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A RESEARCH ON SULTIAM TABLETS BY RP-HPLC METHOD

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ABSTRACT

The estimation of Sultiam in formulation by RP – HPLC method taking water and methanol in the ratio 70:30. Standard substance was dissolved in Methanol was scanned and the spectra was recorded and the spectrum shows that λ_{max} of Sultiam was 245 nm. The calibration curve was plotted using concentration against peak area. With the optimized chromatographic conditions, the drug was linear in the concentration range of 2.5 - 15 µg/ ml. The correlation coefficient was found to be 0.998. By using this method, the main peak of Sultiam was eluted at 5.94 minutes. In this method the optical parameters like Correlation coefficient, Slope, Intercept, LOD and LOQ were calculated.

KEY WORDS: RP-HPLC, Composition, Benzenesulfonamide, Hypromellose.

INTRODUCTION

HPLC is a type of liquid chromatography to quantify and analyse mixtures of chemical and synthetic compound for separating non-volatile species or thermally fragile compounds. This technique is widely use for the separation of materials including Amino acids, proteins, nucleic acids, hydrocarbons, carbohydrates, terpenoids, pesticides, antibiotics, steroids, metals, organic species, and variety of inorganic substances. It is also use for Nano science, molecular detection, pharmaceutical R&D, and finished doses products analysis. The present work was done on sultiam tablets. An analytical method was developed and validated as per ICH [1-7] guidelines.

MATERIALS AND METHODS

Solution of sultiam was scanned in UV region and spectrum was recorded. Methanol is used as a solvent and it was seen that at 245 nm the compound has very good absorbance, which can be used for the estimation of sultiam by HPLC [8].

Based on the literature survey and method development data, The EUTICALS SPA API test method has been adopted with minor changes for estimation of Sultiam in tablet dosage form for In-House Sultiam 50mg & 200 mg tablets.

As the product is official in monograph, but there is no official method for the analysis of sultiam in tablet dosage form. So the method of analysis directly adopted from drug master file[11,12](DMF) of Sultiam with minor changes [9].

Preparation of Mobile phase

Mix water and methanol 70:30 ratio and filter through 0.45µm nylon membrane filter and sonicate for 5 minutes.

Diluent preparation:

Prepare the degassed mixture of water and methanol (70:30 v/v). Weigh accurately 30 mg of Sultiam working standard and transfered into a 100 mL volumetric flask. Add about 50 mL of diluent to dissolve it completely (sonicate if necessary) and dilute up to the mark with diluents [10].

Sample solution Preparation

20 tablets of Sultiam Weighed and crushed. Weight accurately powder equivalent to 30 mg of Sultiam, transfered into a 100mL volumetric flask and add about 50 mL of diluents to dissolve it completely and sonicate for 20 minutes with intermediate shaking, make up the volume with diluent and filtered through 0.45µm membrane filter.

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Validation of Developed Method and Forced Degradation Studies of Sultiam Tablets By RP-HPLC

The validation describes the procedure for validation of assay method of sultiam 50 mg & 200 mg tablets by HPLC as per ICH Guidelines. The analytical procedure shall be validated for the following parameters. System suitability, Specificity, Linearity, Range , Accuracy, Precision, Repeatability, Intermediate Precision, Robustness

RESULTS AND DISCUSSION

Results for Analytical Method development:

Wavelength Fixation

Solution of sultiam was scanned in UV region and spectrum was recorded. Methanol is used as a solvent and it was seen that at 245 nm, the compound has very good absorbance, which can be used for the estimation of sultiam by HPLC method.

Method Development Trials

Trial-III(Optimized)

The main peak was eluted at 5.94 min with the composition of water : methanol(70:30 v/v). The peak shape ,tailing factor and theoretical plates of sultiam peak was found to be satisfactory. The tailing factor of sultiam peak was found to be 1.41. The theoretical plates for sultiam peak was found to be 7211. In trial-II Sultiam peak was eluted at 4.967, but the number of theoretical plates are found to be less as compared to trial-III. Hence, Trial-III is considered as optimized condition.

SYSTEM SUITABILITY RESULTS

The % RSD of area of Sultiam in replicate injections of standard solution should not be more than 2.0. The tailing factor of Sultiam peak should not be more than 2.0. The theoretical plates of Sultiam peak should be more than 2000. The above results reveal that the system is suitable for analysis.

LINEARITY RESULTS

The linearity response of the Sultiam was determined and found to be linear at the concentration levels shown in the following table and was found to be meeting the acceptance criteria.

The Correlation Coefficient for Sultiam is 1**.** From this study, it is found that the method is linear.

RANGE RESULTS

HPLC Chromatogram of Sultiam Range

The % RSD for the individual recoveries of each level and mean recovery are not more than 2.0 %.The % recovery at each level and mean recovery are between 98.0% to 102.0%. This study concludes that the method is accurate in the range of 25% to 150% of working concentration.

Accuracy

The % RSD for the individual recoveries of each level and mean recovery are not more than 2.0 %. The % recovery at each level and mean recovery are between 98.0% to 102.0%.

Repeatability (Method Precision)

The % RSD for the assay of Sultiam for six replicate samples should be less than 2.0 %.

The % RSD of Assay from six test preparations for 50mg and 200mg is 0.806 and 0.846. The study concludes that the test results obtained by this method are repeatable and the method is found to be precise.

Results For Robustness

The method remains unaffected due to deliberate changes to the analytical method. The assay value shall not differ from the initial value by more than 2.0 %. The assay similarity factor should be between 0.98 to 1.02.

The % Assay differs by 1.10%&1.00% from initial value after 24hours and 48 hrs respectively. The similarity factor from the initial value after 24hours & 48hours is 1.00 and 1.00. It is concluded from the above result that the test and standard solutions are stable at room temperature upto 48hours. For sample solution The assay value shall not differ from the centrifuged sample to filtered samples by more than 2.0 %. The similarity factor should be between 0.98 to 1.02.

Table 1. Results for Trial-III

Name	Retention time(min)	Area percent	Theoretical plates (USP)	USP Tailing
Sultiam	5.943	100	7211	1.33

Table 2. Results of Chromatogram of System suitability

Parameter	Sultiam
Tailing Factor	1.33
%RSD of area	0.041
Theoretical plates	7211
Retention Time	5.943

Table 3. Different levels of Linearity solutions and Areas

Linearity Level	Volume of Stock solution (mL)	Final dilution (mL)	Conc. (in ppm)	Response (mean area)
Level 1 (25%)	2.5	50	75	1618205
Level 2 (50%)	5.0	50	150	3203326
Level 3 (75%)	7.5	50	225	4822620
Level 4 (100%)	10.0	50	300	6394527
Level 5 (125%)	12.5	50	375	8053488
Level 6 (150%)	15.0	50	450	9674189
Y – intercept				21742.64
Slope				-12515.41
Correlation Coefficient				1

Table 4. Recovery data Results(25%)

Recovery level I (25%)			
Analysis no.	**Quantity added in ppm**	**Quantity recovered in ppm**	**% Recovery**
1	74.62	73.50	98.5
2	74.42	74.05	99.5
3	74.47	74.05	99.4
		Mean	**99.1**
		SD	**0.551**
		% RSD	**0.56**

Table 5. Recovery data Results(50%)

Recovery level II (50%)			
Analysis no.	**Quantity added in ppm**	**Quantity recovered in ppm**	**% Recovery**
1	149.49	148.40	99.3
2	149.24	148.49	99.5
3	148.97	148.57	99.7
		Mean	**99.5**
		SD	**0.200**
		% RSD	**0.20**

Table 6. Recovery data Results(100%)

Recovery level III (100%)			
Analysis no.	**Quantity added in ppm**	**Quantity recovered in ppm**	**% Recovery**
1	305.72	300.41	98.3
2	304.44	301.49	99.0
3	301.56	301.25	99.0
		Mean	**98.8**
		SD	**0.404**
		% RSD	**0.41**

Table 7. Recovery data Results (150%)

Recovery level IV (150%)			
Analysis no.	**Quantity added in ppm**	**Quantity recovered in ppm**	**% Recovery**
1	446.05	443.67	99.5
2	447.54	444.35	99.3
3	448.13	444.41	99.2
		Mean	**99.3**
		SD	**0.153**
		% RSD	**0.15**

HPLC Chromatogram of Sultiam Range

Table 8. Summary of Recovery data Results

Recovery Level	% Mean Recovery
Recovery level -25%	99.1
Recovery level -50%	99.5
Recovery level -100%	98.8
Recovery level-150%	99.3
Mean Recovery	**99.2**
SD	**0.299**
% RSD	**0.30**

Table 9. Repeatability (Analyst-I)

Sample. No	% Assay For 50mg	% Assay For 200mg
Sample Preparation-1	100.5	98.9
Sample Preparation-2	101.3	98.8
Sample Preparation-3	102.4	99.4
Sample Preparation-4	102.5	98.4
Sample Preparation-5	102.1	100.7
Sample Preparation-6	101.0	99.9
Avg	**101.6**	**99.4**
SD	**0.819**	**0.841**
%RSD	**0.806**	**0.846**

Table 10.Changse in mobile phase flow rate

Robustness Criteria	%RSD	Tailing factor	Theoretical plates
Change in mobile phase flow rate by - 0.2ml/ minutes(Flow rate =0.8 ml/minutes)	0.028	1.38	6586
Actual mobile phase flow rate 1ml/minutes	0.107	1.33	7211
Change in mobile phase flow rate by +0.2ml / minutes(Flow rate =1.2 ml/minutes)	0.017	1.34	6000

Table 11. Changes in wavelength

Robustness Criteria	%RSD	Tailing factor	Theoretical plates
Change in wavelength by-2nm (243nm)	0.199	1.36	6390
Actual wavelength 245nm	0.107	1.33	7211
Change in wavelength by+2nm(247)	0.025	1.35	6413

Table 12. Changes in column temperature

Robustness Criteria	%RSD	Tailing factor	Theoretical plates
Change in column temperature by -5°C (20°C)	0.033	1.34	6275
Actual column temperature (25°C)	0.107	1.33	7211
Change in column temperature by +5°C (30°C)	0.067	1.37	6458

Table 13. Changes in mobile phase organic phase

Robustness Criteria	%RSD	Tailing factor	Theoretical plates
Change in mobile phase organic phase by -10% (270ml)	0.029	1.36	6889
Actual organic phase (300ml)	0.107	1.33	7211
Change in mobile phase organic phase by -10% (330ml)	0.040	1.37	5796

Table 14. Solution stability -sample preparations

Preparation	Sample	
	% Assay	Difference
Initial	98.9	NA
After 24 hours	100.0	1.10
After 48 hours	99.9	1.00

Table 15. Solution stability -standard preparations

Preparation	Standard	
	Standard area	Similarity factor
Initial	6502984	NA
After 24 hours	Fresh std:6529379 24hrs sts: 6529085	1.00
After 48 hours	Fresh std: 6519200 48hrs sts: 6517947	1.00

Table 16. Filter study data results for sample

Sample No.	% Assay for Sultiam	Difference
Centrifuged	99.9	NA
0.45µ GHP filtered	99.9	0.0
0.45µ Nylon filtered	99.8	0.1

Table 17. Filter study data results for Standard

Sample No.	Standard area for Sultiam	Similarity factor
Centrifuged	6502984	NA
0.45µ GHP filtered	6518601	1.00
0.45µ Nylon filtered	6512257	1.00

Fig 1. HPLC Chromatogram of system suitability

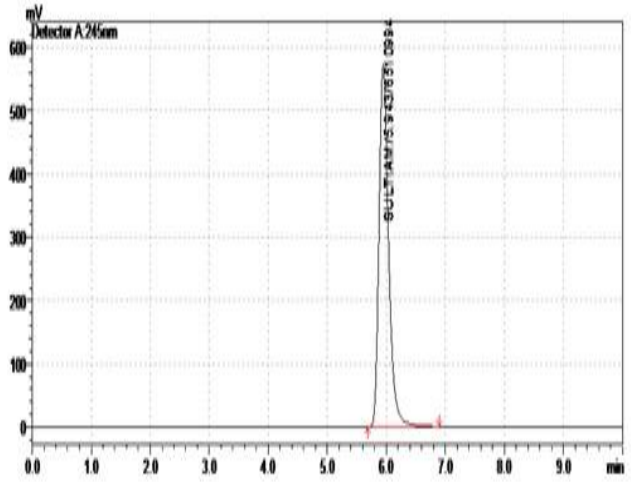


Fig 2. HPLC Chromatogram of Sultiam 243 nm

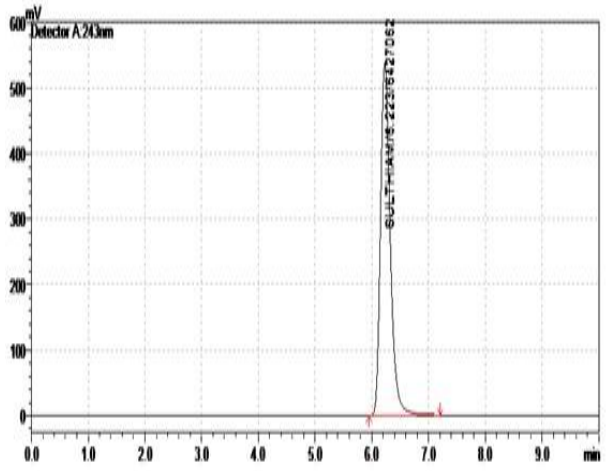


Fig 3. HPLC Chromatogram of Sultiam 247 nm

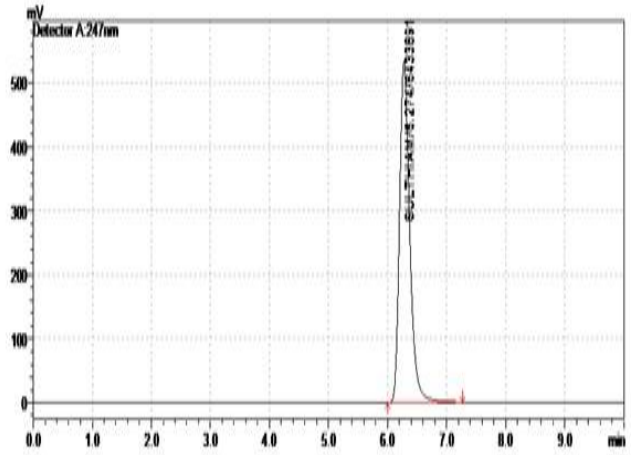


Fig 4. HPLC Chromatogram of Sultiam Low column oven temperature

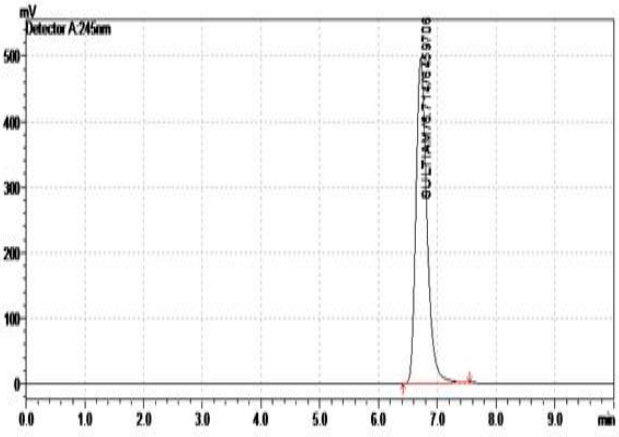
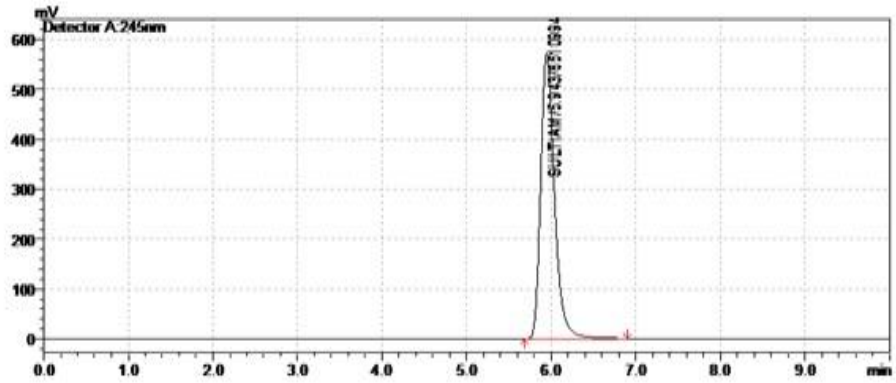


Fig 5. HPLC Chromatogram of Sultiam High column oven temperature



SUMMARY AND CONCLUSION

The determination of Sultiam in tablet dosage forms using chromatographic methods with mobile phase as Water and Methanol in the ratio 70:30 respectively. Literature survey reveals that there are no specific RP-HPLC methods available for the determination of Sultiam in tablet dosage forms. Henceforth we planned to develop a precise, accurate, less time consuming and with low solvent cost RP-HPLC method, was developed and validated as per the ICH guidelines. The assay method for Sultiam in tablet dosage form was developed and validated as per ICH guidelines**.** The results were found to be within the acceptance limit.

The validated method was found to be simple, specific, precise, accurate, Robust and Rugged for the estimation of Sultiam in tablet dosage form. Hence it is concluded that the assay method is found to be valid in terms of reliability, precision, accuracy, suitable for chemist-to-chemist and day-to-day for routine analysis as well as for stability analysis.

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Nil

CONFLICT OF INTEREST

No interest

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