility is an important consideration to take into account.(3) Solubility can be increased by combining and applying several techniques. The drug's characteristics and the formulation's objectives influence the method selection. For example, addressing solubility difficulties with a mix of surfactants and particle size reduction may be more effective than using either technique alone.

Solubility Enhancement:

Drugs that do not dissolve well in water may not be adequately absorbed in the body's gastrointestinal fluids due to their low solubility and slow rate of dissolution. This matter is especially pertinent to class II medications, as increasing the drug's solubility and rate of dissolution in these liquids might improve its effectiveness and absorption. Poor solubility drugs may not dissolve quickly enough at the point of absorption, which could lead to their prolonged residence in the gastrointestinal tract and eventual disintegration in the stomach. A medication that is poorly soluble can be identified using two essential parameters: the dose-to-solubility ratio and an aqueous solubility of less than 100 µg/mL. A substance's solubility at a given temperature is determined by the concentration of dissolved solute in balance with its solid form. The molecular crystal lattice, the properties of water, and the giver and acceptor of hydrogen bonds all affect a molecule's solubility.

Method of solubility:

Surfactants:

Reducing the interfacial tension between the surfaces of the solute and solvent improves wetting and solvation interactions, which is a conventional strategy for making a poorly soluble material more soluble.

pH adjustments:

One of the easiest and most popular methods to increase the water solubility of ionizable substances is to modify the pH of the microenvironment to affect the ionization behavior. The pH of the medium and the drug's pKa have an impact on a compound's ionization, according to the Henderson-Hasselbalch equation and the pH-partition theory.

Salt formation:

For many years, one method of increasing solubility has been to turn poorly soluble medication candidates—such as weak acids and

The polar regions are known to harbor sea salt ions, namely Na+, Cl−, and Br−, in a variety of media, such as aerosols, snow packs, and ice cores (DeAngelis et al., 1997; Rankin and Wolff, 2003; Fischer et al., 2007; Legrand et al., 2016). Sea salts that get stuck in snowpacks produce a significant chemical reservoir and are essential to many different chemical processes(14).

bases—into salts. This method works effectively for both solid dose forms and injectable and other liquid formulations.

Cosolvents:

To dissolve lipophilic medications, a cosolvent system—a combination of suitable solvents—is frequently employed. Today, ethanol, propylene glycol, glycerin, and polyethylene glycol 400 (PEG 400) are a few examples of water-soluble organic solvents in use.

Polymeric Alteration:

A medicine can exist in a variety of crystalline forms, referred to as polymorphs, each possessing unique properties. The physicochemical characteristics of these polymorphs might differ, encompassing aspects such as inherent solubility, dissolving rate, shape, density, biological activity, shelf life, melting point, vapor pressure, physical and chemical stability, and bioavailability.

Co-evaporation/Co-precipitation:

Strongly basic medications, as prochlorperazine maleate, exhibit a considerable decrease in solubility in alkaline circumstances yet a high solubility in acidic situations. When a weak base is administered orally in a conventional formulation, the poorly soluble free base may precipitate in the intestinal fluid. Because of this precipitation, the drug's bioavailability is decreased because it cannot be released from the formulation.

Solvent Deposition/Evaporation:

This method involves dissolving the drug in a solvent such as methylene chloride to create a clear solution. The carrier is then mixed into the solution through stirring, and the solvent is removed by evaporation under controlled temperature and pressure conditions.(9)

Simultaneous estimation:

It is no longer necessary to prepare and analyze several samples independently when using simultaneous estimation, which allows the analysis of multiple substances in a single analytical procedure. When studying complicated samples with several analytes that need to be quantified at the same time, this technique is essential (8) There are other interdependencies between cost categories, and our empirical estimation seeks to demonstrate how various activities are linked to one another and do not happen sequentially or independently. Scrap costs are influenced by the intricacy of the product and process as well as the resources used for assessment and prevention(11). The techniques were found to be simple, quick, extremely sensitive, and economical, which qualified them for the simultaneous estimating (7). If certain requirements are satisfied, simultaneous equations (Vierordt's approach) may be used to quantify both medications (x and y) in a sample if they each absorb at the other's λ_max(6).

Method development metformin use the uv spectrophotometric:

In the development of that novel technique, UV spectrophotometry was used. One of the most often recommended oral drugs for managing type 2 diabetes is metformin. Ensuring the safety and efficacy of pharmaceutical products requires accurate measurement of the amount of metformin present. Because UV spectrophotometry is sensitive and inexpensive, it offers a simple and effective way to measure this.

Choose Your Wavelength:
Examine the metformin's UV spectra between 200 and 400 nm to determine its λ_max. Metformin typically has a λ_max of approximately 232 nm.

Preparing the Standard Solution:
Metformin hydrochloride can be standardized by dissolving a specific quantity of the medication in an appropriate solvent, like water or a buffered solution, to create a stock solution.

The calibration curve
To create a calibration curve, create a number of diluted solutions from the standard stock. To create the calibration curve, plot the absorbance against concentration after measuring each dilution's absorbance at the λ_max.

An Example of Analysis

Using the proper solvent, remove metformin from tablet formulations. Utilizing the calibration curve, determine the extracted solution's absorbance at λ_max and utilize it to determine the concentration (12).

Case study:
Development of a UV Spectrophotometric Method for Analyzing Metformin in Multi-Drug Combinations.

Sr.no	Drug	Solvent	Wavelength	Instrument
1	Vildagliptin and metformin	Distilled water	Vildagliptin 217 nm.metformin 234nm	UV
2	Metformin HCL	Methanol	Metformin HCL233nm	UV
3	Metformin hcl, pioglitazoneHCL, Glimepiride	Acetonitrile, methonal, glacial acetic acid	MET 235nm, PIO 265, GLP227nm	HPLC
4	Teneligliptin hydobromide hydrate, metformin HCL	Methanol	MET 237nm, TEN 246nm	UV
5	Glibenclamide	Methanol	GLB 229.5 nm	UV
6	Paracetamol, aceclofenac and thiocolchicoside	Phosphate buffer	PCM 243nm, ACE 274nm, THC 259NM	UV
7	Dapagliflozin, metformin	Methanol	Dapagliflozin 235nm, metformin 272nm	UV
8	Repaglinide, metformin	Methanol (AR)	REPA 284 nm, MET237 nm	UV

1st case Vildagliptin and metformin by uv spectrophotometery
Vildagliptin and metformin hydrochloride in combination tablet form can now be estimated simultaneously with ease using a simple, accurate, precise, and repeatable procedure. It was necessary to create new procedures for their simultaneous analysis because there are currently no UV methods available for the simultaneous estimation of these two medications in their combination dosage form.

Now, a simple, accurate, exact, and repeatable method may be used to estimate vildagliptin and metformin hydrochloride simultaneously in combination tablet form. Since there aren't any UV techniques available right now for estimating these two medications together in their combination dosage form, this development was required.

Experimental condition
Distilled water was chosen as the analytical solvent based on the drug's solubility properties. The UV spectra of both medications were scanned, and wavelengths of 217 nm15 for Vildagliptin and 234 nm for Metformin were chosen for estimate.

Wavelength selection

The λ max was ascertained by independently scanning the standard solution containing Vildagliptin and Metformin hydrochloride at varying concentrations throughout the 200–400 nm range. Also, the overlap spectrum for both medications was examined.

Method of validation

Recovery research was used to determine accuracy. The recovery experiment was conducted by adding different known concentrations of conventional metformin and vildagliptin to the tablet sample that had already been tested. Assay precision was based on repeatability, intraday accuracy for both medication

Accuracy

By comparing the analytical quantity found with the known amount spiked at the 80%, 100%, and 120% level of LOQ concentration with measurements for each concentration level reached, the procedure's accuracy was ascertained.

Limit of detection and quantitation

Using the following formula, the LOD and LOQ of cefixime trihydrate were calculated based on the slope of the calibration curve, the standard deviation of the response, and both.
LOD is equal to 3.3 x σ / S.
LOQ is equal to 10 times σ / S.

Where
σ is the response's standard deviation.
S is the calibration curve's slope.

Vildagliptin's LOD and LOQ were determined to be 0.023 µg / ml and 0.225 µg / ml, respectively, and 0.44 µg / Metformin 1.35 µg/ml and ml, respectively.

Precision

By measuring the absorbance of six replicates of 0.7 µg/ml of Vildagliptin and 7 µg/ml of Metformin, the repeatability, reproducibility, and intermediate precision of the procedure were used to estimate the precision, which was reported as a percentage RSD. Table 4's % RSD values are less than 2%, demonstrating the good accuracy of the analytical process.

Linearity

Both metformin and vildagliptin showed linearity, with absorbencies between 3.5 and 10.5 mcg ml-1 and 0.35 and 1.05 mcg ml-1, respectively. at the specific wavelengths they have chosen, 217 nm and 234 nm. correspondingly. The Metformin and Vildagliptin calibration graphs

Specificity

A specificity analysis was carried out to assess the possibility of interference between the medication ingredient and the tablet excipients. The commercial formulation's excipients were combined in the appropriate amounts, diluted with methanol, and filtered through Whatman filter paper No. 41. To ascertain whether the excipients and the medication were interfering, the UV spectra of the standard solution and the placebo were compared (7)

Stability of the solution

A specificity study was carried out to ascertain whether the medication ingredient and tablet excipients would interfere with one another. The commercial formulation's excipients were combined in the appropriate amounts, diluted with methanol, and filtered through Whatman filter paper No. 41. The UV spectra of the standard solution and the placebo were then contrasted in order to look for any possible drug-excipient interference. (7)

Results

The recently suggested technique for calculating the molar absorptivity of metformin (Figure 2) and vildagliptin (Figure 1) exhibits noteworthy results. The equations $y = 0.004x + 0.005$ y=0.004x+0.005 for Vildagliptin and $y = 0.006x + 0.012$ y=0.006x+0.012 for Metformin are the results of the linear regression analysis of absorbance against concentration. As Figure 4 illustrates, both of these equations have a high correlation coefficient ($\boldsymbol{U}$ 2 = 0.999 r 2 = 0.999). Table 1 displays the findings of the UV analysis. A high degree of precision is shown by the method's low standard deviation and percentage relative standard deviation (%RSD). Table 2 illustrates that the %RSD is below 2% and complies with ICH criteria.The recovery percentage ranged from 98 to 102%, demonstrating the high degree of specificity and accuracy of the suggested approach. Table 2 displays the recovery study's findings.(13)

Table no 1 Result of uv analysis

Parameters	Vildagliptin	Metformin
Detection wavelength	217nm	234nm
Beers las limit	0.17 µg / ml -1.56 µg / ml	1.19 µg / ml -10.73 µg / ml
Regression equation	y = mx + c	Y=mx +c
slope	0.004	0.006
Intercept	0.005	0.012
Correlation coefficient	0.999	0.999

Table no 2 Recovery

Drug	Level of recovery %	Amount present (in µg / ml)	Amount found (in µg / ml)	% recovery	% RSD
Vildagliptin	50%	0.35	0.3479	99.28	0.0966
	100%	0.7	0.7049	100.77	0.1321
	150%	1.05	1.0542	100.4	0.1443
Metformin	50%	3.5	3.498	99.94	0.1598
	100%	7	7.008	100	0.1072
	150%	10.5	10.507	100.06	0.1218

Table no 3 result of validation parameters

S. No	Validation parameter	Result vidaglipatin	Result metfoemin
1	Linarity	R 2 =0.999	R 2=0.999
2	Precision Interday precision Intermediate precision Intraday precision	%RSD 0.167 0.159 0.136	%RSD 0.147 0.155 0.132
3	Accuracy 50% 100% 150%	%recovery 99.28 100.77 100.4	%recovery 99.94 100 100.06
4	LOD	0.074 µg / ml	0.44µg / ml
5	LOQ	0.225µg / ml	1.35µg / ml
6	Beer lambert's limit	0.17µg / ml-1.56µg / ml	1.192µg / ml- 10.73µg / ml

Conclusion:

The advances in the simultaneous determination of metformin using UV spectroscopy have been emphasized in this study, with particular attention paid to method development and solubility enhancement techniques. A common anti-diabetic medication, metformin, frequently needs exact analytical techniques to guarantee correct dosage and effectiveness. Because UV spectroscopy is easy to use, affordable, and dependable when measuring metformin in different formulations, it has become an invaluable instrument.

Metformin's solubility has been significantly improved in recent research, which is important for increasing its bioavailability and therapeutic efficacy. To address the intrinsic solubility issues with metformin, a number of strategies have been investigated, including the use of solubilizing chemicals and innovative formulation techniques.

Metformin estimate method development has also changed, with new strategies seeking to boost

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