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## A SIMPLE UV SPECTROPHOTOMETRIC METHOD FOR THE ESTIMATION OF HYDROCHLOROTHIAZIDE IN BULK

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### Abstract

A rapid, simple, accurate, and economical UV spectrophotometric method has been developed for the assay of the Hydrochlorthiazide in bulk. The analysis is based on the UV absorption maxima at 273nm wavelength, using 1M sodium hydroxide solution as solvent and water as diluent. The linearity was observed in the range of 1- 60µg/mL, with the correlation coefficient, 0.998. The results of analysis were validated statistically as per ICH guidelines. The method is accurate and reproducible and can be employed for routine analysis of the drug from API.

**Keywords:** Hydrochlorthiazide, UV spectrophotometry and Validation.

### Introduction

Hydrochlorthiazide (2H-1, 2, 4 – Benzothiadiazine -7- sulphonamide – 6 – chloro - 3, 4-dihydro-,1 ,1-dioxide) is a potent thiazide diuretic and anti-Hypertensive agent. It acts by inhibiting kidneys ability to retain water. It acts on distal convoluted tubule and inhibit  $\text{Na}^+/\text{Cl}^-$  symport leading to reduction in plasma volume and cardiac output, therefore widely used alone or in combination with other Hypertensive drugs for the treatment of CVS disorders viz., Hypertension, Angina and Congestive heart failure. Literature survey reveals that few UV methods have been reported for the estimation of Hydrochlorthiazide in single and combination with other drugs, in bulk samples, formulations.<sup>(1-13)</sup> Though few methods are available in literature, these lack in simplicity and cover narrow concentration range. The present study aim to develop a simple, economic method which provides wide range of linearity.

### Materials and Methods

#### Instrumentation

A UV-Visible double beam spectrophotometer (LAB INDIA) with a spectral bandwidth 1nm using matched cuvettes made up of quartz was used. Analytical balance (Shimadzu-AY220) was used for weighing materials.

## Materials

Hydrochlorthiazide standard drug was obtained as a gift sample from Aurobindo Pharma Ltd (Hyderabad, India), NaOH (AR Grade) and double distilled water.

**Method development:** After assessing the solubility of drug in different solvents, methanol and M sodium hydroxide solution were selected as solvents, uv spectra were recorded in these two solvents, hydrochlorothiazide has shown almost similar spectra with these two solvents, finally sodium hydroxide was selected as a solvent (water as a diluent), because these are easily available, economic and eco-friendly.

## Preparation of Standard Solutions

25mg of standard Hydrochlorthiazide was accurately weighed and transferred in to 25ml volumetric flask, sufficient quantity of M NaOH solution was added to dissolve the drug, ultrasonicated for 5 minutes and volume was made up with distilled water (1000 $\mu$ g/ml). From the above standard stock solution different concentrations in the range of 1-100 $\mu$ g/ml were prepared at an interval of 5  $\mu$ g/ml using water as diluent.

## Selection of Wavelength

One of the working standard (30 $\mu$ g/ml) solutions was scanned in range of 200 - 400nm. The spectrum revealed that hydrochlorothiazide had shown a well defined absorption maximum at 273nm.

## Construction of Calibration Curve

Various aliquots of standard solutions ranging from 1-100 $\mu$ g/ml concentration of Hydrochlorthiazide were scanned at 273 nm and the absorbance was noted using water as blank. Graph was plotted by taking concentrations on X-axis and absorbencies on y-axis. The hydrochlorothiazide obeys beer's law in the range of 1-60  $\mu$ g/ml.

## Assay

25 mg of hydrochlorthiazide (bulk) was accurately weighed and transferred in to 25ml volumetric flask. Required quantity of M NaOH was added to dissolve the drug and diluted to volume with distilled water, sonicated for 5min. The concentration of this solution was 1000 $\mu$ g/ml. From this solution, 30 $\mu$ g/ml solution was prepared. The concentration of this solution was determined by measuring absorbance at 273nm, and the percentage of drug was found to be 99.7%.

## Results

### Method Validation

As per ICH guidelines, the method validation parameters were checked for Linearity, Precision, Accuracy, Robustness and Ruggedness.

### Linearity

Linearity of the method was established by plotting a graph of concentration on X-axis and absorbance on Y-axis.

The linearity was obtained in the concentration range of 1-60 µg/ml. The regression equation was  $Y=0.0446X+0.0582$  with correlation coefficient ( $r^2$ ), =0.998. The results were given in Table 1.

**Table-1: Linearity data of Hydrochlorthiazide.**

Concentration (µg/ml)	Absorbance
1	0.044
5	0.259
10	0.537
15	0.730
20	0.985
25	1.177
30	1.422
35	1.642
40	1.873
45	2.085
50	2.251
55	2.48
60	2.728

### Accuracy

Accuracy was ascertained on the basis of recovery studies by standard addition method. Recovery studies were carried out at three different levels (80,100,120) by the addition of standard drug to pre-analyzed sample solution.

These solutions were analyzed by taking absorbance and percentage recoveries were calculated. The recoveries of drug were observed to be close to 100%, representing the method was accurate. The results were given in Table 2.

**Table-2: Recovery data of Hydrochlorthiazide.**

S. No	Level	Amount of drug added	Amount of drug recovered	% recovery
1	80%	30	29.94	99.80
2	100%	40	40.04	100.1
3	120%	50	49.96	99.92

**Precision**

Precision was determined by studying the system precision and method precision. System precision was performed by analysing the standard solution for six times. In method precision, a homogenous sample of single batch was analyzed for 6 times. It was evaluated by assaying six independent test preparations of same concentration (30 $\mu$ g/ml) and %RSD was calculated. The results of statistical evaluation are given in Table 3 & Table 4.

**Table-3: System precision data of Hydrochlorthiazide.**

Concentration ( $\mu$ g/ml)	Absorbance
30	1.324
30	1.315
30	1.309
30	1.326
30	1.318
30	1.356
Average	1.324
%RSD	1.2%

**Table-4: Method precision data of Hydrochlorthiazide.**

Concentration ( $\mu$ g/ml)	Absorbance
30	1.312
30	1.324
30	1.333
30	1.345

<i>G. Vamshi Krishna* et al. International Journal Of Pharmacy &amp; Technology</i>	
30	1.316
30	1.369
<b>Average:</b>	1.333
<b>%RSD</b>	1.5%

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### Robustness

The Robustness of analytical method was studied by the analysis of six replicates of Hydrochlorthiazide standard preparation of same concentration (40µg/ml) at multiple wavelengths i.e. 270nm, 273nm and 276nm. The %RSD was calculated at each wavelength. The %RSD was found to be 1.94%, 1.68% and 1.75% at 270nm, 273nm & 276nm respectively. The results were given in table 5.

**Table-5: Robustness data for Hydrochlorthiazide.**

Concentration (µg/ml)	Absorbance		
	270nm	273nm	276nm
40	1.799	1.912	1.880
40	1.894	1.876	1.842
40	1.807	1.963	1.928
40	1.828	1.885	1.849
40	1.809	1.887	1.849
40	1.808	1.891	1.852
<b>Average</b>	1.824	1.902	1.866
<b>Standard deviation</b>	0.0355	0.0320	0.0382
<b>%RSD</b>	1.94%	1.68%	1.75%

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### Ruggedness

This study was carried out for analyst to analyst variation. Six replicates of Hydrochlorthiazide standard preparation of same concentration (20µg/ml) were prepared by different analysts and %RSD was calculated for each. The %RSD was found to be 1.1% and 0.9%. The results were given in table 6.

**Table-6: Ruggedness data for Hydrochlorothiazide.**

Concentration ( $\mu\text{g/ml}$ )	Absorbance	
	Analyst-1	Analyst-2
20	1.007	1.022
20	1.000	1.022
20	1.023	0.997
20	1.020	1.010
20	1.024	1.008
20	1.029	1.011
<b>Average</b>	1.017	1.011667
<b>%RSD</b>	1.1%	0.9%

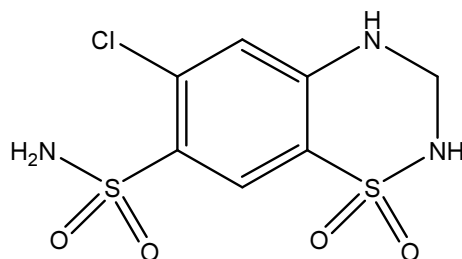
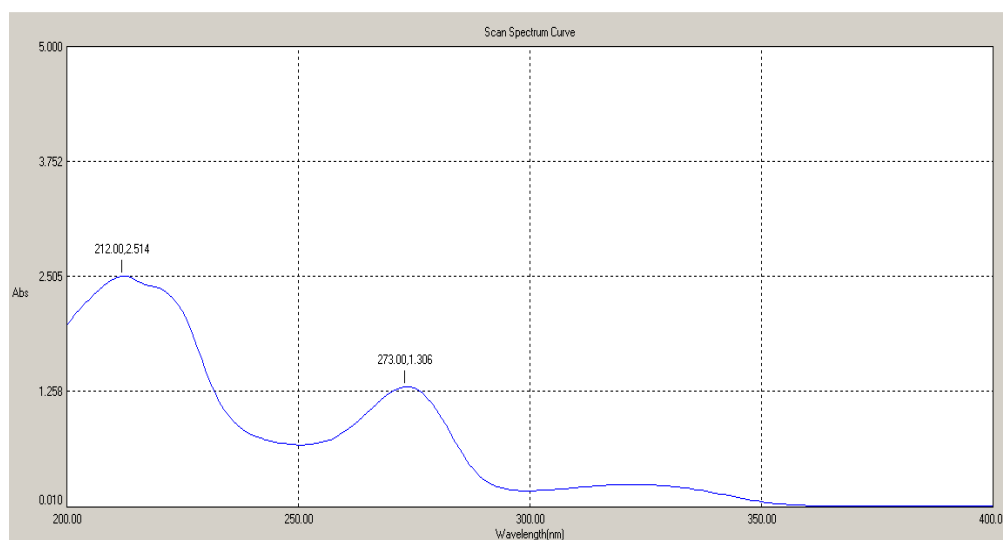
**Figure 1: Hydrochlorothiazide ( $\text{C}_7\text{H}_8\text{ClN}_3\text{O}_4\text{S}_2$ )****Figure 2: UV spectrum for Hydrochlorothiazide in 1M NaOH**

Figure 3: UV spectrum for Hydrochlorothiazide in Methanol

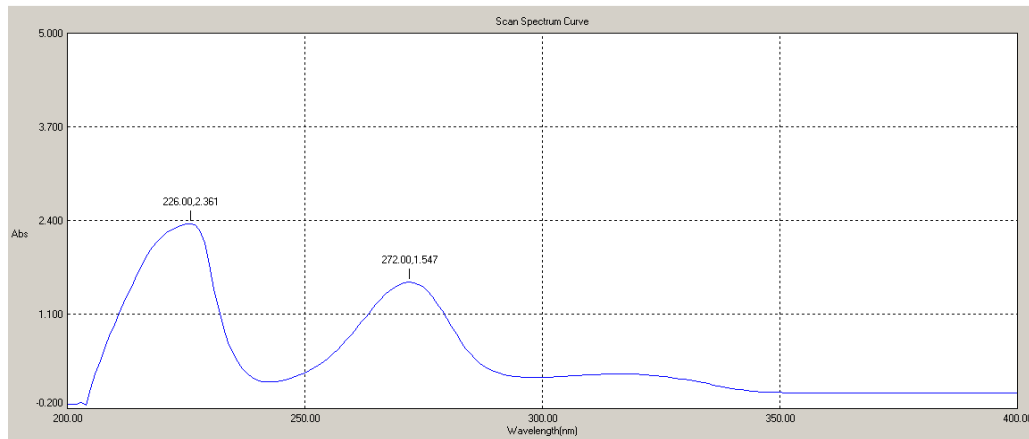
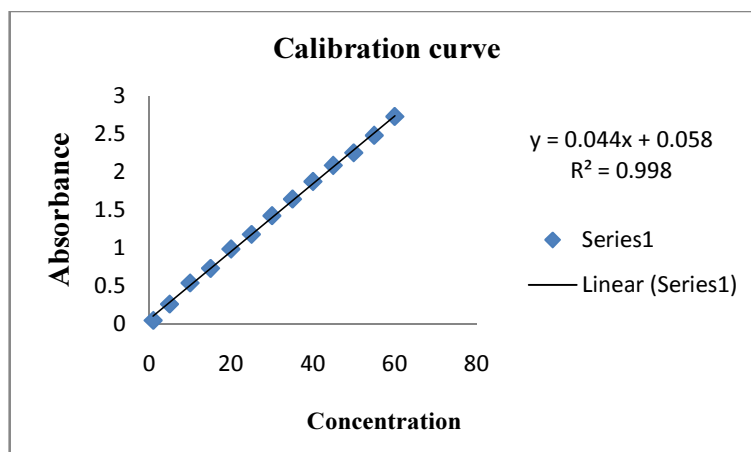


Figure 4: Calibration curve of Hydrochlorothiazide.



### Conclusion

From the above data, it is concluded that the proposed method is simple, accurate, reproducible, and inexpensive. The proposed method can be successfully employed for the routine analysis of hydrochlorothiazide in bulk drug sample.

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