



International Journal of Chemistry and Pharmaceutical Sciences

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Research Article

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Analytical Method Development and Validation for Simultaneous Estimation of Asparin and Atrovastatin Calcium in Combind Dosage form by using UV-Spectrophotometry

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ABSTRACT

A simple, Accurate, precise method was developed for the simultaneous estimation of the Aspirin and Atrovastatin calcium in Tablet dosage form. Solutions of Aspirin and Atrovastatin Calcium were prepared in different solvents like methanol; ethanol, acetonitrile and UV spectrum of each were recorded by scanning between 200-400 nm. Aspirin was found to be linear in a concentration range of 10-100µg/ml. The absorbances of these solutions were noted at wavelengths 275 and 247 nm, respectively. Calibration curves were plotted using concentration Vs absorbance at wavelength of 275 nm and the slope, intercept and correlation coefficient values were found to be 0.008, 0.007 and 0.999 respectively, Atrovastatin was found to be linear at a concentration range of 1-10µg/ml. The absorbances of these solutions were noted at 247 and 275 nm, respectively. Calibration curves were plotted using concentration Vs absorbance. At a wavelength of 247 nm, the slope, intercept and correlation coefficient values were found to be 0.001, 0.046 and 0.999, respectively. At wavelength 275 nm, the slope, intercept and correlation coefficient values were found to be 0.006, 0.005 and 0.999, respectively.

Keywords: Aspirin, Atrovastatin, UV-Spectrophotometry

ARTICLE INFO

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Article History: Received 10 October 2015, Accepted 19 November 2015, Available Online 27 December 2015

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Manuscript ID: IJCPS2789



PAPER-QR CODE

Citation: V. Aparna, et al. Analytical Method Development and Validation for Simultaneous of Asparin and Atrovastatin Calcium in Combind Dosage Form by Using UV- Spectrophotometry. *Int. J. Chem, Pharm, Sci.*, 2015, 3(12): 2081-2089.

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1. Introduction

Aspirin is chemically named as (3R, 5S, 6E)-7-[4-(4-fluorophenyl)-2-(N-methylmethanesulfonamido)-6-(propan-2-yl) pyrimidin-5-yl]-3, 5-dihydroxyhept-6-enoic acid [1]. Rosuvastatin is a competitive inhibitor of the enzyme HMG-CoA reductase, having a mechanism of action similar to that of other statins. Its approximate elimination half life is 19 h and its time to peak plasma concentration is reached in 3–5 h following oral administration [2]. Putative beneficial effects of rosuvastatin therapy on chronic heart failure may be negated by increases in collagen turnover markers as well as a reduction in plasma coenzyme Q10 levels in patients with chronic heart failure [3].

Atorvastatin is chemically named 7-[2-(4-fluorophenyl)-3-phenyl-4-(phenylcarbamoyl)-5-(propan-2-yl)-1H-pyrrol-1-yl]-3,5-dihydroxyheptanoate. Atorvastatin selectively and competitively inhibits the hepatic enzyme HMG-CoA reductase. As HMG-CoA reductase is responsible for converting HMG-CoA to mevalonate in the cholesterol biosynthesis pathway, these results in a subsequent decrease in hepatic cholesterol levels [4]. Decreased hepatic cholesterol levels stimulates upregulation of hepatic LDL-C receptors which increases hepatic uptake of LDL-C and reduces serum LDL-C concentrations [5].

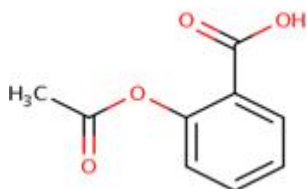


Figure 1: Structure of Aspirin

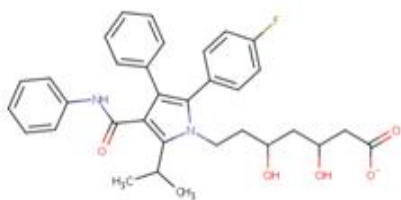


Figure 2: Structure of Atorvastatin

2. Materials and Methods

Preparation of standard solution:

10 mg of Aspirin and 10mg of Atorvastatin were accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicate to dissolve it completely and the volume was made up to the mark with the same solvent to give a concentration of 1000 µg/ml. (Stock solution) Further 0.2 and 0.1 ml were pipette out from the above stock solution into a 10ml volumetric flask and diluted up to the mark with diluent to give a concentration of 20 µg/ml and 10µg/ml respectively[7].

Preparation of sample solution:

10 Tablets of contents were weighed and triturated in glass mortar. The quantity of powder equivalent to 10 mg of active ingredient present in Aspirin and Atorvastatin was

transferred into a 10 ml clean dry volumetric flask, 7 ml of diluent was added to it and was shaken by mechanical stirrer and sonicate for about 30 minutes by shaking at intervals of five minutes each and was diluted up to the mark with diluent to give a concentration of 1000 µg/ml and allowed to stand until the residue settles before taking an aliquot for further dilution (stock solution)[8]. 0.2 ml of upper clear solution was transferred to a 10 ml volumetric flask and diluted with diluent up to the mark to give the respective concentrations as par with standard solution. The solution was filtered through 0.45 µm filter before injecting into HPLC system.

Preparation of Placebo:

The amount of powdered inactive ingredient supposed to be present in 10 tablets were accurately weighed and transferred in to 10 ml volumetric flask, 7 ml of diluent was added and shaken by mechanical stirrer and sonicated for about 30 minutes by shaking at intervals of five minutes and was diluted up to the mark with diluent and allowed to stand until the residue settles before taking an aliquot for dilution. 0.1 ml of upper clear solution was transferred to a 10 ml volumetric flask and diluted with diluent up to the mark and the solution was filtered through 0.45 µm filter before injecting into HPLC system.

Linearity:

Preparation of sample stock solution:

About 20 mg of Aspirin and 10 mg of Atorvastatin samples was weighed in to 10ml volumetric flask, it was dissolved with diluent and the volume was made up to the mark with same diluent (2000µg/ml of Aspirin and 1000µg/ml of Atorvastatin)[12].

Preparation of Level – I (20µg/ml of Aspirin &10µg/ml of Atorvastatin)

0.1ml of stock solution had taken in 10ml of volumetric flask diluted up to the mark with diluent.

Preparation of Level–II (40 µg/ml of Aspirin &15.0 µg/ml of Atorvastatin)

0.2ml of stock solution had taken in 10ml of volumetric flask diluted up to the mark with diluent.

Preparation of Level–III (60 µg/ml of Aspirin &20 µg/ml of Atorvastatin)

0.3ml of stock solution had taken in 10ml of volumetric flask diluted up to the mark with diluent.

Preparation of Level–IV (80 µg/ml of Aspirin &25 µg/ml of Atorvastatin)

0.4ml of stock solution had taken in 10ml of volumetric flask diluted up to the mark with diluent.

Preparation of Level–V (100 µg/ml of Aspirin &30 µg/ml of Atorvastatin).

0.5ml of stock solution had taken in 10ml of volumetric flask diluted up to the mark with diluent.

10µl of each level were injected into the system and recorded the peak response.

Procedure:

Each level solution was injected into the chromatographic system and the peak area was measured. A graph of peak area versus concentration (on X-axis concentration and on Y-axis Peak area) was plotted and the correlation

coefficient was calculated. The linearity of the method was demonstrated over the concentration range of 10-100 µg / ml. Aliquots of five levels were prepared from sample solution and labeled as solution 1, 2, 3, 4 and 5 respectively.

Acceptance criteria

1. Correlation Coefficient should be not less than 0.9990.
2. % RSD of peak areas for Solution 1, 2, 3, 4 and 5 should be not more than 2.0 %.

Precision

Preparation of stock solution:

10 mg of Aspirin and 10mg of Atorvastatin were accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added and sonicate to dissolve it completely. The volume was made up to the mark with the same solvent to give the concentration of 1000 µg/ml. (Stock solution) .Further 0.2 ml and 0.2 ml was pipette out from the above stock solutions into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 20 µg/ml and 20 µg/ml respectively.

Procedure:

The standard solution was injected for five times and the areas for all five injections were measured in HPLC. The %RSD for the area of five replicate injections was found to be within the specified limits

Acceptance criteria

% Relative standard deviation of peak areas and R_t should not be more than 2.0 %.

Intermediate Precision/Ruggedness

To evaluate the intermediate precision (also known as Ruggedness) of the method Precision was performed on different day by using different make column of same Dimensions.

Preparation of stock solution:

10 mg of Aspirin and 10mg of Atorvastatin accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added ,sonicate to dissolve it completely and the volume was made up to the mark with the same solvent to give the concentration of 1000 µg/ml. (Stock solution) Further 0.2ml and 0.1ml were pipette out from the above stock solution into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 20µg/ml and 10 µg/ml respectively[13].

Procedure:

The standard solution was injected for five times and the area was measured for all five injections in HPLC. The %RSD for the area and R_t of five replicate injections was found to be within the specified limits.

Acceptance criteria: % Relative standard deviation of peak areas and R_t should not be more than 2.0 %

Accuracy

Assay was performed in triplicate for various concentrations of Aspirin and Atorvastatin equivalent to 50, 100, and 150 % of the standard amount was injected into the HPLC system as per the test procedure.

Preparation of Standard stock solution:

10 mg of Aspirin and 10mg of Atorvastatin accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicate to dissolve

it completely and volume was made up to the mark with the same solvent to give the concentration of 1000 µg/ml. (Stock solution)

Preparation Sample solutions:

Preparation of 50% solution (15 µg/ml of Aspirin and 90µg/ml of Atorvastatin):

From the above stock solutions take 0.15 ml and 0.9 ml into 10 ml dry volumetric flask, make up to the mark with diluent.

Preparation of 100% solution (30 µg/ml of Aspirin and 180 µg/ml of Atorvastatin):

From the above stock solutions take 0.3 ml and 1.8 ml into 10 ml dry volumetric flask, make up to the mark with diluent

Preparation of 150% solution (45 µg/ml of Aspirin and 270 µg/ml of Atorvastatin):

From the above stock solutions take 0.45 ml and 2.7 ml into 10 ml dry volumetric flask, make up to the mark with diluent. These solutions were filtered through 0.45µ membrane and then each concentration; three replicate injections were made under the optimized conditions

Procedure:

The standard solution, Accuracy -50%, Accuracy -100% and Accuracy -150% solutions were injected. The amount found and amount added for Aspirin and Atorvastatin individual recovery and mean recovery values were calculated and the results were found to be within limits.

Acceptance criteria

The mean % recovery of the Aspirin and Atorvastatin each spike level should be not less than 98.0 % and not more than 102.0 %.

Specificity

A) Aspirin and Atorvastatin identification:

Solutions of Standard and Sample were prepared as per test procedure and injected into the HPLC system.

Acceptance criteria

Chromatogram of standard and sample should be identical with near Retention time.

B) Placebo interference:

A study to establish the interference of placebo was conducted. A sample of placebo was injected into the HPLC system as per the test procedure.

Acceptance criteria

Chromatogram of placebo should not show any peak at the retention time of analyte peak. There is no interference due to placebo at the retention time of analyte. Hence the method is specific.

C) Blank interference:

A study to establish the interference of blank was conducted. Diluent was injected into HPLC system as per the test procedure.

Acceptance criteria

Chromatogram of blank should not show any peak at the retention time of analyte peak. There is no interference due to blank at the retention time of analyte. Hence the method is specific.

Ruggedness

The simultaneous estimation of Aspirin and Atorvastatin was performed by different analysts on different days.

Acceptance criteria

% Relative standard deviation of peak areas and R_f should not be more than 2.0 %.

Robustness

The robustness of the proposed method was determined by analysis of aliquots from homogenous lots by differing physical parameters like flow rate and mobile phase composition, temperature variations which may differ but the responses were still within the specified limits of the assay[14].

a) Effect of variation of flow rate:

A study was conducted to determine the effect of variation in flow rate. The flow rate was varied at 0.6 ml/min to 1.0 ml/min. Standard solution 20 ppm Aspirin and 10 ppm Atorvastatin were prepared and analysed using the varied flow rates along with method flow rate.

The results are summarized on evaluation of the above results, it can be concluded that the variation in flow rate affected the method significantly. Hence it indicates that the method is robust even by change in the flow rate $\pm 10\%$. The method is robust only in less flow condition. The effect of variation of flow rate was evaluated.

b) Effect of variation of mobile phase composition:

A study was conducted to determine the effect of variation in mobile phase ratio by changing the ratio of mobile phase. The Organic composition in the Mobile phase was varied from 30 % to 70 %.

Standard solution 20 $\mu\text{g/ml}$ of Aspirin and 10 $\mu\text{g/ml}$ Atorvastatin were prepared and analysed using the varied mobile phase composition along with the actual mobile phase composition in the method. Standard solution was prepared and injected into the HPLC system and the Chromatograms which were recorded.

Limit of Detection

For Aspirin:

Preparation of 10 $\mu\text{g/ml}$ solution:

10mg of Aspirin was accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicated to dissolve it completely and volume was made up to the mark with the same solvent to give the concentration of 1000 $\mu\text{g/ml}$. (Stock solution). Further 0.1ml of the above stock solution was pipetted out into a 10ml volumetric flask and dilute up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$.

Preparation of 0.15% solution at Specification level (0.004 $\mu\text{g/ml}$ solution):

Further 1ml of the above stock solution was pipette into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$. 0.1 ml of 10 $\mu\text{g/ml}$ solution was pipette into a 10 ml of volumetric flask and diluted up to the mark with diluent give the concentration of 0.004 $\mu\text{g/ml}$ [15].

For Atorvastatin

Preparation of 10 $\mu\text{g/ml}$ solution:

10mg of Atorvastatin working standard was accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicate to dissolve it completely and the volume was made up to the mark with the same solvent to give the concentration of 1000 $\mu\text{g/ml}$. (Stock solution)

Further 0.1 ml of the above stock solution was pipette into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$.

Preparation of 0.22% solution At Specification level (0.006 $\mu\text{g/ml}$ solution):

Further 1ml of the above stock solution was pipette into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$. 0.22 ml of 10 $\mu\text{g/ml}$ solution was pipette into a 10 ml of volumetric flask and diluted up to the mark with diluent to give the concentration of 0.006 $\mu\text{g/ml}$ [16].

LOD sample:

From the 10 $\mu\text{g/ml}$ of Aspirin and 10 $\mu\text{g/ml}$ of Atorvastatin of the individual stock solution we prepared 0.004 $\mu\text{g/ml}$ and 0.006 $\mu\text{g/ml}$ injected into the system.

The LOD is determined by the formula

$$\text{LOD} = \text{S/N}$$

Where

N = Average Baseline Noise obtained from Blank

S = Signal Obtained from LOD solution (0.25% of target assay concentration).

Acceptance Criteria: S/N Ratio value shall be not more than 3 for LOD solution.

Limit of Quantification (LOQ)

For Aspirin:

Preparation of 10 $\mu\text{g/ml}$ solution:

10 mg of Aspirin was accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicate to dissolve it completely and make volume up to the mark with the same solvent to give the concentration of 1000 $\mu\text{g/ml}$. (Stock solution). Further 0.1ml of the above stock solution was pipette into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$ [17].

Preparation of 0.05% solution At Specification level (0.015 $\mu\text{g/ml}$ solution):

Further 1ml of the above stock solution was pipette into a 10ml volumetric flask and dilute up to the mark with diluent to give the concentration of 1 $\mu\text{g/ml}$.

0.05 ml of 10 $\mu\text{g/ml}$ solution was pipetted into a 10 ml of volumetric flask and diluted up to the mark with diluent to give the concentration of 0.015 $\mu\text{g/ml}$.

For Atorvastatin:

Preparation of 10 $\mu\text{g/ml}$ solution:

10mg of Atorvastatin was accurately weighed and transferred into a 10 ml clean dry volumetric flask, about 7 ml of diluent was added, sonicated to dissolve it completely and make volume up to the mark with the same solvent to give the concentration of 1000 $\mu\text{g/ml}$ [18]. (Stock solution).

Further 0.1ml of the above stock solution was pipetted into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$.

Preparation of 0.06% solution At Specification level (0.02 $\mu\text{g/ml}$ solution):

Further 1ml of the above stock solution was pipetted into a 10ml volumetric flask and diluted up to the mark with diluent to give the concentration of 10 $\mu\text{g/ml}$. 0.06 ml of 10 $\mu\text{g/ml}$ solution was pipetted into a 10 ml of volumetric flask and diluted up to the mark with diluent to give the concentration of 0.02 $\mu\text{g/ml}$.

3. Results and Discussion

Selection of solvent: Solutions of Aspirin and Atorvastatin Calcium were prepared in different solvents like methanol; ethanol, acetonitrile and UV spectrum of each were recorded by scanning between 200-400 nm. An overlain spectrum of Aspirin and Atorvastatin Calcium were prepared in different solvents like methanol, water etc. Better absorbances were observed for both the drugs when water is used as a solvent. Hence, Methanol was selected as solvent for present study. From the spectra of drugs in methanol, four trial wavelengths were selected (270, 275, 247 and 236 nm) for their simultaneous estimation. Different concentrations of Aspirin (10 to 100 µg/ml), Atorvastatin Calcium (1 to 10 µg/ml) and mixture of Aspirin and Atorvastatin Calcium were prepared, scanned and absorbances were noted at these three wavelengths. From these data, it was noted that, at the wavelengths 275 and 247 nm, good linearity was observed and hence these wavelengths were fixed for the study.

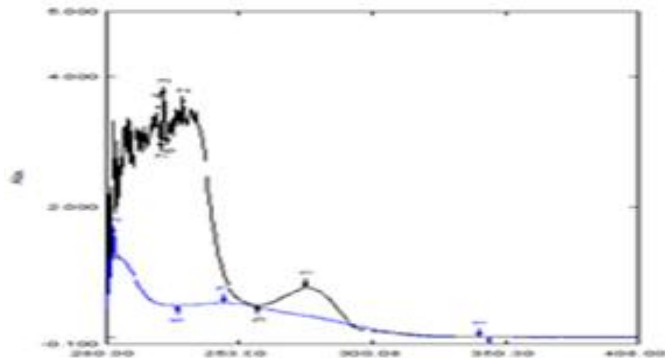


Figure 3: Overlain Normal spectra of Aspirin and Atorvastatin Calcium in Methanol

Validation of the method

Linearity (Aspirin): Aspirin was found to be linear in a concentration range of 10-100 µg/ml. The absorbances of these solutions were noted at wavelengths 275 and 247 nm, respectively. Calibration curves were plotted using concentration Vs absorbance at wavelength of 275 nm and the slope, intercept and correlation coefficient values were found to be 0.008, 0.007 and 0.999 respectively, fig. 7.1.2. At wavelength 247nm, slope, intercept and correlation coefficient values were found to be 0.007, 0.015 and 0.999, respectively.

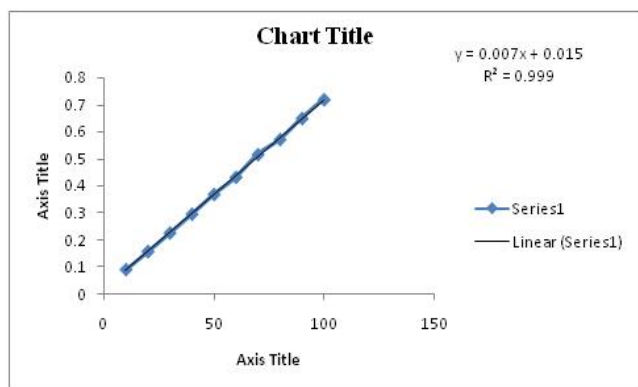


Figure 4: Calibration graph of Aspirin at 275nm

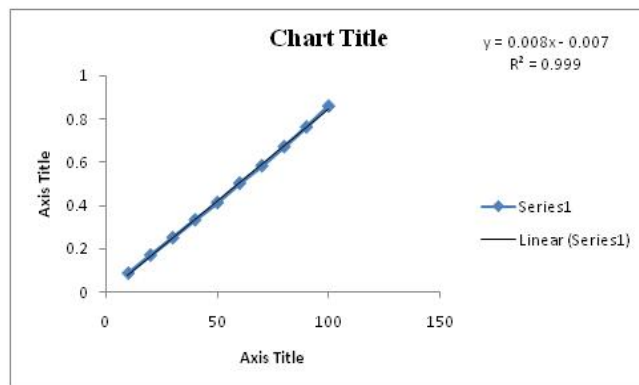


Figure 5: Calibration graph of Aspirin at 247nm

Aspirin was found to be linear at a concentration range of 10-100 µg/ml. The absorbances of these solutions were noted at 275 and 247 nm, respectively. Calibration curves were plotted using concentration Vs absorbance. At a wavelength of 247 nm, the slope, intercept and correlation coefficient values were found to be 0.008, 0.007 and 0.999, respectively, fig. 7.1.3. At wavelength 275 nm, the slope, intercept and correlation coefficient values were found to be 0.007, 0.015 and 0.999, respectively.

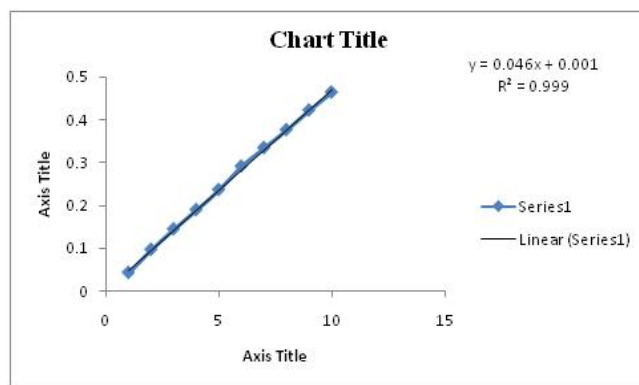


Figure 6: Calibration graph of Atorvastatin at 247nm

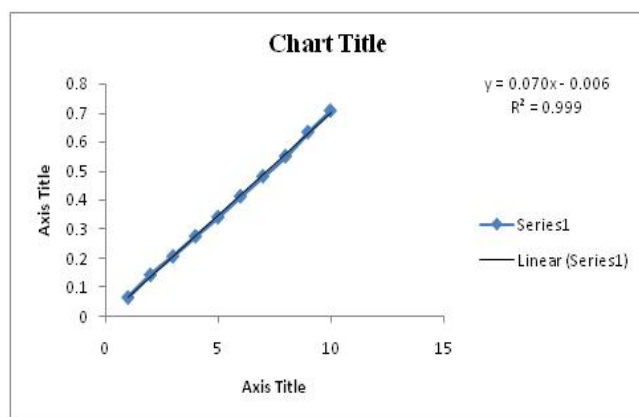


Figure 7: Calibration graph of Atorvastatin at 275nm

Atorvastatin was found to be linear at a concentration range of 1-10 µg/ml. The absorbances of these solutions were noted at 247 and 275 nm, respectively. Calibration curves were plotted using concentration Vs absorbance. At a wavelength of 247 nm, the slope, intercept and correlation

coefficient values were found to be 0.001, 0.046 and 0.999, respectively. At wavelength 275 nm, the slope, intercept and correlation coefficient values were found to be 0.006, 0.005 and 0.999, respectively.

Precision

Precision studies were performed by preparing the standards three times and measuring the absorbances of drugs at 241 nm and 288 nm. Low RSD values indicate that the method is precise.

Recovery studies

In order to ensure the suitability and reliability of proposed method, recovery studies were carried out. To an equivalent quantity of formulation powder (12mg), a known quantity of standard Aspirin Hydrochloride and Atorvastatin were added at 50% and 100% level and the contents were re-analyzed by the proposed method.

Analysis of Formulation

Preparation of standard solutions

Standard stock solutions of Aspirin Hydrochloride and Atorvastatin were prepared by dissolving 10 mg of the drug in water and the volume was made up to 100ml in a standard flask. From the stock solution, concentrations ranging from 2-20 µg/ml was prepared for both Aspirin Hydrochloride and Atorvastatin and scanned in the UV region.

Preparation of sample solution

Twenty tablets (ZENFLOX-UTI) are powdered and the average weight was calculated. A quantity equivalent to 10 mg of drug was dissolved in water. Finally the volume was made up to get a working concentration of 12µg/ml each of Aspirin Hydrochloride and Atorvastatin. Absorbances were noted at 241 nm and 288 nm, respectively.

At λ_1 $A_1 = ax_1bc_x + ay_1bc_y$

λ_2 $A_2 = ax_2bc_x + ay_2bc_y$

$$C_{fix} = \frac{A_2ay_1 - A_1ay_2}{ax_2ay_1 - ax_1ay_2}$$

$$C_{ofx} = \frac{A_1ax_2 - A_2ax_1}{ax_2ay_1 - ax_1ay_2}$$

A_1 = absorbance of formulation at 288nm.

A_2 = absorbance of formulation at 241nm.

ax_1 = Absorptivity of Aspirin Hydrochloride at 288nm.

ax_2 = Absorptivity of Aspirin Hydrochloride at 241nm.

ay_1 = Absorptivity of Atorvastatin at 288nm.

ay_2 = Absorptivity of Atorvastatin at 241nm.

C_{fix} = Concentration of Aspirin Hydrochloride.

C_{ofx} = Concentration of Atorvastatin.

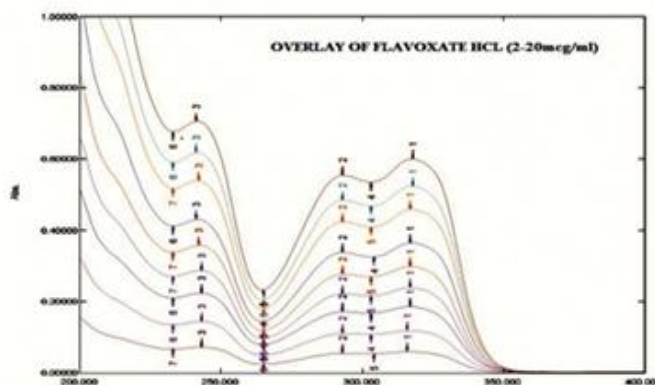


Figure 8: Overlay UV spectra of Aspirin at 288nm

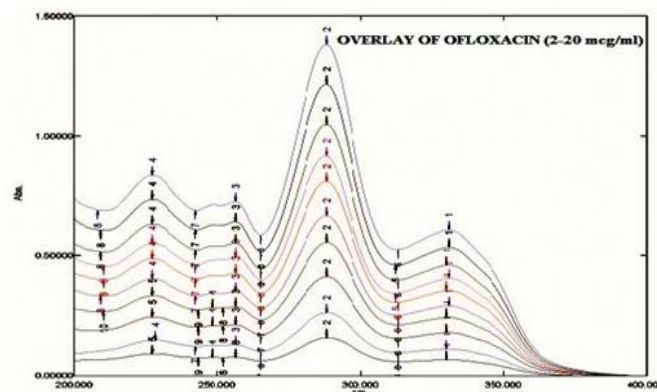


Figure 9: Overlay UV spectra of Atorvastatin at 241nm

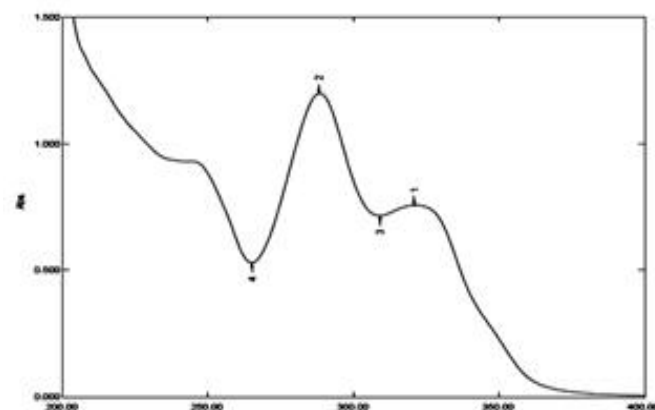


Figure 10: UV spectra of formulation (12µg/ml of Aspirin Hydrochloride and 12µg/ml of Atorvastatin)

Table 1: List of Standard and Sample details

S.no	Name	Batch No.	Manufacturer/supplier
1.	Aspirin working standard	-	KP labs pvt. Ltd
2.	Atorvastatin working standard	-	KP labs pvt. Ltd

Table 2: List of Equipment/Instrument details

S.No.	Instrument name	Model
1.	HPLC system	WATERS, software: Empower, 2695 separation module. 2996 PDA detector.
2.	Semi micro balance	Sartorius ME235P

3.	P ^H Meter	Lab India ph. Meter
4.	Sonicator	Ultrasonic cleaner power sonic 420
5.	UV/VIS spectrophotometer	LABINDIA UV
6.	Constant temperature water bath	Thermolab GMP

Table 3: List of Chemicals and Reagents

S.No	Name	Manufacturer	Grade
1.	Potassium dihydrogenorthophosphate	Merck	GR
2.	Sodium perchlorate	Merck	GR
3.	Perchloric acid	Merck	GR
4.	Ortho phosphoric acid	Merck	GR
5.	Methanol	Merck	HPLC
6.	Acetonitrile	Merck	HPLC
7.	Water	Milli-pore	Milli-Q
8.	0.45 µm Nylon filter	Axivia	S0761009
9.	0.45µm PVDF filter	Rankem	D004A07

Table 4: Calibration data of Aspirin at 275nm and 247 nm

S.No	Concentration (~g/ml)	Absorbance at 275 nm	Absorbance at 247nm
1	10	0.084	0.089
2	20	0.168	0.156
3	30	0.249	0.225
4	40	0.332	0.295
5	50	0.412	0.368
6	60	0.502	0.432
7	70	0.583	0.516
8	80	0.672	0.572
9	90	0.763	0.65
10	100	0.86	0.72

Table 5: Calibration data of Atorvastatin at 247 and 275nm

S. No	Concentration (~g/ml)	Absorbance at 247 nm	Absorbance at 275 nm
1	1	0.042	0.065
2	2	0.096	0.143
3	3	0.144	0.207
4	4	0.189	0.276
5	5	0.236	0.342
6	6	0.291	0.414
7	7	0.334	0.483
8	8	0.376	0.552
9	9	0.422	0.635
10	10	0.464	0.709

Table 6: Intraday Precision studies

concentration	Absorbances				%RSD*			
	Aspirin		Atorvastatin		Aspirin		Atorvastatin	
	247 nm	275 nm	247 nm	275 nm	247 nm	275 nm	247 nm	275nm
10µg/ml	0.433	0.328	0.351	0.676	0.23	0.17	0.16	0.08
	0.434	0.329	0.351	0.677				
	0.432	0.328	0.352	0.676				
12µg/ml	0.515	0.389	0.428	0.815	0.11	0.14	0.13	0.07
	0.515	0.388	0.428	0.814				
	0.516	0.389	0.429	0.815				

Table 7: Inter day Precision studies

concentration	Absorbance				%RSD*			
	Aspirin		Atorvastatin		Aspirin		Atorvastatin	
	247nm	275 nm	247 nm	275 nm	247 nm	275 nm	247 nm	275 nm
10µg/ml	0.433	0.328	0.351	0.676	0.35	1.41	0.16	0.392
	0.430	0.328	0.350	0.672				
	0.432	0.320	0.350	0.677				
12µg/ml	1.877	0.059	0.038	0.133	1.01	0.91	0.90	0.75
	1.881	0.057	0.041	0.134				
	1.889	0.053	0.043	0.130				

Table 8: Recovery studies

Level	% Recovery		%RSD	
	Aspirin	Atorvastatin	Aspirin	Atorvastatin
50%	98.33	98.5	0.1	0.27
100%	98.41	99.25	0.39	0.23

*RSD of three observations

Table 9: Analysis of formulation

Drug	Amount (mg/tab)		% Label Claim	% RSD*
	Labeled	Found		
Aspirin	200	199.8	99.90	0.53
Atorvastatin	200	201.0	100.51	0.95

*RSD of three observations

4. Conclusion

A simple, Accurate, precise method was developed for the simultaneous estimation of the Aspirin and Atorvastatin in Tablet dosage form. Optimized wavelength for Aspirin and Atorvastatin was 254nm. To an equivalent quantity of formulation powder (12mg), a known quantity of standard Aspirin Hydrochloride and Atorvastatin were added at 50% and 100% level and the contents were re-analysed by the proposed method. The % recovery of 50% for Aspirin was found to be 98.33 and Atorvastatin was found to be 98.5. The % recovery of 100% for Aspirin was found to be 98.41 and Atorvastatin was found to be 99.25. %RSD were calculated for 50% for Aspirin was found to be 0.1 and Atorvastatin was found to be 0.27. %RSD were calculated for 100% for Aspirin was found to be 0.39 and Atorvastatin was found to be 0.23. LOD, LOQ values are obtained from Aspirin and Atorvastatin within the limits respectively.

5. References

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