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REASEARCH ARTICLE ON RP-HPLC METHOD DEVELOPMENT AND VALIDATION OF TRABECTEDIN FOR THE ESTIMATION OF API AND PHARMACEUTICAL FORMULATION

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ABSTRACT
A reliable and efficient Reverse Phase High-Performance Liquid Chromatography (RP-HPLC) method was developed and validated for the quantitative determination of Trabectedin in active pharmaceutical ingredients (API) and pharmaceutical formulations. The method employed a [specify column type, e.g., C18] column with a mobile phase comprising [specify solvents, e.g., acetonitrile and buffer] in an optimized ratio. The detection wavelength was set at [specify wavelength], ensuring high sensitivity. Validation was performed in accordance with ICH guidelines, covering key parameters such as linearity, accuracy, precision, robustness, and specificity. The method demonstrated excellent linearity across the tested concentration range with a correlation coefficient (R^2) > [value]. Accuracy was confirmed through recovery studies, and precision was validated with low variability in intra-day and inter-day analyses. This validated method is robust and suitable for routine quality control and analysis of Trabectedin in APIs and pharmaceutical products.

KEYWORDS: RP-HPLC, Trabectedin, method development, method validation, pharmaceutical formulations, API analysis, ICH guidelines.

Analytical chemistry, a crucial branch of science, is employed to understand the composition and structure of matter by acquiring, practicing, and conveying information. It is not limited to specific compounds or reactions but encompasses the study of both natural and synthetic materials. Analytical chemistry's properties include geometrical features like molecular morphologies and species identity. This field's development includes concepts and theories related to food safety and quality, pharmaceuticals, water, environmental monitoring, biomedical applications, forensic support, and disease diagnosis, among others. Analytical chemists are essential in supporting these areas.

To identify and measure chemical species in a sample, analytical chemists utilize various techniques. One common method involves comparing the sample to a standard reference material, a known substance whose concentration is established. Most techniques in analytical chemistry rely on this comparative approach.

Every year, new or modified drugs are introduced to the market. However, there is often a delay between a drug's market introduction and its inclusion in pharmacopoeias, leading to a lack of established analytical methods for these drugs. This gap necessitates the development of new analytical methods.

In existing literature, appropriate analytical methods for certain drugs may not be available. Interferences from excipients in drug formulations can further complicate the analysis, making suitable methods scarce. Additionally, the use of expensive reagents and solvents can complicate extraction and separation procedures, reducing their reliability.

Chromatography- an overview
To resolve a multi-component mixture into its individual components, chromatography, is a new, well-known and a primary tool of separation and it can be applied both quantitatively and qualitatively. Despite, some other methods like IR spectroscopy, Nuclear Magnetic

resonance spectroscopy or Mass spectroscopy etc. are required for the final identification and confirmation.

Tswett. M, in 1806, in Warsaw, invented a new technique, while separating the plant pigments by a column of calcium carbonate, which acted as an adsorbent and the different substances get adsorbed to different extent and this give rise to the different coloured bands, at different positions on the column. In Greek, chroma means colour and graphos means writing. Hence, he termed the system of coloured bands as the chromatogram and the method as chromatography.

To separate coloured as well as colourless substances, recent advances have been made there after. Thus, in general, a sample moves over a stationary phase through the mobile phase in chromatography. Chromatography is one of the best and most likely used analytical techniques, now-a-days and in foreseeable future. It is the cornerstone of molecular analytical chemistry. Recent advancements of chromatography have been introduced by A.P.J. Martin and R.L.M. Synge in 1941, made them noble prize winners.

High Performance Liquid Chromatography (HPLC)

HPLC is used to figure out the amount of specific compound in a solution. It supports reliable quantitative range to allow the determination of substances in a single run. This method is considered to be rapid, accurate, precise and specific and offers the ease of automation. It is because methods using HPLC have more advantages over the conventional methods.

Principle of HPLC

A mixture of sample is dissociated into components for its identification, quantification and purification by HPLC, due to the differences in their relative affinities for the mobile phase and stationary phase used. Especially, RP-HPLC, relies on the principle of hydropohobic interactions, as the more non-polar the material is, longer it will be retained. Due to their low affinities and polar nature, most of the drugs elute at a faster rate through the column and so they are separated and detected easily.

The optimization of laboratory resources is ensured by the effective method development, while methods meet the objectives required at each stage of drug development. To approve the drug, at certain stages, method development is required by the regulatory agencies.

Types of HPLC

There are following variants of HPLC, depending upon the phase system (stationary) in the process:

1. Normal Phase HPLC

This method separates analytes on the basis of polarity. NP-HPLC uses polar stationary phase and non-polar mobile phase. Therefore, the stationary phase is usually silica and typical mobile phases are hexane, methylene chloride, chloroform, diethyl ether, and mixtures of these. Polar samples are thus retained on the polar surface of the column packing longer than less polar materials.

2. Reverse Phase HPLC

The stationary phase is nonpolar (hydrophobic) in nature, while the mobile phase is a polar liquid, such as mixtures of water and methanol or acetonitrile. It works on the principle of hydrophobic interactions hence the more nonpolar the material is, the longer it will be retained.

3. Size-exclusion HPLC

The column is filled with material having precisely controlled pore sizes, and the particles are separated according to it's their molecular size. Larger molecules are rapidly washed through the column; smaller molecules penetrate inside the porous of the packing particles and elute.

Application of HPLC

The information that can be obtained by HPLC includes resolution, identification and quantification of a compound. It also aids in chemical separation and purification. The other applications of HPLC include:

- Pharmaceutical Applications

1. To control drug stability.
2. Tablet dissolution study of pharmaceutical dosages form.
3. Pharmaceutical quality control.

- Environmental Applications

1. Detection of phenolic compounds in drinking water.
2. Bio-monitoring of pollutants.

- Applications in Clinical Tests

1. Urine analysis, antibiotics analysis in blood.
2. Analysis of bilirubin, biliverdin in hepatic disorders.
3. Detection of endogenous Neuropeptides in extracellular fluid of brain etc.

UV-Vis spectroscopy

UV-Vis spectroscopy is an analytical technique that measures the amount of discrete wavelengths of UV or visible light that are absorbed by or transmitted through a sample in comparison to a reference or blank sample. This property is influenced by the sample composition, potentially providing information on what is in the sample and at what concentration. Since this spectroscopy technique relies on the use of light, let's first consider the properties of light.

Origen and Characteristics of UV-Visible Spectrum

UV-VIS spectrum results from the interaction of electromagnetic radiation in the UV-Visible region with molecules, ions or complexes. It forms the basis of analysis of different substances such as, inorganic, organic and biomolecules. These determinations find applications in research, industry, clinical laboratories and in the chemical analysis of environmental samples. It is therefore important to learn about the origin of the UV-VIS spectrum and its characteristics.

Radiation and energy

Radiation is a form of transmitted energy Electromagnetic radiation is so-named because it has electric and magnetic fields that simultaneously oscillate in planes mutually perpendicular to each other and to the direction of propagation through space Electromagnetic radiation has the dual nature: its exhibits wave properties and particulate properties.

The nature of light

Light is a form of energy. Energy can be transferred from one point to another point either by particle motion or by wave motion. Accordingly, different theories on the nature of light have been proposed. The important theories are as follows: -

1. Particular like theory.
2. Electromagnetic wave theory.

Absorption of radiation

Absorption of radiation by matter always involves the loss of energy by the radiation and a corresponding gain in energy by the atoms or molecules of the medium. The energy absorbed from radiation appears as increased internal energy, or in increased irrational and rotational energy of the atoms and molecules of the absorbing medium.

Interaction of Matter with radiation

The word spectroscopy is used to refer to the broad area of science dealing with the absorption, emission, or scattering of electromagnetic radiation by molecules, ions, atoms, or nuclei. Spectroscopic techniques are some of the most widely used analytical methods in the world today.

Absorption: A transition from a lower level to a higher level with transfer of energy from the radiation field to an absorber, atom, molecule, or solid.

Emission: A transition from a higher level to a lower level with transfer of energy from the emitter to the radiation field. If no radiation is emitted, the transition from higher to lower energy levels is called nonradioactive decay. The data that is obtained from spectroscopy is called a spectrum. Spectrum is a plot of the intensity of energy detected versus the wavelength (or mass or momentum or frequency, etc.) of the energy.

Instrumentation

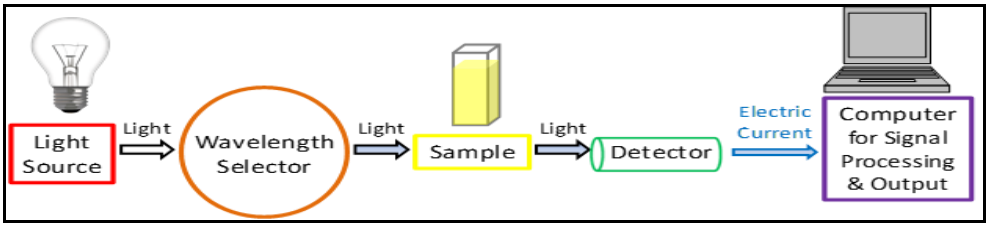


Figure 1.2: Components in a UV-Vis spectroscopy.

Light source
As a light-based technique, a steady source able to emit light across a wide range of wavelengths is essential. A single xenon lamp is commonly used as a high intensity light source for both UV and visible ranges. Xenon lamps are, however, associated with higher costs and are less stable in comparison to tungsten and halogen lamps, can be made more smoothly.

Wavelength selection
In the next step, certain wavelengths of light suited to the sample type and analyte for detection must be selected for sample examination from the broad wavelengths emitted by the light source. Available methods for this include:

Monochromators
A monochromator separates light into a narrow band of wavelengths. It is most often based on diffraction gratings that can be rotated to choose incoming and reflected angles to select the desired wavelength of light.

Method validation of drugs
According to ISO definition, validation is defined as "Verification, where the specified requirements are adequate for an intended use." Method validation can be used for qualitative, semi-quantitative or quantitative methods.

The scientific soundness of the measurement or characterization and also to varying extents throughout the regulatory submission process, the validation of analytical method is required. The method development includes the measurement of the correct substance, in correct amount and in appropriate range. The goal of method validation is to identify the critical parameters and to establish the acceptance criteria of system suitability.

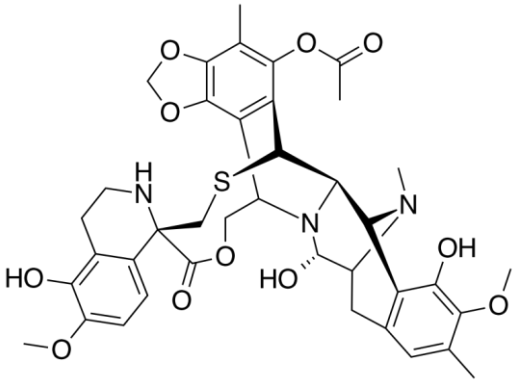
Method Validation
This process consists of establishments of the performance characteristics and the limitation of the method.

Method Performance Parameters are Determined using Equipment that is:
1. Within specification
2. Working correctly
3. Adequately calibrated

Method Validation is required when:
1. A new method is being developed
2. Revision of the established method
3. When established method are used in different laboratories and by different analysts etc.
4. Comparison of method
5. When quality control indicates method changes

Drug Profile
Trabectedin
Trabectedin, also referred as ET-743 during its development, is a marine-derived antitumor agent discovered in the Carribean tunicate Ecteinascidiaturbinata and now produced synthetically. Trabectedin has a unique mechanism of action. It binds to the minor groove of DNA interfering with cell division and genetic transcription processes and DNA repair machinery. It is approved for use in Europe, Russia and South Korea for the treatment of advanced soft tissue sarcoma.

Administration (FDA) have approved trabectedin as an orphan drug in soft tissue sarcomas and ovarian cancer. On October 23, 2015, the FDA approved trabectedin, (as Yondelis), for the treatment of specific soft tissue sarcomas.



Structure of Trabected in

Molucular Weight	761.84 g•mol−1
Color	White Powder
Melting point	205 °C

Water Solubility Solubility in water is low (0.01 mg/mL), but higher in acid (up to 1.1 mg/mL) and sufficient to allow administration in aqueous solution.

Uses Trabectedin is used is in the treatment of overactive bladder. It works equally well to antimuscarinic medication such as solifenacin or tolterodine. In the United Kingdom it is less preferred to these agents.

Trabectedin is also indicated to treat neurogenic detrusor overactivity (NDO), a bladder dysfunction related to neurological impairment, in children ages three years and older.

Mechanism of Action -Trabectedin interacts with the minor groove of DNA and alkylates guanine at the N2 position, which bends towards the major groove. In this manner, it is thought that the drug affects various transcription factors involved in cell proliferation, particularly via the transcription-coupled nucleotide excision repair system. Trabectedin blocks the cell cycle

at the G2 phase, while cells at the G1 phase are most sensitive to the drug. It also inhibits overexpression of the multidrug resistance-1 gene (MDR-1) coding for the P-glycoprotein that is a major factor responsible for cells developing resistance to cancer drugs. The agent is also thought to interfere with the nucleotide excision repair pathways of cancer cells, suggesting that it could be effective in the treatment of many cancer types including melanoma and sarcoma, as well as lung, breast, ovarian, endometrial and prostate cancers; clinical evaluations are underway in these indications.

Literature Survey

1. Analytical Method Development and Validation for the Estimation of Trabectedin in Bulk and Parenteral Dosage Form by RP-HPLC.

A new RP-HPLC method for the quantitative determination of Trabectedin was developed and validated as per ICH guidelines. The drugs were injected into Zorbax SB, C18,(150x4.6mm); 3.5µm column maintained at ambient temperature and effluent monitored at 215nm. The mobile phase consisted of phosphate buffer (pH 3.0) and Acetonitrile in the ratio of 70:30 V/V. The flow rate was maintained at 0.8 ml/min.

2. RP-HPLC Method Development and Validation for Determination of Eptifibatide Acetate in Bulk Drug Substance and Pharmaceutical Dosage Forms

A new, rapid, economical and isocratic reverse phase high performance liquid chromatography (RP-HPLC) method was developed for the determination of eptifibatide acetate, a small synthetic antiplatelet peptide, in bulk drug substance and pharmaceutical dosage forms. The developed method was validated as per of ICH guidelines. The chromatographic separation was achieved isocratically on C18 column (150 x 4.60 mm i.d., 5 µM particle size) at ambient temperature using acetonitrile (ACN), water and trifluoroacetic acid (TFA) as mobile phase at flow rate of 1 mL/min and UV detection at 275 nm

3. Analytical Method Development and Validation forthe Estimation of Trabectedin in Bulk And Parenteral Dosage Form By RP-HPLC.

A new RP-HPLC method for the quantitative determination of Trabectedin was developed and validated as per ICH guidelines. The drugs were injected into Zorbax SB, C18,(150x4.6mm); 3.5µm column maintained at ambient temperature and effluent monitored at 215nm. The mobile phase consisted of phosphate buffer (pH 3.0) and Acetonitrile in the ratio of 70:30 V/V.

4. Development and validation of RP HPLC method for the estimation of Sofosbuvir and related impurity in bulk and pharmaceutical dosage form.

The present work is aimed at development and validation of RP HPLC method which is simple, specific, precise, and accurate for estimation of Sofosbuvir and its process-related impurity in bulk and pharmaceutical dosage forms. Extensive literature survey revealed no method for estimation of the above said.

5. Development and Validation of RP - HPLC Method for the Estimation of Tylosin Tartrate in Pure and Pharmaceutical Formulation.

A simple, fast, precise, selective and accurate RP-HPLC method was developed and validated for thesimultaneous determination of tylosin tartrate from pharmaceutical formulation. Chromatographic separationwas achieved gradient on a phenomenex c18 column (250 x 4.6 mm, 5 µ particle size) using a mobile phase. Acetonitrile and water in the ratio of 90:10.the flow rate was 1.5ml / min and effluent was detected at 292 nm. The retention time of tylosin tartrate was found to be 2min. linearity was observed in the concentration range of50 -250µg /ml.

AIM AND OBJECTIVE

Aim

The aim of this study is to develop and validate a robust RP-HPLC method for the accurate quantification of trabectedin in both its active pharmaceutical ingredient (API) form and pharmaceutical formulations.

Objectives

- Literature Survey.
- Selection of Drug.
- Selection of Method and Instrument.
- To developed sensitive, rapid, accurate, precise and desirable method for estimation of drug by RP-HPLC method.
- To develop RP-HPLC method forMirabegron.
- To validate Novel Qualitative and Quantitative Analytical method development for estimation of Mirabegron in bulk and pharmaceutical formulation as per ICH guidelines.
- Statistical analysis of the recovery data obtained from different techniques for Mirabegron.

Validation

Linearity

The linearity of peak area response for Trabectedinwas determined from 10% to 50% level of working concentration of Trabectedin. The stock solutions of standard Trabectedinwas diluted to six different known concentrations. Linearity graph of concentration (as x-value) versus area (as y-value) were plotted and correlation coefficient, y-intercept and slope of the regression were calculated.

Table: Linearity Result of Trabectedin.

Linearity		
Sr. No	**Concentration (µg/mL)**	**Peak Area**
1	30	723810
2	60	1447620
3	90	2121430
4	120	2895240
5	150	3619050
6	180	4342860

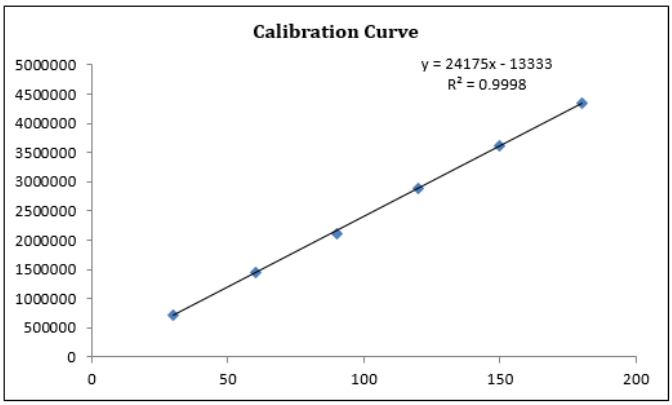
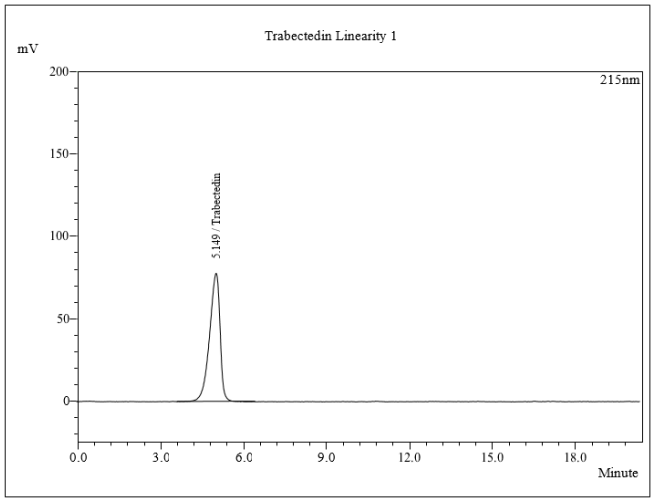
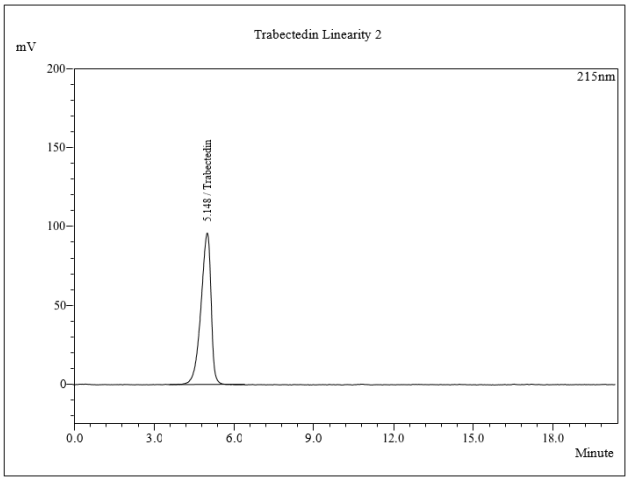


Figure: Calibration Curve of Trabectedin.



A typical Chromatogram of Linearity level 1.



A typical Chromatogram of Linearity level 2.

Characteristic parameters of Trabectedinfor the proposed HPLC method.

Parameter	Result
	Trabectedin
Calibration range (µg/ml)	30-180
Detection wavelength (nm)	215
Solvent (Acetonitrile: Buffer)	55:45
Regression equation (y*)	Y = 24175x - 13333
Slope (b)	24175
Intercept (a)	13333
Correlation coefficient(r2)	0.9998
Limit of Detection (µg/ml)	3.0885
Limit of Quantitation (µg/ml)	9.3591

System Suitability

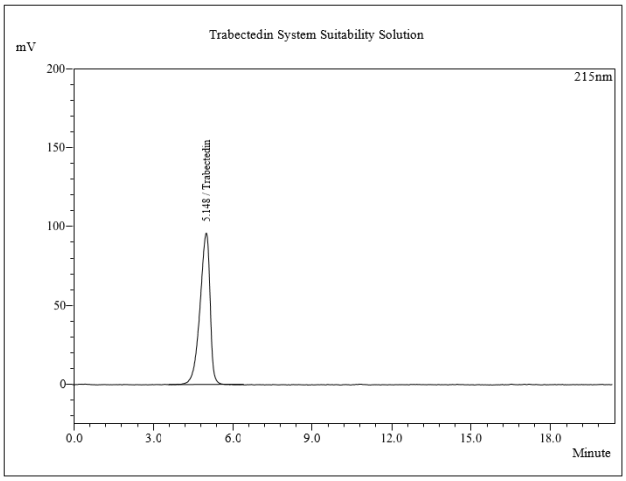
System-suitability tests are an integral part of method development and are used to ensure adequate performance of the chromatographic system. Retention time (Rt), number of theoretical plates (N) and tailing factor (T) were evaluated for six replicate injections of the drug at a concentration of 20 µg/ml. The results which are given in Table No 7.15.were within acceptable limits.

Table: System suitability studies of Trabectedinby HPLC method.

System Suitability Parameter		
Retention time (min)	Concentration (µg/mL)	5.145
Peak area	30	723810
Theoretical plates		12849
Asymmetric Factor		1.3

Table: System suitability studies of Trabectedin by HPLC method.

Formulation	
Name of Formulation	Trabectedin Powder For Concentrate For Solution for Infusion
Type of Formulation	Infusion
Concentration (mg)	1



A typical Chromatogram of System Suitability Solution

Specificity

Chromatogram of blank was taken as shown in Fig. Chromatogram of Trabectedin showed peak at a retention time of 2.910 min. The mobile phase designed for the method resolved the drug very efficiently. The Retention time of Trabectedin was 2.910 ± 0.0078min. The wavelength 315 nm was selected for detection because; it resulted in better detection sensitivity for the drug. The peak for Trabectedin from the tablet formulation was Trabectedin.

Table: Specificity of Trabected in by HPLC method.

Specificity				
Sample	Label Claim (mg)	Amount Found	Recovery	Retention Time
Infusion	1	0.91	91	5.145

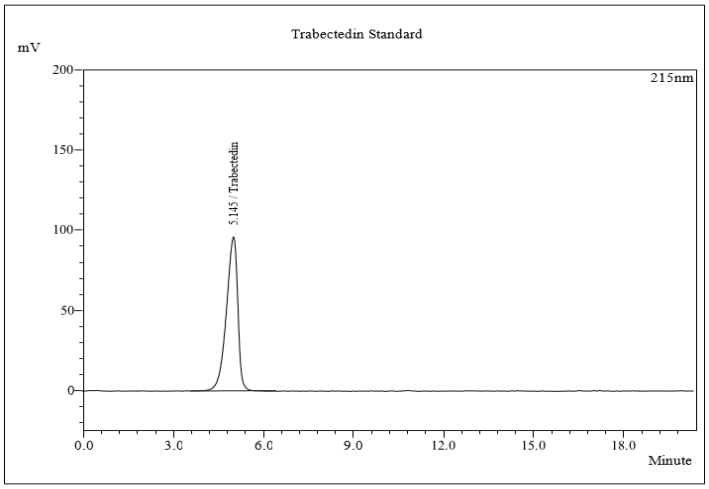
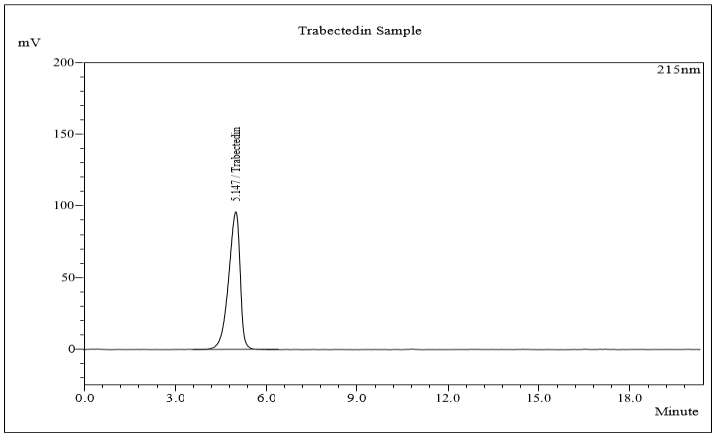


Figure: A typical chromatogram of Trabectedin standard [Concentration 60ug/ml].



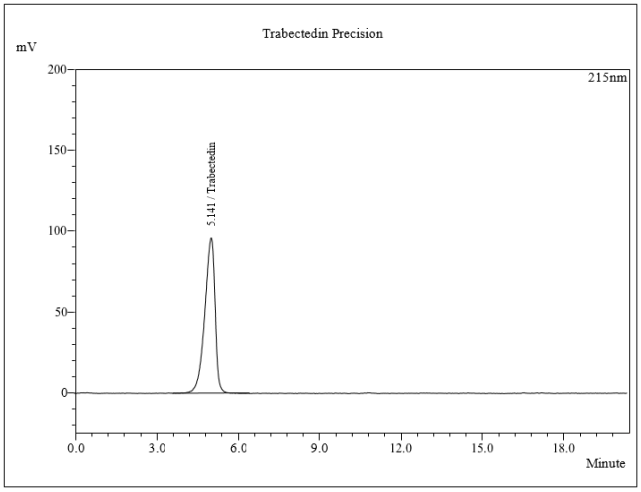
A typical chromatogram of Trabectedin Sample [Concentration 60ug/ml].

Precision

Demonstration of precision was done under two categories. The injection repeatability (System Precision) was assessed by using six injections of the standard solution of Trabectedin and the % RSD of the replicate injections was calculated. In addition, to demonstrate the precision of method (Method Precision), six samples from the same batch of formulation were analyzed individually and the assay content of each sample was estimated.

Table: Intraday Precision of Trabectedin.

Precision			
Sr. No	**Concentration (µg/mL)**	**Intraday**	**Interday**
1	120	2900745	2950745
2	120	2893596	2960946
3	120	2898275	3022160
4	120	2879568	3037630
5	120	2950487	3047528
6	120	2898228	3057409
Average		2903483.2	3012736



A typical chromatogram of Intraday Precision of Trabectedin[Concentration 60ug/ml].

Accuracy

Recovery studies by the standard addition method were performed with a view to justify the accuracy of the proposed method. Previously analysed samples of Trabectedin (20 µg/ml) were spiked with 80, 100, and 120 % extra Trabectedin standard and the mixtures were analysed by the proposed method.

Recovery Table

Recovery				
Sr. No	**Amount of Sample (µg/ml)**	**Amount of Drug Added (µg/ml)**	**Amount of Drug Recovered (µg/ml)**	**Recovery %**
1	120	60	59.989	99.98
2	120	120	119.99	99.99
3	120	180	180.01	100.01

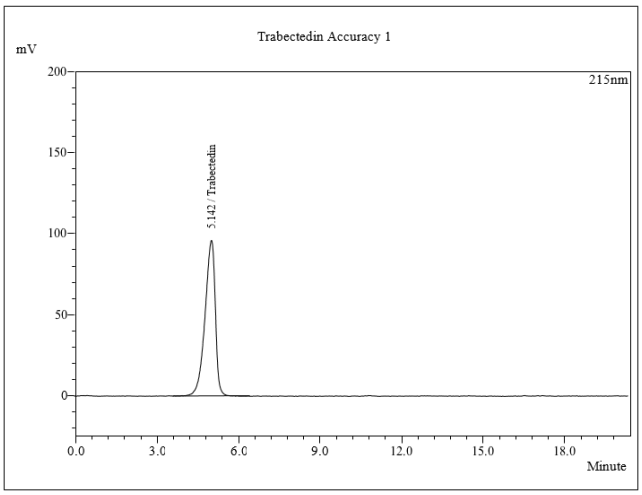


Figure: Chromatogram of Recovery level 1.

Summary

The contents of the thesis have been divided into Nine chapters and appropriate references have been placed after the 9th chapter.

Spectrophotometric method was developed for the estimation of Trabectedinin Pharmaceutical Formulation.

- Designed of Experiment.
- Optimized and Developed method for Spectrophotometry.
- Spectrophotometric method was validated for Linearity, Accuracy, Interday & Intraday Precision, Specificity & Selectivity, Sensitivity, Robustness.
- Optimized and Developed method for Chromatography.
- Chromatographic method was validated for Linearity, Accuracy, Interday & Intraday Precision, Specificity & Selectivity, Sensitivity, Robustness.
- All the developed methods were successfully applied to determine the drugs in Pharmaceutical preparation.

Trabectedin is a potent antineoplastic agent used in the treatment of soft tissue sarcomas and ovarian cancer. The accurate quantification of trabectedin in both its active pharmaceutical ingredient (API) and pharmaceutical formulations is essential for ensuring therapeutic efficacy and safety. This study focused on the development and validation of a reverse-phase high-performance liquid chromatography (RP-HPLC) method for the precise estimation of trabectedin.

Method Development

- The RP-HPLC method was developed using a suitable chromatographic column (typically C18 phase) and an optimized mobile phase composition.
- Parameters such as pH, buffer concentration, and organic solvent ratio were optimized to achieve efficient separation and good peak resolution of trabectedin from potential impurities and matrix components.

Validation Parameters

- The developed RP-HPLC method was validated according to International Conference on Harmonisation (ICH) guidelines.
- Validation parameters included specificity, linearity, accuracy, precision, limit of detection (LOD), limit of quantitation (LOQ), and robustness.
- Specificity was confirmed by demonstrating separation of trabectedin from other components in the formulation or matrix.
- Linearity was established over a range of concentrations, ensuring that the method could accurately quantify trabectedin within this range.
- Accuracy and precision studies verified the reliability of the method in terms of recovery and reproducibility.
- LOD and LOQ were determined to assess the method's sensitivity, indicating the lowest detectable and quantifiable concentrations of trabectedin.
- Robustness studies evaluated the method's robustness against minor variations in analytical conditions, such as mobile phase composition or flow rate.

Applications

- The validated RP-HPLC method was successfully applied to quantify trabectedin in API samples and various pharmaceutical formulations, including injections and lyophilized powders.
- Sample preparation techniques were optimized to ensure efficient extraction and minimal matrix interference, enhancing the method's applicability and reliability in routine analysis.

CONCLUSION

For routine analytical purpose, it is always necessary to establish methods capable of analyzing huge number of samples in a short time period with due accuracy and precision. Mirabegron is official in Indian Pharmacopoeia.

A very few analytical methods appeared in the literature for the determination of Mirabegron includes HPLC, HPTLC and UV- Visible spectrophotometric methods. In view of the above fact, some simple analytical methods were planned to develop with sensitivity, accuracy, precision and economical. In the present investigation HPLC method (Using Quality by Design) for the quantitative estimation of Mirabegron in bulk drug and per ICH guidelines pharmaceutical formulations has been developed. HPLC methods were validated as and results of linearity, precision, accuracy, Specificity, System suitability and robustness pass the limit. The HPLC method is more sensitive, accurate and precise compared to the previously reported method. There was no any interference of excipients in the recovery study. The low value of %RSD, molar extinction coefficient (L mol-1 cm-1) suggested that the developed methods are sensitive. The proposed high-performance liquid chromatographic method has also been evaluated over the accuracy, precision and robustness and proved to be convenient and effective for the quality control of Mirabegron. Developed method was found simple and cost effective for the quality control of Mirabegron.

Moreover, the lower solvent consumption leads to a cost effective and environmentally friendly Spectroscopic procedure. Thus, the proposed methodology is rapid, selective, requires a simple sample preparation procedure, and represents a good procedure for Mirabegron.

The development and validation of an RP-HPLC method for the estimation of trabectedin in both active pharmaceutical ingredient (API) and pharmaceutical formulations have been successfully achieved. This study aimed to establish a reliable analytical method that ensures accurate quantification of trabectedin, a potent antineoplastic agent used in the treatment of soft tissue sarcomas and ovarian cancer.

Through meticulous method development, various parameters such as mobile phase composition, column type, and detection wavelength were optimized to achieve efficient separation and sensitivity. The method demonstrated excellent specificity, with trabectedin being well resolved from potential impurities and excipients present in pharmaceutical formulations. Linearity studies confirmed the method's ability to accurately measure trabectedin over a wide concentration range, essential for both quality control during manufacturing and pharmacokinetic studies.

Validation of the RP-HPLC method included rigorous assessment of accuracy, precision, robustness, and system suitability parameters. The results met acceptance criteria set by regulatory guidelines, ensuring the method's reliability and reproducibility in routine

analysis. Limits of detection (LOD) and quantitation (LOQ) were determined, indicating the method's sensitivity in detecting trabectedin at trace levels in complex matrices.

Applications of the validated RP-HPLC method in pharmaceutical formulations demonstrated its practical utility in drug analysis. Sample preparation techniques, including extraction and dilution methods, effectively recovered trabectedin from matrices while minimizing interference. The method's performance in pharmaceutical formulations, such as injections and lyophilized powders, underscored its versatility and applicability across different dosage forms.

Overall, the RP-HPLC method developed and validated for trabectedin analysis represents a robust tool for pharmaceutical quality control and pharmacokinetic studies. Its successful implementation ensures accurate determination of trabectedin content, contributing to the efficacy, safety, and regulatory compliance of trabectedin-based therapies in clinical settings.

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