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## Development and Validation of Analytical Methods for Simultaneous Estimation of Rosuvastatin, Clopidogrel and Aspirin in Pharmaceutical Dosage Form

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### ABSTRACT:

A newer, simple, accurate and sensitive Simultaneous Equation method is developed for the simultaneous estimation of Rosuvastatin (ROS), Clopidogrel (CLOP) and Aspirin (ASP) in pharmaceutical dosage form. Simultaneous Equation method was developed using three wavelengths which are 243.56 nm ( $\lambda_{max}$  of ROS), 223.38 nm ( $\lambda_{max}$  of CLOP) and 276.44 nm ( $\lambda_{max}$  of ASP). In Simultaneous Equation method ROS, CLOP and ASP obeyed Beer's law in the concentration range of 1-5  $\mu\text{g/ml}$  for ROS, 7.5-37.5  $\mu\text{g/ml}$  for CLOP and 7.5-37.5  $\mu\text{g/ml}$  for ASP. Methanol was used as a solvent. The results of the analysis were analyzed and validated statistically and recovery studies were carried out as per ICH guidelines. It can be used for routine analysis of three drugs in bulk as well as in pharmaceutical formulations.

**KEY WORDS:** Rosuvastatin (ROS), Clopidogrel (CLOP), Aspirin (ASP), Simultaneous Equation Method.

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### INTRODUCTION:

Rosuvastatin belong to a class called statin. It is a selective and competitive inhibitor of HMG-CoA reductase and used for the treatment of dyslipidemia. It is chemically (E)-(3R,5S)-7-{4-(4-fluorophenyl)6-isopropyl-2{methyl (methyl sulphonylamino)}pyrimidin5-yl}-3,5-dihydroxyhepten-6-oic acid. Clopidogrel is a thienopyridine class antiplatelet agent. Chemically it is methyl (S)-a-(o-chlorophenyl)-6,7-dihydrothieno[3,2-c] pyridine-5-(4H)-acetate. Aspirin is a salicylate drug. It is non-selective cyclo-oxygenase inhibitor. Chemically it is 2-(acetyloxy)benzoic acid. Clinically a combination is being used in the treatment of Acute coronary syndrome, Myocardial infraction, Stroke and Angina for better therapeutic effect.

The combination of Rosuvastatin, Clopidogrel and Aspirin is not official in any official pharmacopoeia. A literature survey revealed that no analytical methods were reported for the simultaneous estimation of Rosuvastatin, Clopidogrel and Aspirin in pharmaceutical dosage form Hence in the present study a physical mixture of Rosuvastatin, Clopidogrel and Aspirin was being taken for simultaneous estimation by UV method. This present investigation describes a rapid, accurate and precise UV method of Rosuvastatin, Clopidogrel and Aspirin in combination using Methanol as a solvent. In which three wavelengths are used 243.56 nm ( $\lambda_{max}$  of ROS), 223.38 nm ( $\lambda_{max}$  of CLOP) and 276.44 nm ( $\lambda_{max}$  of ASP).

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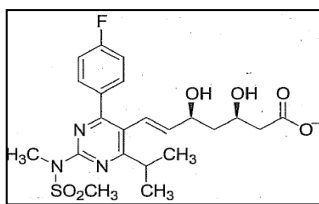


Figure: 1 Structure of Rosuvastatin

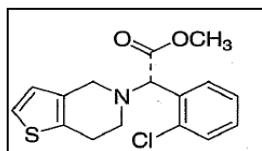


Figure: 2 Structure of Clopidogrel

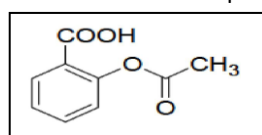


Figure: 3 Structure of Aspirin

## MATERIALS AND METHODS:

### Instruments and Apparatus:

Instrument used was an UV-Visible double beam spectrophotometer, SHIMADZU (model UV-1800, software – UV probe, version 2.42) with a pair of 1 cm matched quartz cells. All weighing was done on Reptech electronic analytical balance. All the apparatus used were calibrated.

### Reagents and Chemicals:

Rosuvastatin (Gift sample, Sun Pharma, Silvassa.), Clopidogrel (Gift sample, Aarti Drugs Ltd., Vapi.) and Aspirin (Gift sample, Sidmak Laboratories Pvt., Ltd., Valsad). Methanol were used as a solvents.

Marketed Formulation: Combined Tablet Formulation was purchased from Local market.

### Preparation of Standard solution:

Rosuvastatin (ROS) standard stock solution: (1000 µg/ml)  
100 mg of ROS standard was weighed and transferred to a 100 ml volumetric flask. The drug was dissolved by adding 25 ml of methanol and volume was made up to the mark with methanol to give a solution containing 1000 µg/ml ROS. From this solution 10 ml was transfer to 100 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 100

µg/ml ROS. From this solution 25 ml was transfer to 50 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 50 µg/ml ROS.

Clopidogrel (CLOP) standard stock solution: (1000 µg/ml)

100 mg of CLOP standard was weighed and transferred to a 100 ml volumetric flask. The drug was dissolved by adding 25 ml of methanol and volume was made up to the mark with methanol to give a solution containing 1000 µg/ml ROS. From this solution 10 ml was transfer to 100 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 100 µg/ml CLOP. From this solution 25 ml was transfer to 50 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 50 µg/ml CLOP.

Aspirin (ASP) standard stock solution: (1000 µg/ml)

100 mg of ASP standard was weighed and transferred to a 100 ml volumetric flask. The drug was dissolved by adding 25 ml of methanol and volume was made up to the mark with methanol to give a solution containing 1000 µg/ml ROS. From this solution 10 ml was transfer to 100 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 100 µg/ml ASP. From this solution 25 ml was transfer to 50 ml volumetric flask. The volume was adjusted to the mark with the methanol to give a solution containing 50 µg/ml ASP.

### Selection of Analytical Wavelength

1-5µg/ml solutions of ROS were prepared in Methanol and spectrum was recorded between 200-400 nm. Spectrums for above concentration were obtained with n=5. Similarly 7.5-37.5 µg/ml solutions of CLOP were prepared in Methanol and spectrum was recorded between 200-400nm and 7.5-37.5 µg/ml solutions of CLOP were prepared in Methanol and spectrum was recorded between 200-400nm. ROS showed  $\lambda_{max}$  at wavelength 243.56 nm, CLOP showed  $\lambda_{max}$  at wavelength at 223.38 nm and ASP CLOP showed  $\lambda_{max}$  at wavelength at 276.44 nm.

The overlain spectrums of ROS, CLOP and ASP at different concentration were recorded

### Method:

Calibration curve for the ROS (1 - 5 µg/ml)

Appropriate volume of aliquot from standard ROS stock solution was transferred to different volumetric flasks of 10 ml capacity. The volume was adjusted to the mark with the methanol to obtain concentration of 1, 2, 3, 4, and 5µg/ml. The curve of each solution against the Methanol was recorded. Absorbance at 243.56 nm was measured and the plot of absorbance vs. concentration was plotted. The straight-line equation was determined.

Calibration curve for the CLOP (7.5 - 37.5 µg/ml)

Appropriate volume of aliquot from standard CLOP stock solution was transferred to different volumetric flasks of 10 ml capacity. The volume was adjusted to the mark with the methanol to obtain concentration of 7.5, 15, 22.5, 30, and 37.5µg/ml. The curve of each solution against the Methanol was recorded. Absorbance at 223.38 nm was measured and the plot of absorbance vs. concentration was plotted. The straight-line equation was determined.

Calibration curve for the ASP (7.5 - 37.5 µg/ml)

Appropriate volume of aliquot from standard ASP stock solution was transferred to different volumetric flasks of 10 ml capacity. The volume was adjusted to the mark with the methanol to obtain concentration of 7.5, 15, 22.5, 30, and 37.5µg/ml. The curve of each solution against the Methanol was recorded. Absorbance at 223.38 nm was measured and the plot of absorbance vs. concentration was plotted. The straight-line equation was determined.

Preparation of Sample solution:

Twenty tablets were weighed and finely powdered. The powder equivalent to 10 mg ROS, 75 mg CLOP and 75 mg ASP was accurately weighed and transferred to volumetric flask of 100ml capacity. Powder was dissolved in methanol in volumetric flask. The flask was shaken and volume was made up to the mark with methanol. The solution was filtered through whatmann filter paper (0.45µ). 10 ml of aliquot was taken and transferred to volumetric flask of 100 ml capacity to obtained a solution of 10µg/ml of ROS, 75µg/ml of CLOP and 75µg/ml of ASP. Volume was made up to the mark with methanol. Further 3.0 ml of this solution was transferred to volumetric flask of 10ml capacity. Volume was made up to the mark with methanol to give a solution containing 3µg/ml ROS,

22.5µg/ml CLOP and 22.5µg/ml ASP. This solution was used for the estimation of ROS, CLOP and ASP in tablet dosage form.

**Estimation of ROS, CLOP and ASP by Simultaneous Equation Method.**

Absorbance of the resulting solution was measured at 243.56 nm for determination of ROS, at 223.38 nm for determination of CLOP and at 276.44 nm for determination of ASP. The amounts of the ROS, CLOP and ASP present in the sample solution were calculated by as follow,

$$C_{ROS} = (A1(ay2az3 - az2ay3) - ay1(A2az3 - az2A3) + az1(A2ay3 - ay2A3)) / (ax1(ay2az3 - az2ay3) - ay1(ax2az3 - az2ax3) + az1(ax2ay3 - ay2 ax3)) \dots\dots(1),$$

$$C_{CLOP} = (ax1(A2az3 - az2A3) - A1(ax2az3 - az2ax3) + az1(ax2A3 - az2ax3)) / (ax1(ay2az3 - az2ay3) - ay1(ax2az3 - az2ax3) + az1(ax2ay3 - ay2ax3)) \dots\dots(2),$$

$$C_{ASP} = (ax1(ay2A3 - A2ay3) - ay1(ax2A3 - A2ax3) + A1(ax2ay3 - ay2ax3)) / (ax1(ay2az3 - az2ay3) - ay1(ax2az3 - az2ax3) + az1(ax2ay3 - ay2ax3)) \dots\dots(3)$$

Where,  
 A1, A2 and A3 are the absorbances of mixture at λ1, λ2 and λ3 respectively,  
 ax1, ax2 and ax3 are absorptivities of ROS at λ1, λ2 and λ3 respectively,  
 ay1, ay2 and ay3 are absorptivities of CLOP at λ1, λ2 and λ3 respectively,  
 az1, az2 and az3 are absorptivities of ASP at λ1, λ2 and λ3 respectively,  
 C<sub>ROS</sub>, C<sub>CLOP</sub> and C<sub>ASP</sub> are the concentrations of ROS, CLOP and ASP, respectively in mixture.

Validation of spectrophotometric method:

- (1) Accuracy  
 Accuracy was determined by calculating recovery of ROS, CLOP and ASP by the standard addition method. Known amounts of standard solutions of ROS, CLOP and ASP were added to a pre-quantified test solutions. Each solution was measured in triplicate, and the recovery was calculated by measuring absorbance.
- (2) Precision  
 The precision of an analytical method is the degree of agreement among individual test results when the method is applied repeatedly to multiple samplings of homogenous samples.

### (3) Repeatability

Standard solutions of ROS were prepared of linearity range and spectrums were recorded. Absorbance was measured at 243.56 nm, 223.38 nm and 276.44 nm. The absorbance of the same concentration solution was measured six times and RSD was calculated.

In the similar manner solutions of CLOP and ASP were prepared and spectrums were recorded. Absorbance was measured at 243.56 nm, 223.38 nm and 276.44 nm. The procedure was repeated for six times and RSD was calculated.

### (4) Intra and inter day precision

Variation of results within the same day (intraday), variation of results between days (interday) were analyzed.

Intraday precision was determined by analyzing ROS, CLOP and ASP individually for three times in the same day at 243.56 nm, 223.38 nm and 276.44 nm.

Inter day precision was determined by analyzing ROS, CLOP and ASP individually daily for three day at 243.56 nm, 223.38 nm and 276.44 nm.

### (5) Linearity and Range

The linearity of analytical method is its ability to elicit test results that are directly proportional to the concentration of analyte in sample within a given range. The range of analytical method is the interval between the upper and lower levels of analyte that have been demonstrated to be determined within a suitable level of precision, accuracy and linearity.

## RESULT AND DISCUSSION:

From overlain spectra of ROS, CLOP and ASP it is clear that ROS exhibited at  $\lambda_{max}$  243.56 nm, CLOP at  $\lambda_{max}$  223.38 nm, and ASP at  $\lambda_{max}$  276.44 nm. The overlain spectra of ROS, CLOP and ASP reveals that the both the drug exhibits distinct  $\lambda_{max}$  for estimation of ROS, CLOP and ASP using Simultaneous Equation method. In this method the  $\lambda_{max}$  of three drug is required.

Calibration data at 243.56, 223.38 and 276.44 nm for ROS, CLOP and ASP are shown in Table. Calibration curves for ROS, CLOP and ASP were plotted between absorbance and concentration. The following equations for straight line were obtained for ROS, CLOP and ASP.

Linear equation for ROS at 243.56 nm,  $Y = 0.0837x + 0.0917$  Linear equation for ROS at 223.38 nm,  $Y = 0.0549x + 0.0641$  Linear equation for ROS at 276.44 nm,  $Y = 0.0306x + 0.0388$  Linear equation for CLOP at 243.56 nm,  $Y = 0.0050x + 0.0333$  Linear equation for CLOP at 223.38

nm,  $Y = 0.0235x + 0.0719$  Linear equation for CLOP at 276.44 nm,  $Y = 0.0032x + 0.0090$  Linear equation for ASP at 243.56 nm,  $Y = 0.0116x + 0.0381$  Linear equation for ASP at 223.38 nm,  $Y = 0.0100x + 0.0568$  Linear equation for ASP at 276.44 nm,  $Y = 0.0238x + 0.0617$

The developed Simultaneous Equation method was validated. The linear range, correlation coefficient, detection limit and standard deviation for ROS, CLOP and ASP by Spectroscopy method are shown in Table. Accuracy was determined by calculating the recovery. The method was found to be accurate with % recovery 98.63-101.91 % at 243.56nm, 223.38nm and 276.44 nm for ROS, 98.73-101.45% at 243.56nm, 223.38nm and 276.44 nm for CLOP and 98.13-101.70% at 243.56nm, 223.38nm and 276.44 nm for ASP. Precision was calculated as repeatability and intraday and interday variation for three drugs. The LOD and LOQ for ROS was found to be 0.022 $\mu$ g/ml and 0.126  $\mu$ g/ml at 243.56 nm, 0.044 $\mu$ g/ml and 0.134  $\mu$ g/ml at 223.38 nm and 0.056 $\mu$ g/ml and 0.169  $\mu$ g/ml at 276.44 nm. The LOD and LOQ for CLOP was found to be 0.343  $\mu$ g/ml and 1.040  $\mu$ g/ml at 243.56 nm, 0.495 $\mu$ g/ml and 1.289 $\mu$ g/ml at 223.38nm and 0.515 $\mu$ g/ml and 1.562  $\mu$ g/ml at 276.44 nm. The LOD and LOQ for ASP was found to be 0.685 $\mu$ g/ml and 2.018  $\mu$ g/ml at 243.56 nm, 0.772  $\mu$ g/ml and 2.340  $\mu$ g/ml at 223.38 nm and 0.894 $\mu$ g/ml and 2.485  $\mu$ g/ml at 276.44 nm respectively. Summary of validation parameters are shown.

Marketed formulation was analyzed by the proposed method and assay result of marketed formulation is shown.

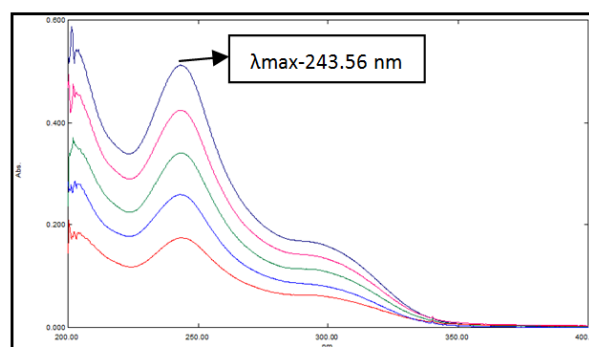


Figure:4: Overlain Spectra of ROS at (1-5  $\mu$ g/ml)

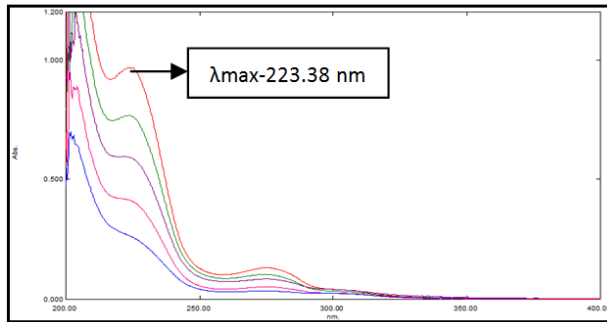


Figure:5: Overlay Spectra of CLOP at (7.5-22.5 µg/ml)

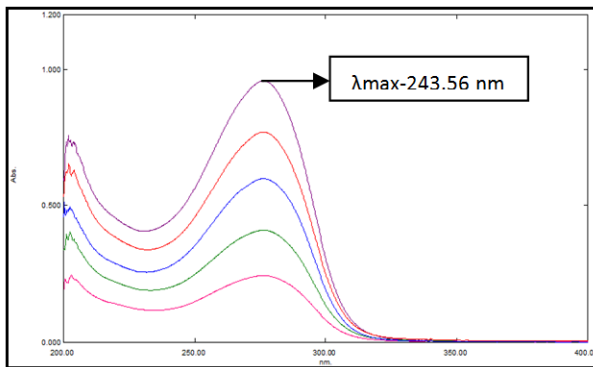


Figure:6: Overlay Spectra of ASP at (7.5-22.5 µg/ml)

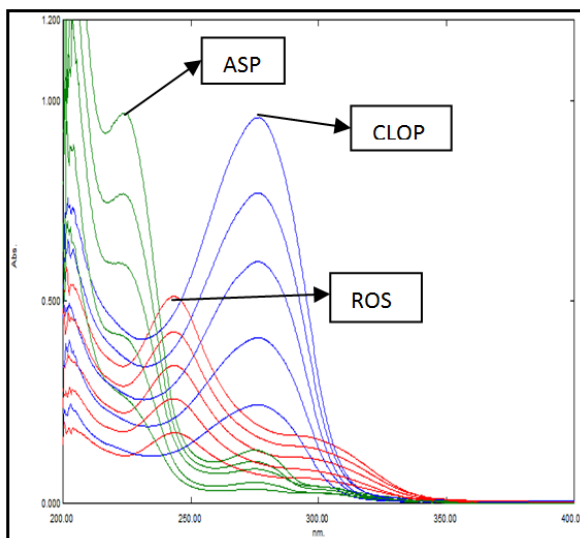


Figure:7: Overlay spectrum of ROS, CLOP and ASP

Table:1: Result of calibration readings of ROS at 243.56 nm.

Concentration (µg/ml)	Absorbance at 243.56 nm Mean ± S.D. (n=5)
1	0.1742 ± 0.0004
2	0.2614 ± 0.0005
3	0.3440 ± 0.0008
4	0.4241 ± 0.0007
5	0.5116 ± 0.0005

Concentration (µg/ml)	Absorbance at 276.44 nm Mean ± S.D. (n=5)
1	0.0713 ± 0.0004
2	0.0991 ± 0.0005
3	0.1296 ± 0.0004
4	0.1610 ± 0.0008
5	0.1932 ± 0.0006

Table:2: Result of calibration readings of ROS at 223.38

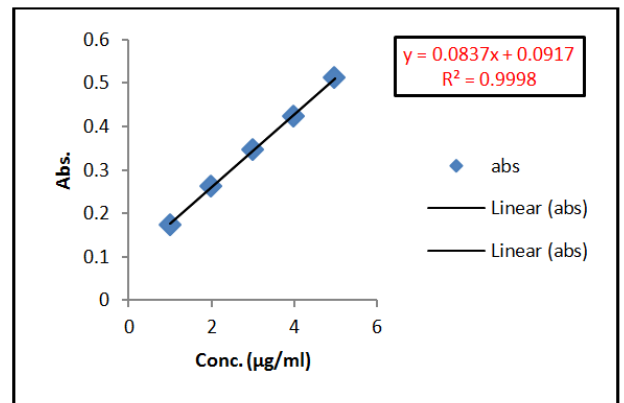


Figure:8: Calibration curve of ROS at 243.56 nm

Table:3: Result of calibration readings of ROS at 276.44 nm.nm.

Concentration (µg/ml)	Absorbance at 223.38 nm Mean ± S.D. (n=5)
1	0.1183 ± 0.0005
2	0.1771 ± 0.0007
3	0.2267 ± 0.0010
4	0.2842 ± 0.0007
5	0.3390 ± 0.0008

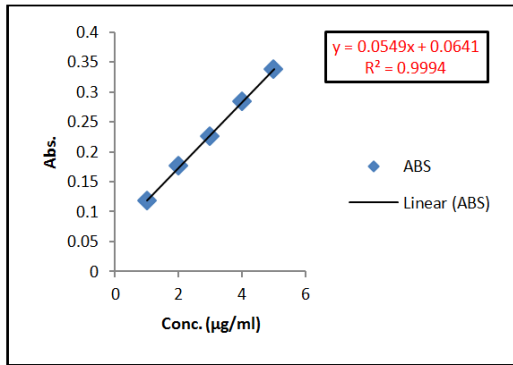


Figure:9: Calibration curve of ROS at 223.38 nm

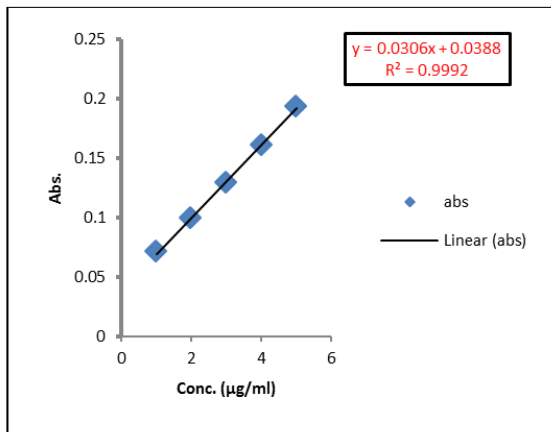


Figure:10: Calibration curve of ROS at 276.44 nm

Table:4: Result of calibration readings of CLOP at 243.56 nm.

Concentration (µg/ml)	Absorbance at 243.56 nm Mean ± S.D. (n=5)
7.5	0.0713 ± 0.0003
15	0.0991 ± 0.0005
22.5	0.1296 ± 0.0004
30	0.1610 ± 0.0008
37.5	0.1932 ± 0.0005

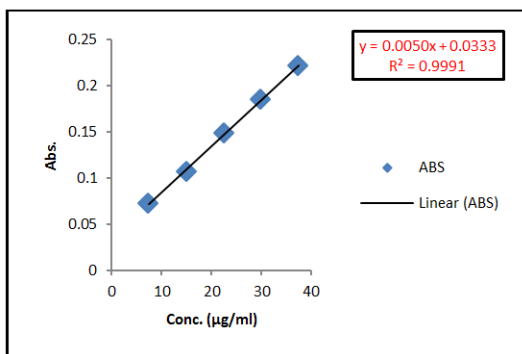


Figure:11: Calibration curve of CLOP at 243.56 nm

Table:5: Result of calibration readings of CLOP at 223.38 nm.

Concentration (µg/ml)	Absorbance at 223.38 nm Mean ± S.D. (n=5)
7.5	0.2613 ± 0.0007
15	0.4185 ± 0.0004
22.5	0.5971 ± 0.0008
30	0.7760 ± 0.0007
37.5	0.9672 ± 0.0008

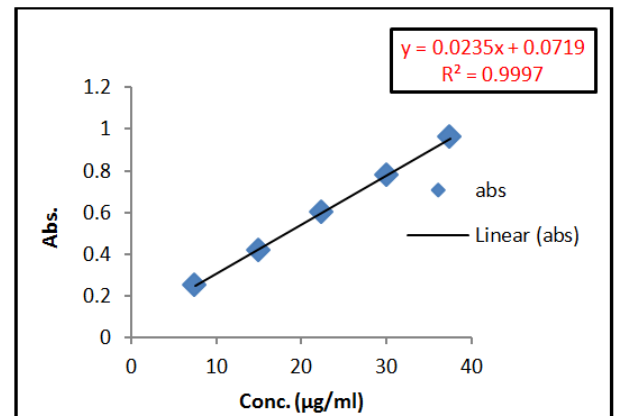


Figure:12: Calibration curve of CLOP at 223.38 nm

Table:6: Result of calibration readings of CLOP at 276.44 nm.

Concentration (µg/ml)	Absorbance at 276.44 nm Mean ± S.D. (n=5)
7.5	0.0342 ± 0.0004
15	0.0567 ± 0.0004
22.5	0.0811 ± 0.0005
30	0.1043 ± 0.0007
37.5	0.1300 ± 0.0005

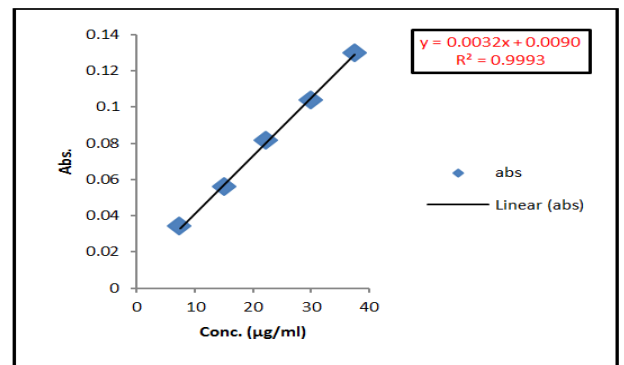


Figure:13: Calibration curve of CLOP at 276.44 nm

Table:7: Result of calibration readings of ASP at 243.56 nm.

Concentration (µg/ml)	Absorbance at 243.56 nm Mean ± S.D. (n=5)
7.5	0.1272 ± 0.0004
15	0.2116 ± 0.0005
22.5	0.2941 ± 0.0008
30	0.3864 ± 0.0007
37.5	0.4731 ± 0.0004

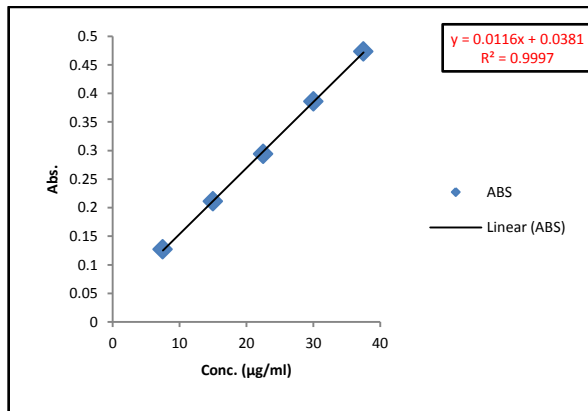


Figure:14: Calibration curve of ASP at 243.56 nm

Table:9: Result of calibration readings of ASP at 276.44 nm.

Concentration (µg/ml)	Absorbance at 276.44 nm Mean ± S.D. (n=5)
7.5	0.2463 ± 0.0005
15	0.4127 ± 0.0004
22.5	0.5990 ± 0.0008
30	0.7736 ± 0.0007
37.5	0.9591 ± 0.0010

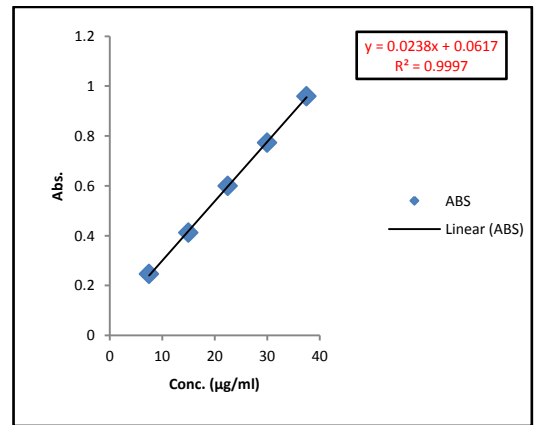


Figure:16: Calibration curve of ASP at 276.44 nm

Table:8: Result of calibration readings of ASP at 223.38 nm.

Concentration (µg/ml)	Absorbance at 223.38 nm Mean ± S.D. (n=5)
7.5	0.1325 ± 0.0005
15	0.2083 ± 0.0008
22.5	0.2761 ± 0.0004
30	0.3603 ± 0.0005
37.5	0.4300 ± 0.0007

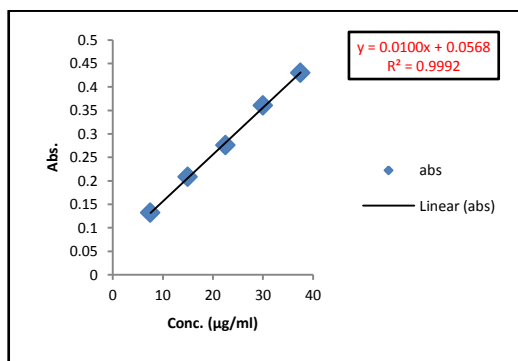


Figure:15: Calibration curve of ASP at 223.38 nm

Table:10: Result for precision of intraday at 243.56 nm.

Drugs	Concentration (µg/ml)	Absorbance at 243.56 nm Mean ± S.D. (n=3)	%RSI
ROS	2	0.2621 ± 0.0005	0.5203
	3	0.3454 ± 0.0006	0.5970
	4	0.4240 ± 0.0008	0.6758
CLOP	7.5	0.1073 ± 0.0006	0.5379
	15	0.1476 ± 0.0005	0.4802
	22.5	0.1861 ± 0.0009	0.8185
ASP	7.5	0.2100 ± 0.0006	0.6044
	15	0.2952 ± 0.0008	1.1511
	22.5	0.3877 ± 0.0007	0.5167

Table:11: Result for precision of intraday at 223.38 nm.

Drugs	Concentration (µg/ml)	Absorbance at 223.38 nm Mean ± S.D. (n=3)	%RSI
ROS	2	0.1782 ± 0.0009	0.8617
	3	0.2255 ± 0.0013	1.2768
	4	0.2841 ± 0.0006	0.6028

CLOP	7.5	0.4193 ± 0.0005	0.6376
	15	0.5960 ± 0.0010	1.0739
	22.5	0.7756 ± 0.0015	0.9870
ASP	7.5	0.2091 ± 0.0010	0.9569
	15	0.2778 ± 0.0007	0.5501
	22.5	0.3613 ± 0.0009	0.7596

Table:12: Result for precision of intraday at 276.44 nm.

Drugs	Concentration (µg/ml)	Absorbance at 276.44 nm Mean ± S.D. (n=3)	%RS
ROS	2	0.0985 ± 0.0007	0.6851
	3	0.1291 ± 0.0009	0.8028
	4	0.1603 ± 0.0011	0.9686
CLOP	7.5	0.0554 ± 0.0006	0.5434
	15	0.0826 ± 0.0008	0.9984
	22.5	0.1050 ± 0.0010	1.1523
ASP	7.5	0.4117 ± 0.0005	0.4710
	15	0.5981 ± 0.0010	0.9672
	22.5	0.7740 ± 0.0008	0.7685

Table:13: Result for precision of interday at 243.56 nm.

Drugs	Concentration (µg/ml)	Absorbance at 243.56 nm Mean ± S.D. (n=3)	%RSD
ROS	2	0.2593 ± 0.0007	0.7226
	3	0.3401 ± 0.0009	0.9392
	4	0.4186 ± 0.0010	1.0711
CLOP	7.5	0.1042 ± 0.0005	0.6853
	15	0.1444 ± 0.0007	0.8741
	22.5	0.1820 ± 0.0013	1.2191
ASP	7.5	0.2073 ± 0.0008	0.8007
	15	0.2911 ± 0.0010	1.3236
	22.5	0.3840 ± 0.0008	0.9313

Table:14: Result for precision of interday at 223.38 nm.

Drugs	Concentration (µg/ml)	Absorbance at 223.38 nm Mean ± S.D. (n=3)	%RS
ROS	2	0.1741 ± 0.0011	1.0827
	3	0.2210 ± 0.0017	1.3209
	4	0.2809 ± 0.0008	0.9142
CLOP	7.5	0.4168 ± 0.0007	0.8284
	15	0.5941 ± 0.0013	1.1941
	22.5	0.7730 ± 0.0018	1.2093

ASP	7.5	0.2062 ± 0.0012	1.152
	15	0.2737 ± 0.0009	0.7789
	22.5	0.3583 ± 0.0012	0.9127

Table:15 Result for precision of interday at 276.44 nm.

Drugs	Concentration (µg/ml)	Absorbance at 276.44 nm Mean ± S.D. (n=3)	%RS
ROS	2	0.0942 ± 0.0009	0.8098
	3	0.1253 ± 0.0012	1.0627
	4	0.1576 ± 0.0015	1.1679
CLOP	7.5	0.0520 ± 0.0008	0.7029
	15	0.0789 ± 0.0010	1.0675
	22.5	0.1015 ± 0.0017	1.2174
ASP	7.5	0.4086 ± 0.0009	0.6416
	15	0.5963 ± 0.0013	1.1791
	22.5	0.7718 ± 0.0010	0.9006

Table:16: Result for Accuracy study at 243.56 nm.

Drugs	Level	Amt. of Sample (µg/ml)	Amt. of STD. Spiked	Total Amt.	Amt Found (µg/ml)
ROS	80	2	1.6	3.6	99.44
	100	2	2	4	100.25
	120	2	2.4	4.4	100.45
CLOP	80	15	12	27	101.14
	100	15	15	30	99.10
	120	15	18	33	100.3
ASP	80	15	12	27	99.92
	100	15	15	30	99.13
	120	15	18	33	100.21

Table:17: Result for Accuracy study at 223.38 nm.

Drugs	Level	Amt. of Sample (µg/ml)	Amt. of STD. Spiked	Total Amt.	Amt Found (µg/ml)
ROS	80	2	1.6	3.6	99.16
	100	2	2	4	100.50
	120	2	2.4	4.4	99.77
CLOP	80	15	12	27	99.62
	100	15	15	30	98.73
	120	15	18	33	100.3
ASP	80	15	12	27	101.45
	100	15	15	30	101.56
	120	15	18	33	99.66



Table:18: Result for Accuracy study at 276.44 nm.

Drugs	Level	Amt. of Sample (µg/ml)	Amt. of STD. Spiked	Total Amt.	Amt Found (µg/ml)
ROS	80	2	1.6	3.6	101.94
	100	2	2	4	100.73
	120	2	2.4	4.4	98.63
CLOP	80	15	12	27	98.74
	100	15	15	30	99.63
	120	15	18	33	100.36
ASP	80	15	12	27	101.70
	100	15	15	30	99.56
	120	15	18	33	100.09

## CONCLUSION

The low value of relative standard deviation for repeated measurement indicates that the method is precise. The value of % recovery is approximately 100%, which indicates that these methods can be used for estimation of these three drugs in combined dosage forms without any interference due to the other components present in the formulations. Hence this study presents simple, accurate, precise and rapid spectroscopic analytical method for the simultaneous estimation of these three drugs in pharmaceutical dosage form.

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