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SIMPLE AND PRECISE UV SPECTROPHOTOMETRIC METHOD FOR THE ESTIMATION AND VALIDATION OF ALBENDAZOLE

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Abstract:
UV spectroscopic method for estimation of Albendazole was developed and validated as per international conference on Harmonization guidelines. It showed the absorption maxima at 237 nm in diluted glacial acetic acid. The developed method was found linear over the concentration range of 5 to 25 µg/ml. The regression coefficient was found to be 0.999. Intraday precision and Interday precision was found to be 0.37 and 0.46 respectively. %Recovery was obtained as 99.4%,99.3% and 99.3% for 80%,100% and 120% respectively. Therefore simple, linear, accurate, precise and highly sensitive method was developed.

Key Words: *Albendazole, UV Spectrophotometric method, Acetic acid, Validation, ICH guidelines*

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1.INTRODUCTION:

Albendazole is a benzimidazole anthelmintic used to treat parenchymal neurocysticercosis and other helminth infections. Albendazole causes degenerative alterations in the tegument and intestinal cells of the worm by diminishing its energy production, ultimately leading to immobilization and death of the parasite. It works by binding to the colchicine-sensitive site of tubulin, thus inhibiting its polymerization or assembly into microtubules. As cytoplasmic microtubules are critical in promoting glucose uptake in larval and adult stages of the susceptible parasites, the glycogen stores of the parasites are depleted. Degenerative changes in the endoplasmic reticulum, the mitochondria of the germinal layer, and the subsequent release of lysosomes result in decreased production of adenosine triphosphate (ATP), which is the energy required for the survival of the helminth.

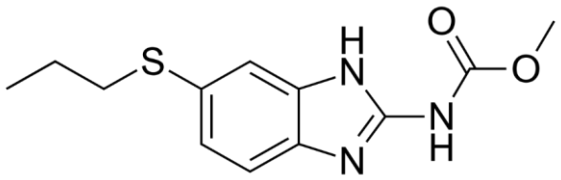

Figure 1 ALBENDAZOLE STRUCTURE

2.MATERIAL AND METHODS:

A. Solvent used

The solubility of albendazole in various solvents was checked in order to select an appropriate solvent for spectrophotometric determinations. The drug Albendazole was soluble in DMF, glacial acetic acid. In this spectrophotometric method glacial acetic acid grade and distilled water was used as the solvents.

B. Preparation of standard stock solution

An accurate weighed quantity of 10 mg of albendazole was transferred to a 10 ml volumetric flask and dissolved in diluted glacial acetic acid and the volume was made to the mark with diluted glacial acetic acid to obtain a final concentration of 1000ug/ ml. (standard stock solution ‘a’)

From stock solution ‘a' 1ml aliquot was pipetted out in a 10ml volumetric flask and made up to the mark with diluted glacial acetic acid to obtain a final concentration of 100ug/ml. (standard stock solution ‘b’)

C. Determination of lambda max

Appropriate dilution of standard stock solution ‘b' was scanned in the entire range of ultra violet light in order to obtain the wavelength of maximum absorption. The dilution was selected in such a way to give an absorbance of approximately 0.9 around which the repeatability of the measurement is optimal. The wavelength of maximum absorption selected was 237nm.

D. Selection of analytical concentration range

Appropriate aliquot of stock solution ‘b' was pipette out into a series of 10ml volumetric flasks. The volume was made up to the mark with distilled water to obtain solutions of concentration ranging from 5-25ug/ml.

E. Preparation of calibration graph

Absorbance of the above solutions was measured at 237nm. A calibration graph of concentration versus observance was established. The data obtained was analysed statistically.

The drug follows the beer’s lambert law in the concentration range of 5-25ug/ml. Regression equation was established and the correlation coefficient was determined.

3.RESULTS AND DISCUSSIONS:

Method validation: Method validation is the process used to confirm that the analytical procedure employed for a specific test is suitable for its intended use. Results from method validation can be used to judge the quality, reliability and consistency of analytical results; it is an integral part of any good analytical practice.

A. Linearity range: The linearity of analytical method is its ability to elicit test results that are directly proportional to the concentration of analysts in the sample with in a given range.

Appropriate aliquots were reported out from the standard stock solution ‘b' out in to a series of 10ml volumetric flasks. The volume was made up to the mark with distilled water to obtain a concentration range, ranging from 5 – 25ug/ml (5, 10, 15, 20, 25 ug/ml). Absorbance of above solutions was measured at 237nm. A calibration graph of the calibration versus absorbance was established.

The drug follows the beer’s lambert law in the concentration range of 5-25ug/ml. Regression equation was established and the correlation coefficient was determined.

Table 1 linearity

CONCENTRATION	ABSORBANCE
5	0.062
10	0.265
15	0.456
20	0.653
25	0.854

INFERENCE: The response was found to be linear and the correlation coefficient was found to be 0.999

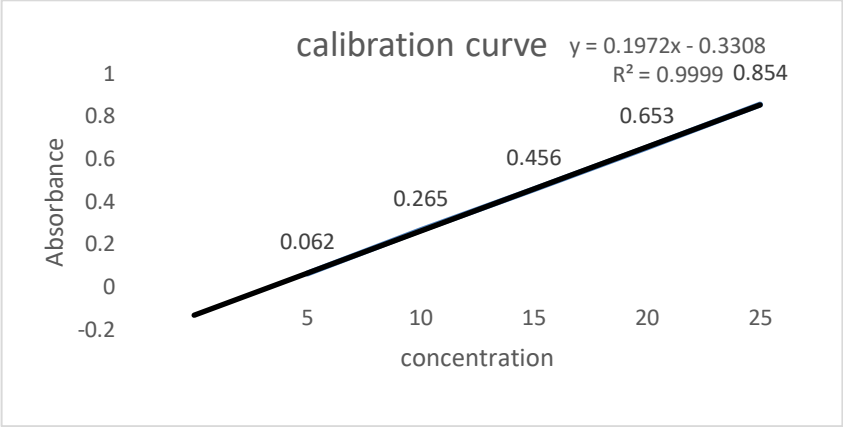

Figure 2 calibration curve

B. Accuracy: To the preanalyzed sample three different amounts of 80%, 100% and 120 % of working standard was added, at each level 3 replicate samples were prepared and samples were analyzed to determine percentage recovery from the sample. Percentage recovery is calculated for all nine readings from the ratio of amount of drug found.

Table 2 Accuracy

% LEVEL	STANDARD AMOUNT	SPIKED AMOUNT	AMOUNT FOUND	% RECOVERY	MEAN %RECOVERY
80	20	16	35.71	99.1	99.4
	20	16	35.86	99.6	
	20	16	35.89	99.6	
100	20	20	39.79	99.4	99.3
	20	20	39.89	99.7	
	20	20	39.63	99.0	
120	20	24	43.71	99.3	99.3
	20	24	43.69	99.2	
	20	24	43.76	99.4	

C. Precision: The precision was determined for Albendazole in terms of intraday precision and interday precision. Sample solution of 20 µg/ml was prepared and absorbance was measured six times in different time intervals within a day (intraday) and at 6 different days (interday). Statistical parameters such as mean, standard deviation and percentage relative standard deviation were calculated.

INTRADAY PRECISION

Table 3 Intraday precision

s.no	ABSORBANCE
1	0.7546
2	0.7521
3	0.7547
4	0.7589
5	0.7584
Mean	0.7557
SD	0.00283
%RSD	0.37

INTERDAY PRECISION

Table 4 Interday precision

S.NO	ABSORBANCE
1	0.7529
2	0.7610
3	0.7528
4	0.7531
5	0.7561
MEAN	0.7551
SD	0.00353
%RSD	0.46

D. Ruggedness The ruggedness of developed method was determined by analyst variation (analyst 1 and analyst 2).The results were analysed statistically and the effect of variation where estimated.

Table 5 Ruggedness

Analyst	ABSORBANCE
Analyst I	0.7521
Analyst II	0.7547
MEAN	0.7534
SD	0.001838
% RSD	0.24

INFERENCE: The %RSD was found to be 0.24

5. CONCLUSION:

A simple, rapid, reliable, UV spectroscopic method for estimation of Albendazole was developed and validated as per international conference on Harmonization guidelines. It showed the absorption maxima at 237 nm in diluted glacial acetic acid. The developed method was found linear over the concentration range of 5 to 25 µg/ml. The regression coefficient was found to be 0.999. Intraday precision and Interday precision was found to be 0.37 and 0.46 respectively. %Recovery was obtained as 99.4%,99.3% and 99.3% for 80%,100% and 120% respectively. Therefore, the developed method was said to be simple, linear, accurate and precise.

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