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ANALYTICAL METHOD DEVELOPMENT AND VALIDATION OF SECNIDAZOLE IN ITS PURE AND PHARMACEUTICAL DOSAGE FORM BY USING UV-VISIBLE SPECTROPHOTOMETRIC METHOD

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Abstract

UV-spectrophotometry refers to absorption spectroscopy in the ultraviolet-visible spectral region. This method of analysis is gaining importance as it is rapid, simple, precise, less time consuming and accurate. The objective of present investigation was to develop an accurate, rapid and robust method for determination of Secnidazole in API and in pharmaceutical preparations by using UV spectrophotometric method. Secnidazole shows maximum absorbance at a wavelength of 305nm, which is used for this study. The method provides a linear response from a quantitation range of 06 µg/ml to 14 µg/ml in ethanol with regression equation y = 0.032x - 0.002 and r^2 0.999 Interday precision and accuracy was found to be below 0.2 and above 99.00% respectively for the developed method. Thus the developed method suitably applied for regular quality control of Secnidazole in pure and pharmaceutical preparations.

Keywords: UV-VIS Spectrophotometer, Secnidazole, Ethanol.

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1. Introduction [1, 2]

Ultraviolet Visible Spectroscopy is concerned with the study of absorption of UV radiation which ranges from 200 nm to 800 nm. Compounds which are colorless absorb radiation in the UV region. In both UV as well as visible spectroscopy, only the valence electrons absorb the energy, thereby the molecule undergoes transition from ground state to excited state. This absorption is characteristic and depends on the concentration and path length as given by Beer – Lambert's law. Any molecule has n, π or σ or a combination of these electrons. These bonding (σ and π) and non-bonding (n) electrons absorb the characteristic radiation and undergoes transition from ground state to excited state.

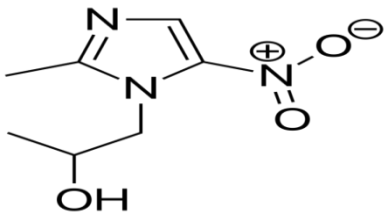
Components of a Spectrophotometer

Instruments for measuring the absorption of U.V. or visible radiation are made up of the following components;

1. Sources (UV and visible)
2. Wavelength selector (monochromator and filters)
3. Sample containers (cuvettes)
4. Solvents
5. Detector
6. Signal processor and readout

2. Drug Profile [3]

Structure of Secnidazole



IUPAC Name

1-[2-methyl-5-nitro-1H-imidazole-1-yl]propan-2-ol.

Solubility

Secnidazole soluble in methanol, ethanol, chloroform, dimethyl formamide, NAOH and chloroform.

Category

Antibiotics

Mechanism of Action

Secnidazole is selective against many anaerobic Gram - positive and Gram -negative bacteria as well. Once it enters bacteria and parasites, secnidazole is activated by bacterial or parasitic enzymes to form a radical anion, there by damaging and killing the target pathogen.

3. Materials and Methods

Table No. 1:- Chemicals used for the Study:

Chemicals	Company Name
Secnidazole Tablet	Secnilforte(Abbott)
Secidazole(Pure Drug)	S D Fine Chemicals
Ethanol	Merck
Distilled Water	Merck

Table No.2 :- Equipment's and Glassware's used For the study

Equipment And Glass wares	Company Name
Double beam UV –VIS spectrophotometer	LABINDIA
Analytical balance	WENSAR
Standard flask	Borosil
Pipette	Borosil
Beakers	Borosil
Measuring cylinder	Borosil

Preparation of stock solution

Preparation of standard stock solution

Weigh accurately 10mg of standard substance of Secnidazole and transfer it into 10ml volumetric flask and dissolve it in ethanol and make up the solution up to 10 ml with ethanol(Stock A-1000µg/ml). Pipette out 1ml from the above solution and transfer into 10ml volumetric flask and dissolve it in ethanol and make up the solution up to 10 ml with ethanol(Stock B-100µg/ml

Preparation of Sample Stock Solution

Transfer 10mg equivalent weight of tablet powder of secnidazole into 10ml volumetric flask and dissolve it in ethanol and make up the solution up to 10 ml with ethanol (Stock A -1000µg/ml).

Pipette out 1ml from the above solution and transfer into 10ml volumetric flask and dissolve it in ethanol and make up the solution up to 10 ml with ethanol (Stock B-100µg/ml).

4. Result and Discussions

4.1. Solubility profile

Table no 3: showing solubility profiles

Solvent name	Solubility
Ethanol	Freely soluble
Methanol	Freely soluble
Chloroform	Freely soluble
0.1N NAOH	Freely soluble
0.1N HCL	Freely soluble
Water	Sparingly soluble

4.2.Selection of wavelength

The absorbance of the solution containing Secnidazole at 10 µg/ml was determined in the UV range 200-400 nm using ethanol as blank.

The maximum absorbance was found to be 305nm.

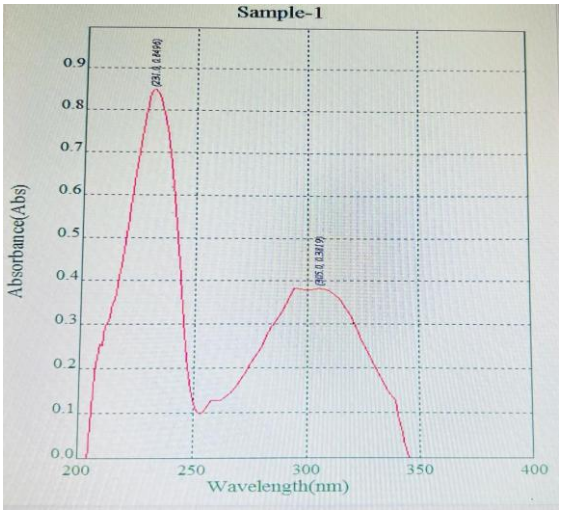


Figure no 1: λ_{max} of Secnidazole.

The maximum wavelength was found to be 305 nm, andmaximum absorbance was 0.3819.

4.3. Assay

Procedure

Place the working standard solution and sample solution into the UV spectrophotometric system and measure the absorbance of the Secnidazole and calculate the %Assay by using the formulae.

Calculation

$$\frac{AT}{AS} \times \frac{WS}{WT} \times \frac{DT}{DS} \times \frac{P}{100} \times \frac{Avg\ Wt.}{Label\ Claim} \times 100$$

Where,

AT =test Absorbance of Secnidazole

AS = standard Absorbance of Secnidazole

WS = Weight of working standard

WT = Weight of sample

DS = Dilution of Standard solution

DT = Dilution of sample solution

P = Percentage purity of working standard.

$$= \frac{0.3900}{0.3819} \times \frac{1}{1} \times \frac{0}{16} \times \frac{99.8}{100} \times \frac{1600}{1000} \times 100$$

= 101.8%

The % Purity of Secnidazole in Pharmaceutical dosage form was found to be 101.8%.

Acceptance criteria

The percentage assay should be between 98-102%.

Conclusion:

It was concluded that percentage assay was found within the limits (101.8%).

4.4. Validation Parameters

4.4.1. Specificity

The blank solution was placed in the UV and spectrum was recorded.

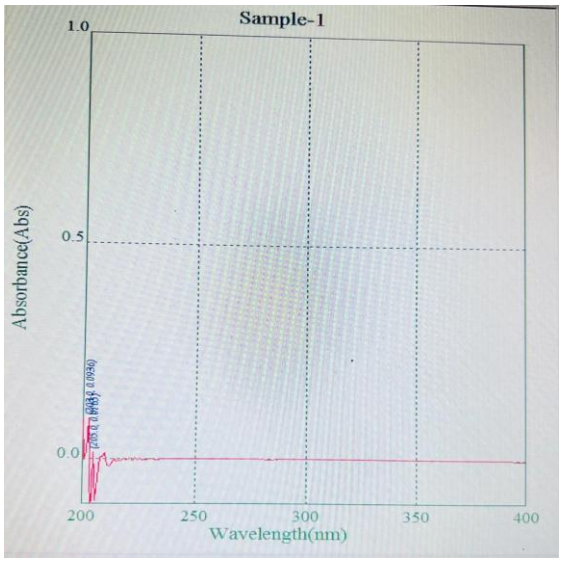


Figure no.2: Spectrum showing blank

Observation

From the spectrum we can conclude that excipients or solvents are not interfering the spectrum of secnidazole.

4.4.2. Linearity

Table No-4 calibration curve of Secnidazole

Conc. [µg/ml]	Absorbance
6	0.1919
8	0.2548
10	0.3256
12	0.3856
14	0.4511

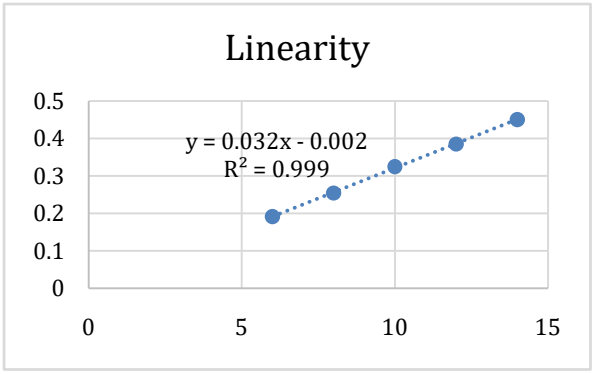


Fig no.3- Calibration Curve of Secnidazole

Acceptance Criteria

The Correlation Coefficient should be not less than 0.999.

Conclusion: Correlation coefficient (r) is 0.99.Regression Coefficient (r^2) is 0.99. These values meet the Validation Criteria. Hence this method is applicable

4.4.3. ACCURACY

To the check the accuracy of the method, recovery studies were carried out by addition of standard drug solution to pre-analysed sample solution at three different levels 80%, 100%, 120%.

Table no. 5:- Table showing results for accuracy

Accuracy level	Conc. taken (ug/ml)	Absorbance	Conc. found	Recovery	% Mean
80	18	0.5799	18.18	101	100.8
80	18	0.5793	18.16	100.8	
80	18	0.5788	18.15	100.8	
100	20	0.6374	19.98	99.9	99.8
100	20	0.6371	19.97	99.8	
100	20	0.6370	19.96	99.8	
120	22	0.6946	21.76	98.9	99.03
120	22	0.6964	21.82	99.1	
120	22	0.6960	21.81	99.1	

Acceptance criteria: The Percentage recovery should lie between 98-102%.

Conclusion: The results obtained for recovery at 80%, 100%.120% are within the limits. Hence method is accurate.

4.4.4. PRECISION:

Repeatability

Precision was determined by analyzing standard preparations of Secnidazole(10ug/ml) for five times.

Table No.6:- Results of Repeatability

Concentration	Absorbance
10 µg/ml	0.3809
10 µg/ml	0.3810
10 µg/ml	0.3806
10 µg/ml	0.3815
10 µg/ml	0.3815
Average	0.3811
SD	0.000394
%RSD	0.1033

Acceptance Criteria:

The % Relative Standard Deviation of Secnidazole from the five sample preparations should be not more than 2.0%.

Conclusion

The %RSD obtained is within the limits (0.1033) .Hence the method is precise.

Intermediate precision

Inter-day Precision

The inter-day precision of the proposed method was determined on samples of drug solutions at different days.

Table no.7:-Results of Method Precision (Day-1)

Concentration	Absorbance
10 µg/ml	0.3809
10 µg/ml	0.3810
10 µg/ml	0.3806
10 µg/ml	0.3815
10 µg/ml	0.3815
Average	0.3811
SD	0.000394
%RSD	0.1033

Acceptance Criteria:

The % Relative Standard Deviation of Secnidazole from the five sample preparations should be not more than 2.0%.

Conclusion: The %RSD obtained is within the limits (0.1033) .Hence the method is precise.

Table no: 8 - Results of Method Precision (Day-2)

concentration	Absorbance
10 µg/ml	0.3911
10 µg/ml	0.3902
10 µg/ml	0.3910
10 µg/ml	0.3914
10 µg/ml	0.3909
Average	0.39092
SD	0.000444
%RSD	0.1135

Acceptance Criteria

The % Relative Standard Deviation of Secnidazole from the six sample preparations should be not more than 2.0%.

Conclusion: The %RSD obtained is within the limits (0.11).Hence the method is precise.

4.4.5. Limit of Detection (LOD)

$$\mathbf{LOD} = \mathbf{3.3X}\frac{\boldsymbol{\sigma}}{\boldsymbol{S}}$$

Where

σ = the standard deviation of the response

S = the slope of the calibration curve

The limit of Detection for the method was found to be 10.425µg/ml

4.4.6. Limit of Quantification (LOQ)

$$\mathbf{LOQ} = \mathbf{10X}\frac{\boldsymbol{\sigma}}{\boldsymbol{S}}$$

The limit of Detection for the method was found to be 31.59µg/ml

4.4.7. Robustness

To demonstrate the robustness of the method, prepared standard solution were placed in the UV and spectrums and absorbance values were recorded by changing the parameters like wavelength and temperature. There was no significant change in absorbance values.

Table no.9: Result of Robustness

Parameter	Absorbance
Wavelength	
303nm	0.3148
305nm	0.3809
307nm	0.3182
Temperature	
26^0C	0.3815
28^0C	0.3810
30^0C	0.3799

5. Summary and Conclusion

Summary

A UV spectrophotometric method has been developed and validated for the determination of secnidazole in its pure and pharmaceutical dosage form. The process was done by using ethanol as a solvent with the detection wavelength set at 305nm. The method was linear with the correlation coefficient 0.999 in the concentration range of 6-14ug/ml. The limit detection and the limit quantification were 10.425ug/ml and 31.59ug/ml respectively. The repeatability of inter day 1 and day 2 precision were satisfactory, the relative standard deviation did not exceed 2%. The accuracy of the method is high as can be seen from mean recovery values of secnidazole which were in the range of 100.8, 99.8,99.03% and the method is robusted. The method met the ICH regulatory requirements.

Conclusion

A simple, economical, rapid, precise and UV spectrophotometric method was developed for the estimation of secnidazole in its pure and pharmaceutical dosage form. The method was developed by using ethanol

as solvent. The developed method was validated for parameters viz accuracy, precision, linearity, robustness, limit of detection and limit of quantification as per ICH guidelines. All the parameters were found to be within the acceptance limits. The results indicated that the proposed method for estimation of secnidazole is very accurate and cost effective and can be employed in routine sample analysis of secnidazole in its pure and pharmaceutical dosage form.

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9. Conflict of Interest
Authors are declared that n conflict on interest.

10. Informed Consent & Ethical Statement
Not Applicable

11. Author Contribution
All authors are contributed equally.

12. References
1. Sharma B. Instrumental Methods of Chemical Analysis. 7th ed. Meerut: Goel Publishing House; 2000. p. 1-2.
2. Beckett H., Stenlake JB., Practical pharmaceutical chemistry. 4th edition. New Delhi (Ind): C.B.S Publications; 2002. Page.1.
3. Indian pharmacopoeia vol.4 page no-2713-2715.
4. A F EL WALLILY *et.al* [1] Spectrophotometric and HPLC determination of secnidazole in pharmaceutical tablets.j pharm biomed anal.2000,july
5. Oksan et al Development and Validation of UV-Spectrophotometric Procedures for Secnidazole Quantitative Determination. Journal of pharmaceutical science and Research, Vol.9[4],2017