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RESEARCH ARTICLE

R. Veereswara Rao al, The Experiment, 2018, Vol 46(1), 2619-2632

STABLE AND NOVEL HPLC METHOD FOR THE DETERMINATION OF CLOPIDOGREL AND ATORVASTATIN IN CAPSULE FORMULATION

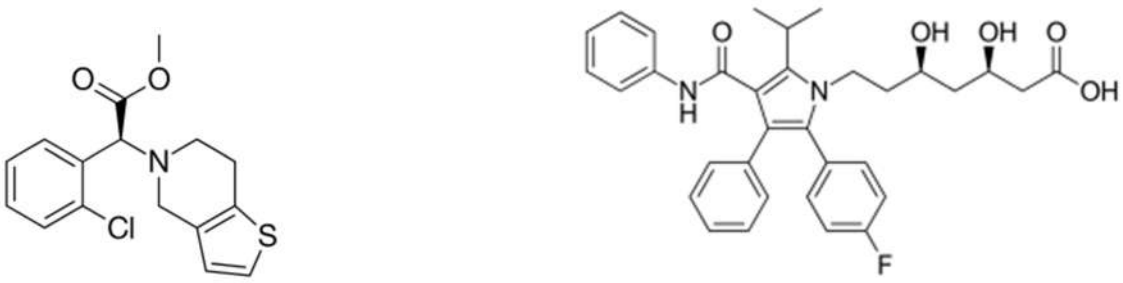
ABSTRACT

Clopidogrel and atorvastatin are available in capsule formulation to treat heart related diseases and abnormalities. Simple HPLC method was developed and validated. Chromatographic conditions are Mobile phase: 1ml of OPA solution in a 1000ml of of water and mix the above OPA solution with acetonitrile in the ratio of 60:40 v/v; Flow rate: 1 ml/min; Column: Discovery C18 150 x 4.6 mm, 5m; Detector wave length: 240nm; Column temperature: 30°C; Injection volume: 10mL; Run time: 6min; Diluent: Water: Acetonitrile (50:50). Method validation results were confirmed the method extendedness with precision % RSD, linearity correlation coefficient above 0.999, accuracy recovery between 98% to 102%, specificity interference, peak purity and % degradation, ruggedness and robustness results. Eventually the finalized method can be used for the determination of Clopidogrel and atorvastatin in capsule formulation.

INTRODUCTION

Clopidogrel is used as antiplatelet medication to reduce the risk of heart related diseases and stroke[1-3]. Clopidogrel chemical name is (+)-(*S*)-methyl 2-(2-chlorophenyl)-2-(6,7-dihydrothieno[3,2-*c*]pyridin-5(4*H*)-yl)acetate. Clopidogrel is used with the combination of Aspirin for the placement of coronary artery stent (dual antiplatelet therapy)[4-5]. Clopidogrel is used for heart stroke who has history of myocardial infarction and other forms of acute coronary syndrome, stroke and peripheral artery disease[6].

Atorvastatin is used to treat cardiovascular disease and for abnormal lipid levels (dyslipidemia)[7-8]. Chemical name is (3*R*,5*R*)-7-[2-(4-Fluorophenyl)-3-phenyl-4-(phenylcarbamoyl)-5-propan-2-ylpyrrol-1-yl]-3,5-dihydroxyheptanoic acid. Primary prevention of heart attack, stroke and revascularization procedures in people who have risk factors such as age, smoking, high blood pressure, low HDL-C and family history of early heart disease[9-10]. Secondary prevention is myocardial infarction, stroke, unstable angina and revascularization[11-12]. Chemical structures of Clopidogrel and atorvastatin were represented in figure-1.



Clopidogrel Atorvastatin

Figure-1: Chemical structure of Clopidogrel Atorvastatin

Clopidogrel and atorvastatin has many analytical methods to determine individually and combination with other products such as Clopidogrel and aspirin[13-18]. Reported methods have large run time and consumption of buffer salts and organic modifier are high[19-20]. The main objective of this study was to develop a simple and less runtime method with less mobile phase consumption.

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MATERIALS AND METHOD

Product information:

Clopidogrel and Atorvastatin are available in capsule formulation each capsule contains 75mg of Clopidogrel and 10mg of Atorvastatin.

Mobile phase:

1ml of Ortho phosphoric acid (OPA) solution in a 2000ml of volumetric flask add 1000ml of water and stir with glass rod to get homogenization. Mix the above OPA solution with acetonitrile in the ratio of 60:40 v/v and filter with 0.2µ PVDF filter.

Chromatographic conditions:

Flow rate	**:**	1 ml/min
Column	**:**	Discovery C18 150 x 4.6 mm, 5m.
Detector wave length	**:**	240nm
Column temperature	**:**	30°C
Injection volume	**:**	10mL
Run time	**:**	6min
Diluent	**:**	Water: Acetonitrile (50:50)

Preparation of Solutions:

Standard Preparation: Accurately Weighed and transferred 18.75mg of Clopidogrel and 2.5mg of Atorvastatin standard materials into a 25 ml & 25ml clean dry volumetric flasks, add 10ml of diluent, sonicated for 10 minutes and make up to the final volume with diluents.

Sample Preparation: 10 capsules were weighed and calculated the average fill weight of each capsule and crushed the content in to fine powder. Weighed equivalent to 75mg of Clopidogrel and 10mg of Atorvastatin was transferred into a 100ml volumetric flask, 7ml of diluent added and sonicated for 30 min, further the volume made up with diluent and filtered. (750µg/ml of Clopidogrel; 100µg/ml of Atorvastatin). 1ml of filtered sample stock solution was transferred to 10ml volumetric flask and made up with diluent. (75µg/ml of Clopidogrel 10µg/ml of Atorvastatin)

RESULTS AND DISCUSSION

METHOD DEVELOPMENT:

HPLC method development was initiated based on the literature and chemical and physical properties of the both analytes. Initial method development was started with water and acetonitrile with pH 3.0 adjustment.

Method development trial-1:

Conditions: M.P.: water and acetonitrile 8:20 v/v pH 3.0 with acetic acid; column YMC pack C18 150*4.6mm, 3µm; flow rate 1.0ml/min; 240 nm wavelength; 50µL injection volume; 25°C column temp.; standard solution 0.2mg/ml concentration.

Results: Clopidogrel and atorvastatin were eluted but atorvastatin peak shape was very poor. Method development trial chromatogram was represented in figure-2.

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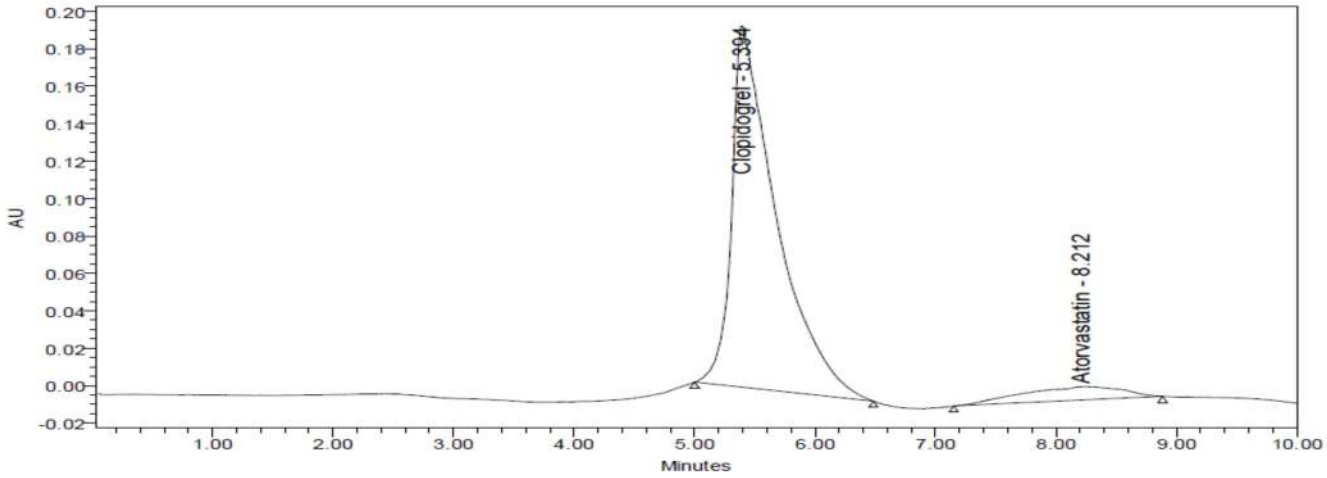

Figure-2: Method development trial-1 chromatogram

Method development trial-2:

Conditions: M.P. A: 0.03M ammonium acetate in water; M.P. B: ACN; column X terra RP18 250*4.6mm, 5µm; flow rate 1.0ml/min; 240 nm wavelength; 20µL injection volume; 30°C column temp.; gradient program: 0-5 min M.P. A 80% B 20%, 30-40 min M.P. A 10% B 90%, 41-50 min M.P. A 80 % B 20%; standard solution 0.2 mg/ml concentration.

Results: Both analytes were separated and elution is acceptable but at 7.4 min blank peak was observed. Method development trial chromatogram was represented in figure-3.

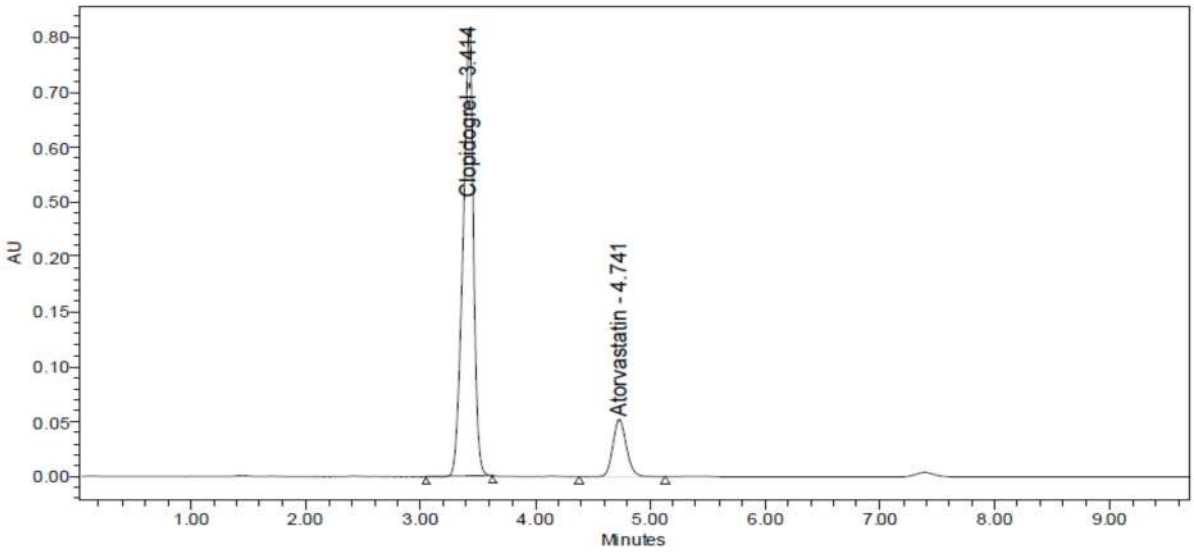

Figure-3: Method development trial-2 chromatogram

Method development trial-3:

Conditions: M.P. A: 0.03M KH_2PO_4 in water; M.P. B: ACN; column X terra RP18 250*4.6mm, 5µm; flow rate 1.0ml/min; 240 nm wavelength; 20µL injection volume; 30°C column temp.; gradient program: 0-5 min M.P. A 80% B 20%, 30-40 min M.P. A 10% B 90%, 41-50 min M.P. A 80 % B 20%; standard solution 0.2 mg/ml concentration.

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Results: Both analytes were separated and elution is acceptable but run time is high. Method development trial chromatogram was represented in figure-4.

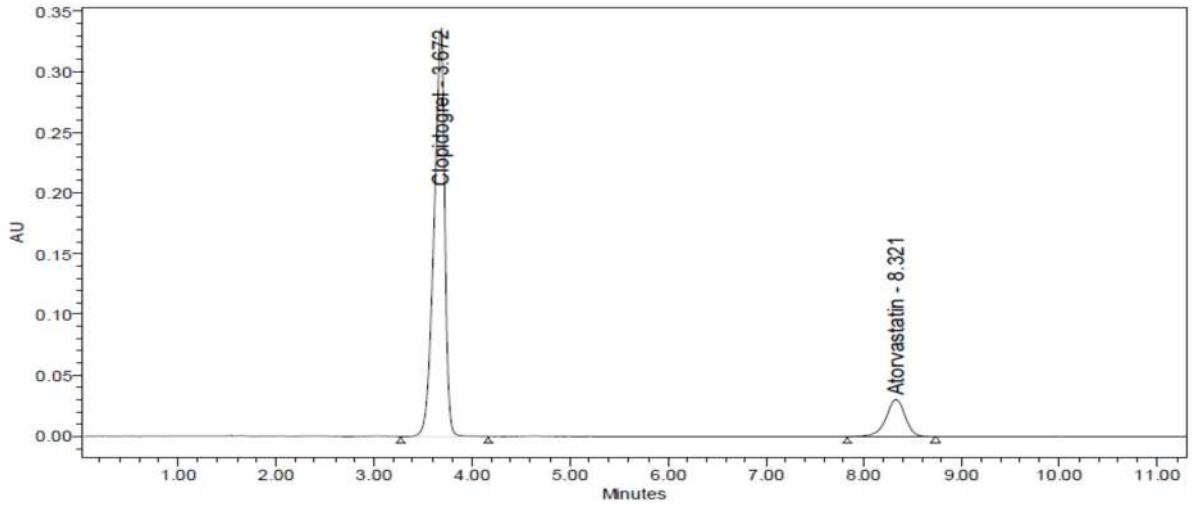

Figure-4: Method development trail-3 chromatogram

Method development trial-4:

Conditions: M.P. A: 1.0ml OPA in water and mixed this solution with acetonitrile in 60:40 v/v ratio; column X terra RP18 250*4.6mm, 5µm; flow rate 1.0ml/min; 240 nm wavelength; 20µL injection volume; 30°C column temp.; standard solution 0.2 mg/ml concentration.

Results: Both analytes were separated and elution is acceptable and runtime also good. Hence this method conditions were finalized.

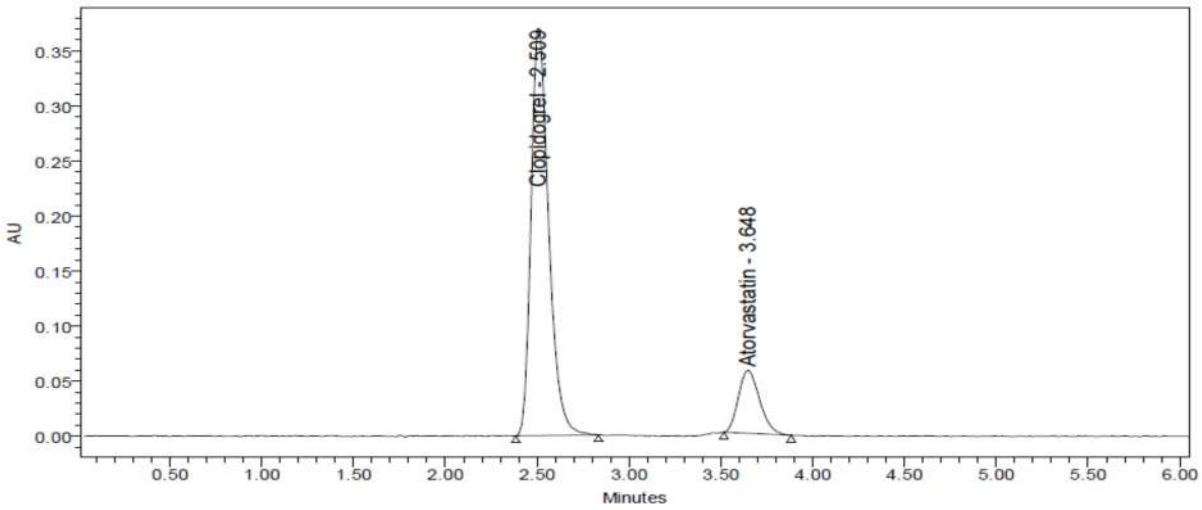

Figure-5: Final trial method development chromatogram

METHOD VALIDATION:

Precision:

Method and system precision was performed with six replicate test solutions. Six replicates % assay values were calculated for clopidogrel and atorvastatin. Intermediate precision was performed with different HPLC instrument, column and analyst.

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% assay and %RSD values were calculated and reported in table-1. Blank, placebo, standard solution and test sample chromatograms were represented in figure-6 to 9.

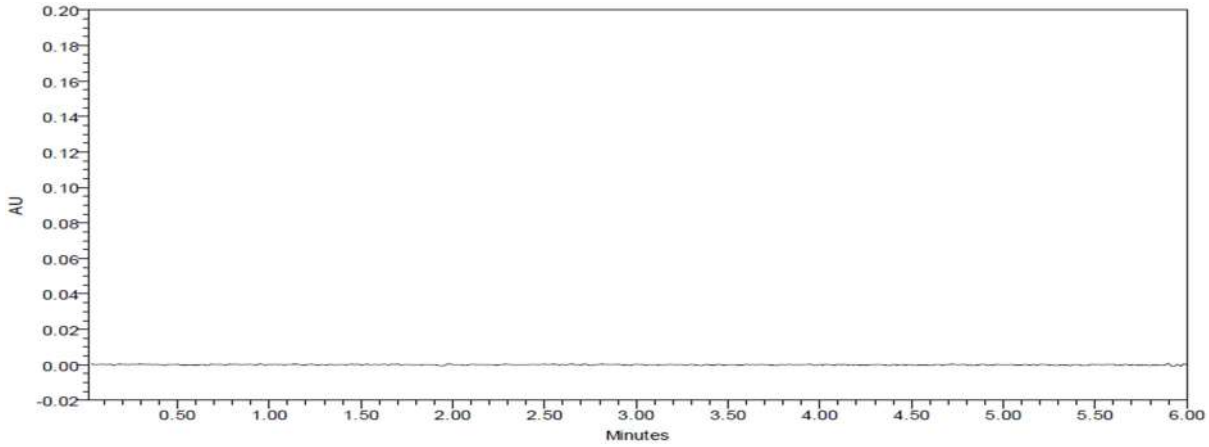

Figure-6: Blank chromatogram

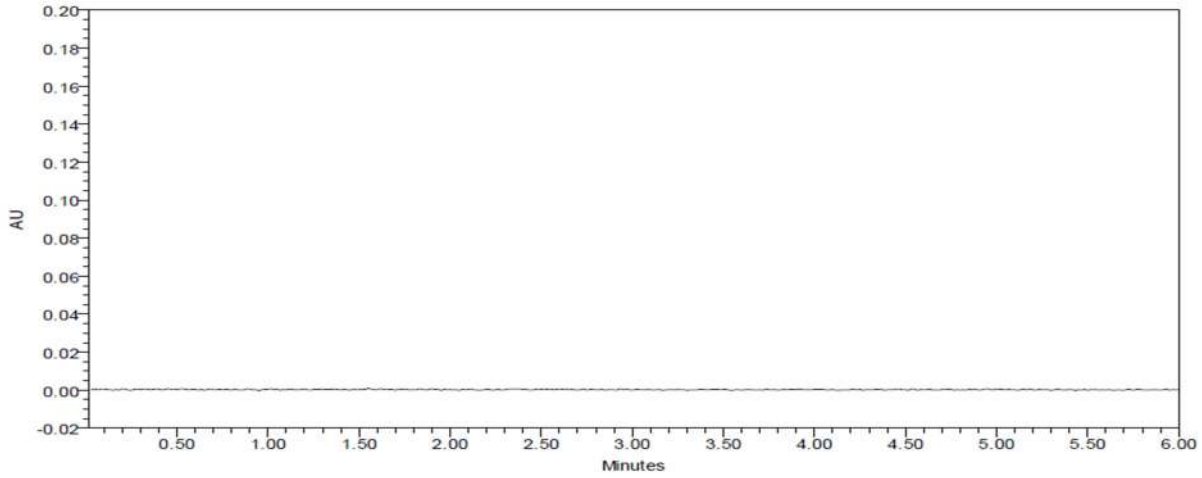

Figure-7: Placebo chromatogram

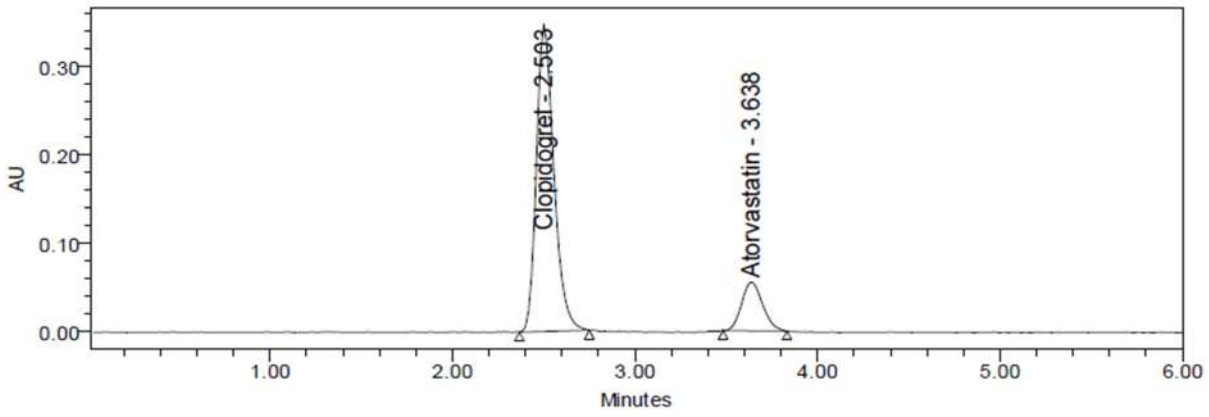

Figure-8: Standard solution chromatogram

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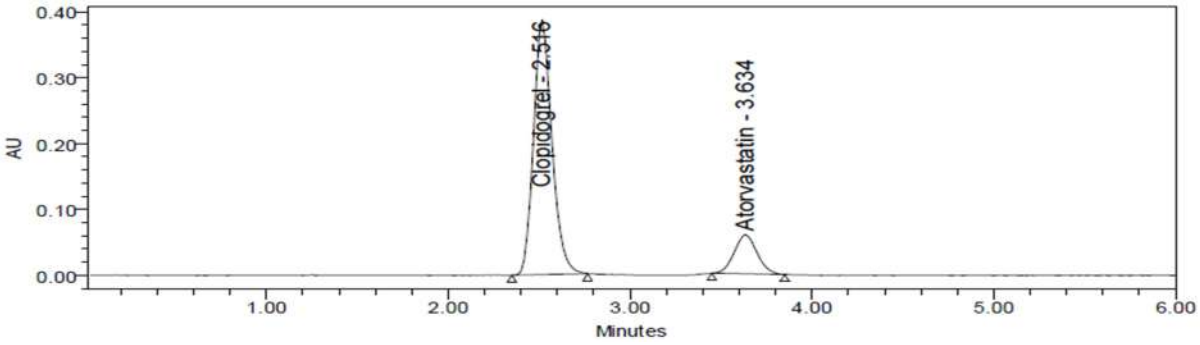

Figure-9: Precision sample chromatogram

Table-1: Precision and intermediate precision results

Precision results								
Assay %	**1**	**2**	**3**	**4**	**5**	**6**	**Avg.**	**%RSD**
Clopidogrel	231110	2311087	2301524	2314945	2317695	2301524	2309681	0.29
	99.73	99.72	99.30	99.88	100.0	99.72	99.72	0.23
Atorvastatin	446279	446501	446518	445975	443036	442764	445178	0.39
	99.93	99.98	99.98	99.86	99.20	99.14	99.68	0.40
Intermediate precision								
Clopidogrel	2308598	2303202	2286194	2303110	2309161	2289465	2299955	0.42
	99.61	99.8	98.64	99.7	99.63	98.78	99.36	0.51
Atorvastatin	426192	429766	427453	428578	420036	421163	425531	0.94
	100.06	100.89	100.35	100.62	98.61	98.87	99.9	0.94

Specificity:

Specificity was performed with different stress degradation conditions such as acid, base, peroxide, thermal, water and UV/visible light. Blank, placebo and impurities interference was evaluated and found no interference with product peaks Clopidogrel and atorvastatin. % assay and degradation was calculated. Degradation stress conditions, % assay, % degradation and peak purity results were tabulated in below table-2. Stress study chromatograms were represented in figure-10 to 15.

Table-2: Stress study results

Clopidogrel / Atorvastatin Stress study results						
S. No.	**Degradation condition**	**% of assay**	**% deg.**	**Purity Angle**	**Purity Threshold**	**Peak purity**
1.	Acid (2N HCl, 30min, 60°C)	99.82 / 96.13	5.18 / 6.85	0.139 / 0.167	0.393/ 0.433	Pass
2.	Base (2N NaOH, 30min, 60°C)	95.12 / 96.00	4.88/ 4.00	0.253 / 0.153	0.526/ 0.716	Pass
3.	Peroxide (20% H_2O_2, 30min, RT)	96.47 / 96.44	3.53/3.56	0.144 / 0.198	0.451 / 0.500	Pass
4.	Water (Water, 6hr, 60°C)	97.11 / 97.47	2.89 / 2.53	0.258 / 0.153	0.458 / 0.478	Pass
5.	Thermal (6hr, 105°C)	98.90 / 98.12	1.10 / 1.88	0.241 / 0.179	0.399 / 0.397	Pass
6.	UV/ visible light (UV light 200 watt hr/sq. meter) (Visible light 1.2 million lux hrs)	99.45 / 99.83	0.55 / 0.17	0.234 / 0.240	0.404 / 0.407	Pass

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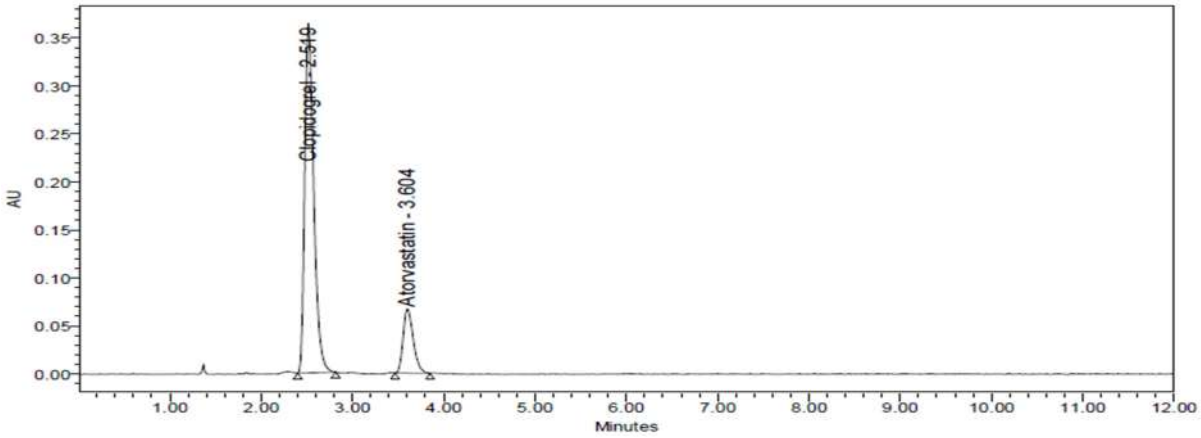

Figure-10: Acid degradation chromatogram

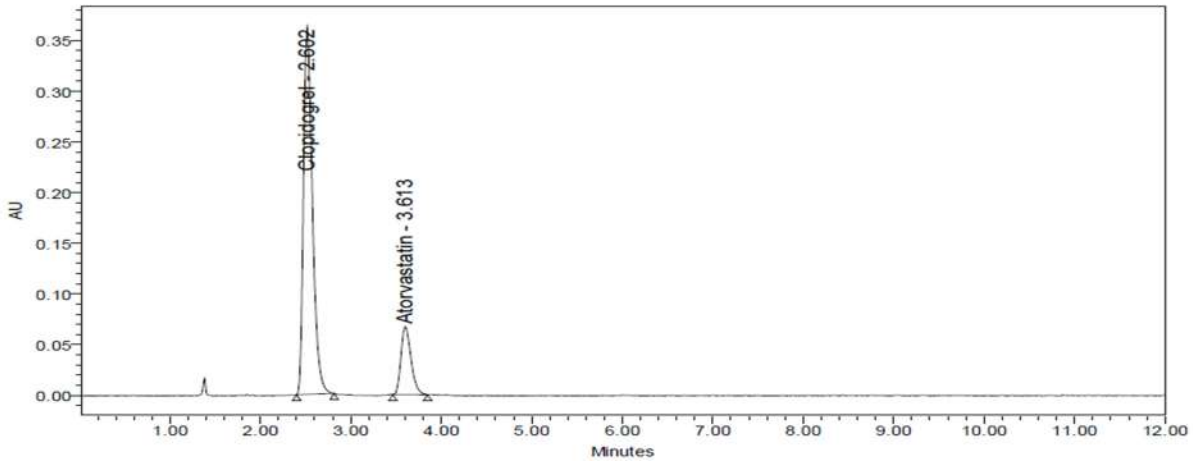

Figure-11: Base degradation chromatogram

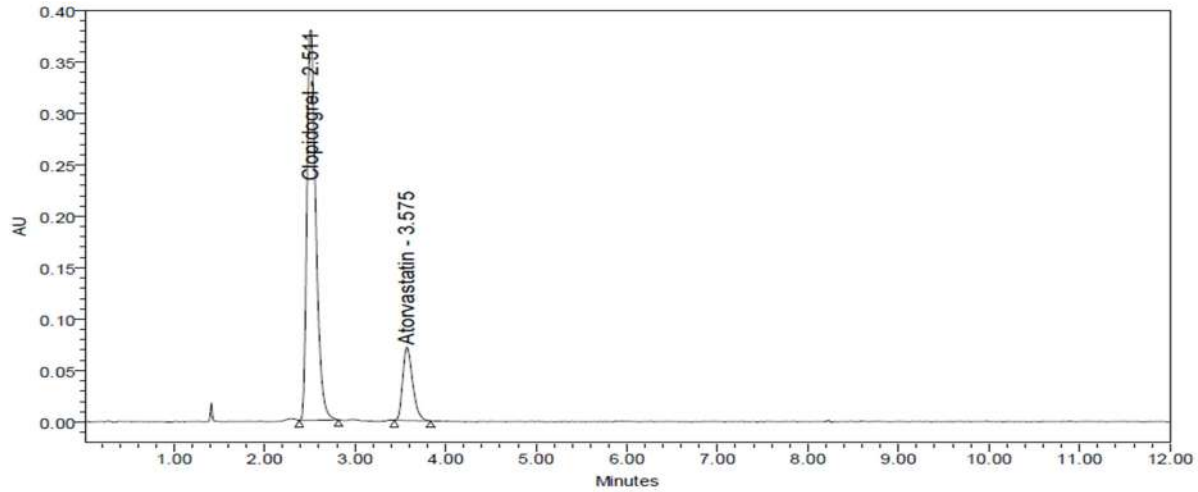

Figure-12: Peroxide degradation chromatogram

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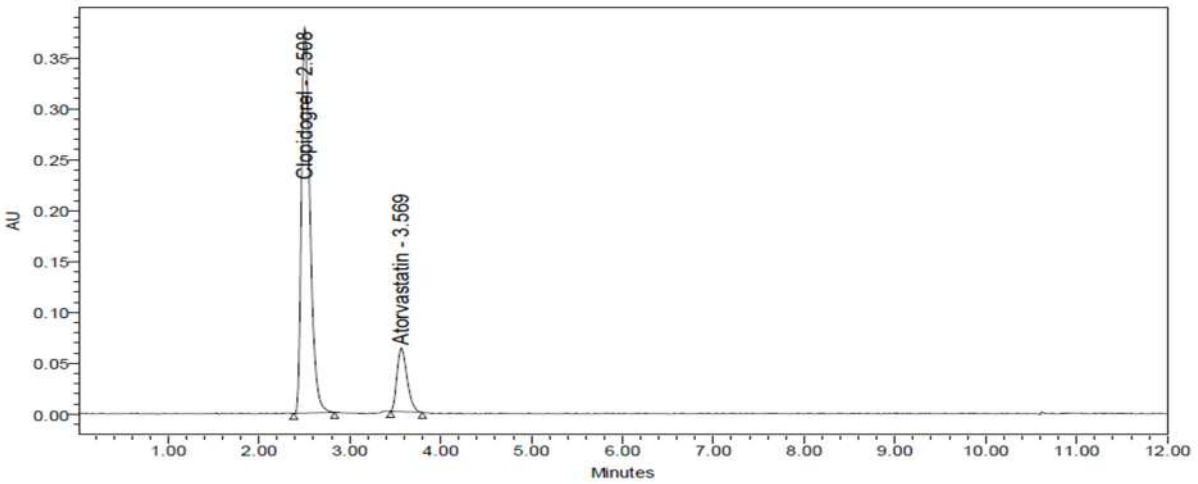

Figure-13: Thermal degradation chromatogram

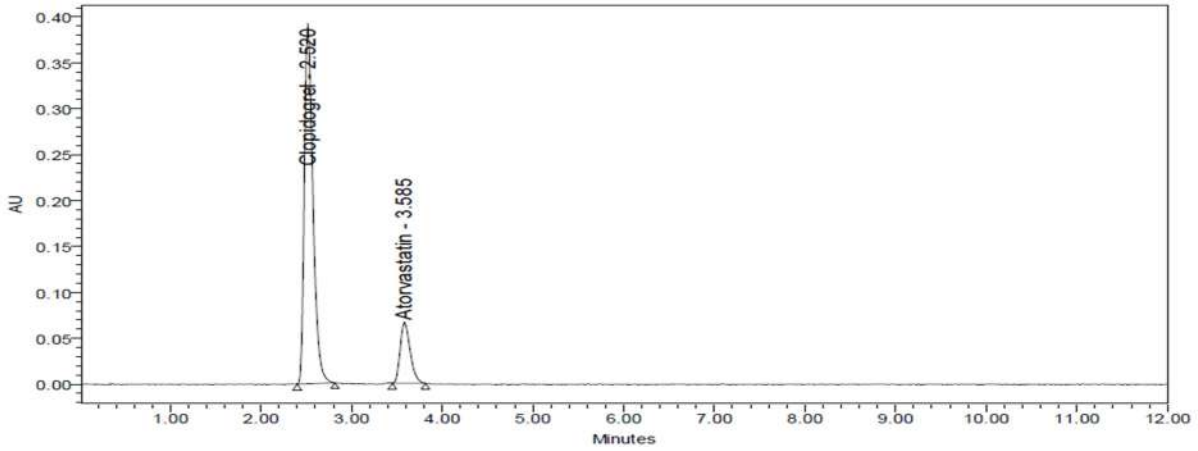

Figure-14: Water degradation chromatogram

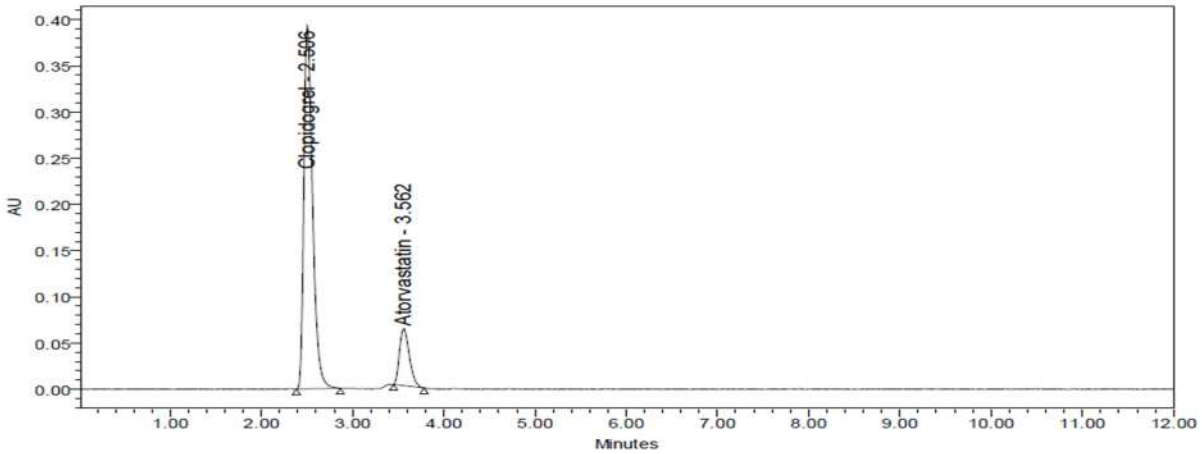

Figure-15: UV/visible light degradation chromatogram

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Linearity:

Linearity validation parameter was evaluated with six different concentration standard preparations. Clopidogrel linearity was evaluated from 18.75 ppm to 112.5 ppm concentration and atorvastatin linearity concentration from 2.5 ppm to 15.0 ppm. Linearity correlation coefficient value found satisfactory i.e. for Clopidogrel 0.9999 and atorvastatin 0.9996. Linearity results were tabulated in table-3 and chromatograms were represented in figure-16 to 21.

Table-3 Linearity results

Clopidogrel	Level	1	2	3	4	5	6	Corr. of coeffi.
	Conc. (µg/ml)	18.75	37.5	56.25	75	93.75	112.5	
	Area	574434	1159977	1735079	2317965	2864080	3430752	0.9999
Atorvastatin	Conc. (µg/ml)	2.5	5.0	7.5	10.0	12.5	15.0	
	Area	112863	228360	323609	445776	547411	659095	0.9996

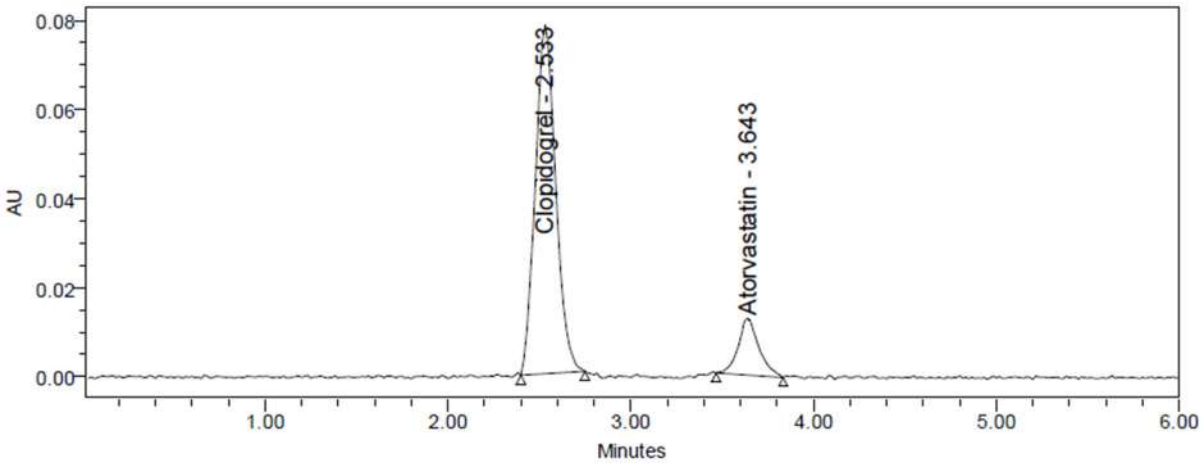

Figure-16: 25% linearity level chromatogram

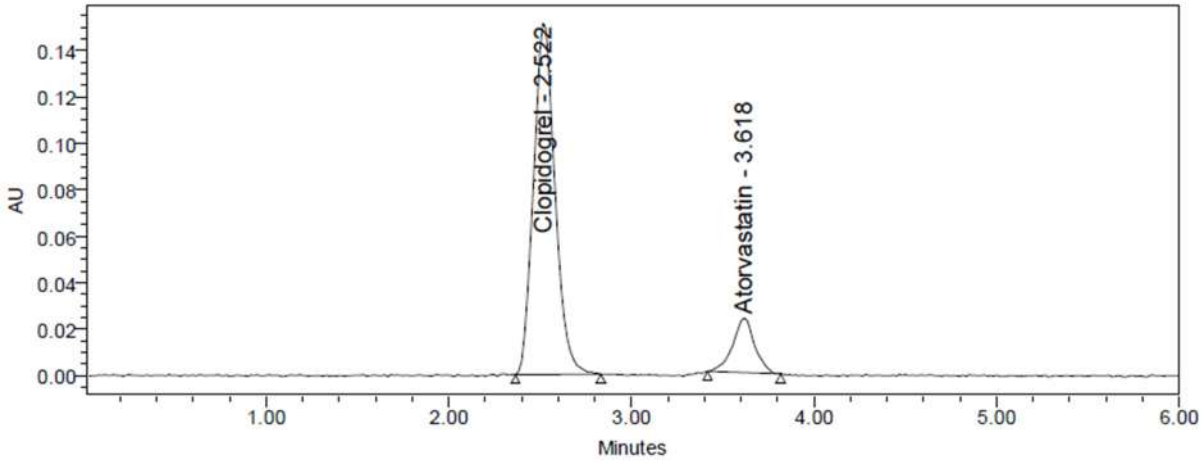

Figure17: 50% linearity level chromatogram

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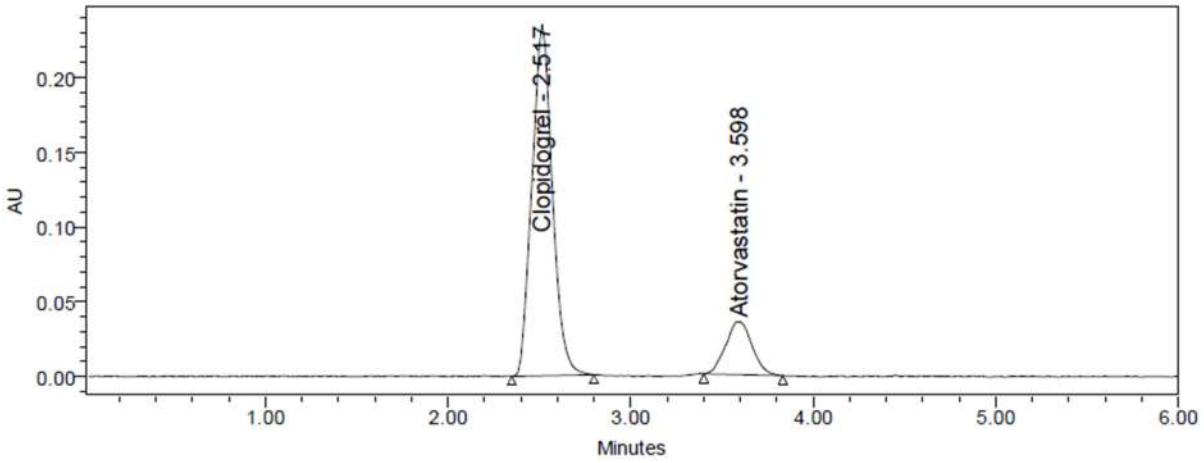

Figure-18: 75% linearity level chromatogram

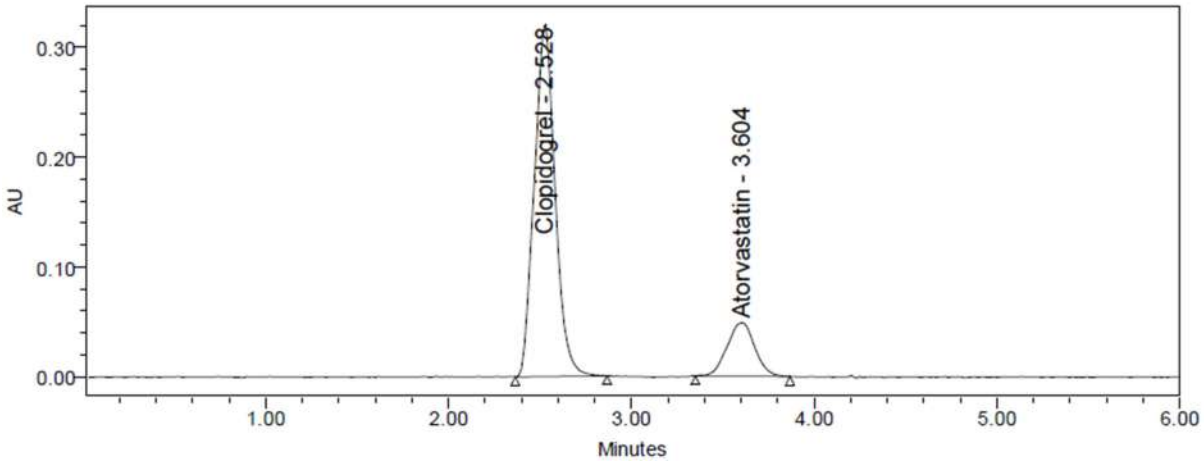

Figure-19: 100% linearity level chromatogram

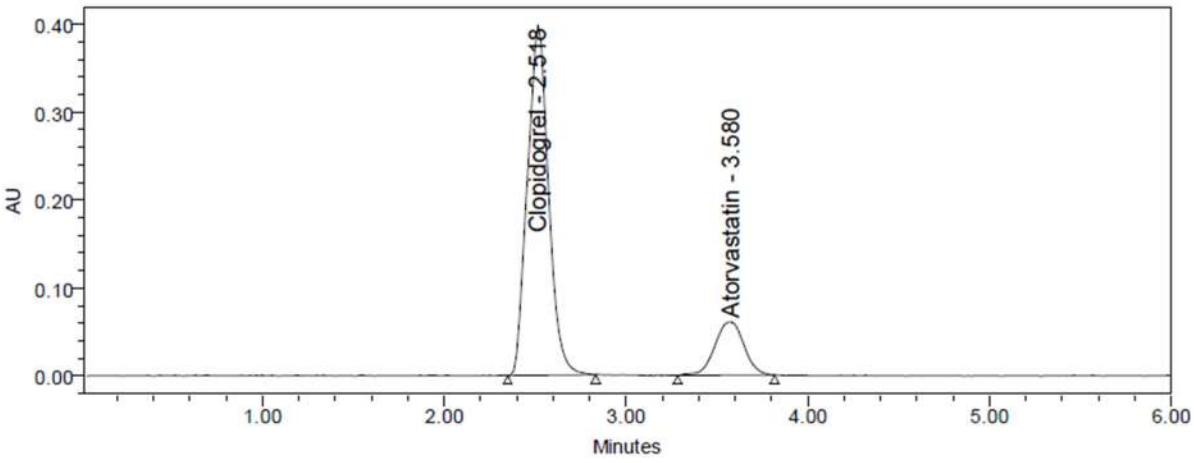

Figure-20: 125% linearity level chromatogram

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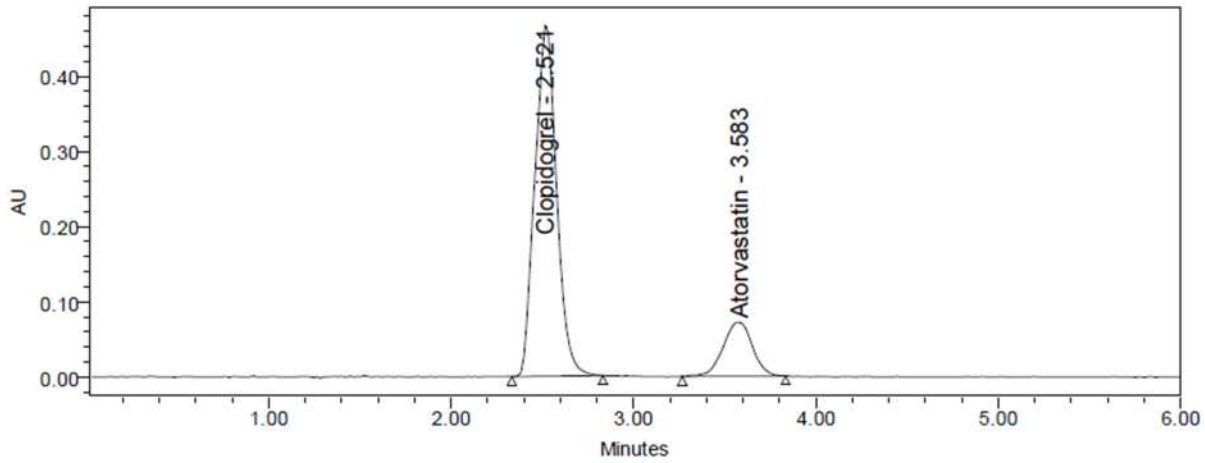

Figure-21: 150% linearity level chromatogram

Accuracy:

Method accuracy was performed with three different concentration levels such as 50% level, 100 % level and 150% level. Three different concentration levels were replicated with three preparations and % recovery was calculated. % recovery results found satisfactory and results were tabulated in below table-4.

Table-4: Accuracy results

Clopidogrel accuracy results									
Level	50%			100%			150%		
Recovery (%)	99. 77	98.78	99.38	99.71	99.23	99.86	99.30	99.28	99.33
Mean (%)	99.08			99.60			99.30		
Atorvastatin accuracy results									
Level	50%			100%			150%		
Recovery (%)	99.72	99.86	98.65	100.66	99.40	99.05	99.20	98.80	99.21
Mean (%)	99.41			99.70			99.07		

Ruggedness:

Ruggedness was evaluated with bench top and refrigerator storage conditions. Test solution stability and system suitability was performed on day-0. Day-1 and day-3 for bench top storage conditions and day-0, day-3 and day-5 for refrigerator storage conditions. Ruggedness results were tabulated in below table-5.

Table-5: Ruggedness results

Clopidogrel ruggedness results							
Time in day	**Bench top stability test solution**				**Tailing factor**	**%RSD**	**Bench top stability standard solution**
	Test-1	Test-2	Difference				Similarity factor
			Test-1	Test-2			
Initial	99.73	99.72	NA	NA	1.21	0.06	0.98
Day-1	99.68	99.75	0.05	0.03	1.26	0.08	0.99
Day-3	99.78	99.70	0.05	0.02	1.28	0.03	
	Refrigerator stability test solution						**Refrigerator stability standard solution**

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Initial	99.73	99.72	NA	NA	1.26	0.09	0.03
Day-3	99.76	99.70	0.03	0.02	1.24	0.05	0.08
Day-5	99.63	99.65	0.1	0.07	1.26	0.04	0.04
Atorvastatin ruggedness results							
Time in day	**Bench top stability test solution**				**Tailing factor**	**%RSD**	**Bench top stability standard solution**
	Test-1	Test-2	Difference				Similarity factor
			Test-1	Test-2			
Initial	99.93	99.98	NA	NA	1.09	0.06	0.99
Day-1	99.86	99.89	0.07	0.09	1.06	0.04	1.00
Day-3	99.90	99.95	0.03	0.03	1.04	0.08	
	Refrigerator stability test solution						**Refrigerator stability standard solution**
Initial	99.93	99.98	NA	NA	1.06	0.06	0.06
Day-3	99.96	99.91	0.03	0.07	1.08	0.08	0.04
Day-5	99.90	99.94	0.03	0.04	1.06	0.04	0.09

Robustness:

Robustness of the method was validated with chromatographic conditions variation and filter validation. Flow rate, column oven temperature and mobile phase solvent ratio variations were evaluated. Filter validation was conducted with PVDF and NYLON filters. Robustness results found satisfactory and results were tabulated in table-6 and 7.

Table-6: Results of Effect of variations

Variation condition		**Flow rate ml/min**			**Column temperature**		
Variation changes		0.8	1.0	1.2	25°C	30°C	35°C
Clopidogrel	Tailing factor	1.26	1.2	1.36	1.29	1.24	1.30
	% RSD	0.06	0.04	0.03	0.08	0.09	0.04
Atorvastatin	Tailing factor	1.06	1.02	1.10	1.06	1.08	1.04
	% RSD	0.09	0.08	0.06	0.05	0.04	0.09
Variation condition		**M.P organic solvent ratio**					
Variation changes		55:45	60:40	65:35			
Clopidogrel	Tailing factor	1.12	1.16	1.18			
	% RSD	0.08	0.03	0.08			
Atorvastatin	Tailing factor	1.08	1.04	1.02			
	% RSD	0.09	0.04	0.06			

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Table-7: Filter Variability results

Clopidogrel filter validation									
Centrifuged		**Nylon filter**				**PVDF filter**			
% assay		% assay		% Difference		% assay		% Difference	
Spl-1	Spl-2	Spl-1	Spl-2	Spl-1	Spl-2	Spl-1	Spl-2	Spl-1	Spl-2
99.69	99.64	99.71	99.69	0.02	0.05	99.61	99.82	0.08	0.18
Atorvastatin filter validation									
99.60	99.81	99.74	99.69	0.14	0.12	99.68	99.69	0.08	0.12

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CONCLUSION:

Simple and less run time HPLC method was developed for the determination of Clopidogrel and atorvastatin in capsule formulation. Method validation was performed with precision, linearity, accuracy, specificity, ruggedness and robustness. Method validation results found satisfactory. Optimized method can be applied for routine analysis.

REFERENCES:

1. Hiatt WR, Fowkes FG, Heizer G, Berger JS, Baumgartner I, Held P, Katona BG, Mahaffey KW, Norgren L, Jones WS, Blomster J. Ticagrelor versus clopidogrel in symptomatic peripheral artery disease. New England Journal of Medicine. 2017 Jan 5;376(1):32-40.
2. Johnston SC, Easton JD, Farrant M, Barsan W, Conwit RA, Elm JJ, Kim AS, Lindblad AS, Palesch YY. Clopidogrel and aspirin in acute ischemic stroke and high-risk TIA. New England Journal of Medicine. 2018 Jul 19;379(3):215-25.
3. Mega JL, Close SL, Wiviott SD, Man M, Duvvuru S, Walker JR, Sundseth SS, Collet JP, Delaney JT, Hulot JS, Murphy SA. PON1 Q192R genetic variant and response to clopidogrel and prasugrel: pharmacokinetics, pharmacodynamics, and a meta-analysis of clinical outcomes. Journal of thrombosis and thrombolysis. 2016 Apr 1;41(3):374-83.
4. Schulz-Schüpke S, Byrne RA, Ten Berg JM, Neumann FJ, Han Y, Adriaenssens T, Tölg R, Seyfarth M, Maeng M, Zrenner B, Jacobshagen C. ISAR-SAFE: a randomized, double-blind, placebo-controlled trial of 6 vs. 12 months of clopidogrel therapy after drug-eluting stenting. European heart journal. 2015 Jan 23;36(20):1252-63.
5. Sahlen A, Varenhorst C, Lagerqvist B, Renlund H, Omerovic E, Erlinge D, Wallentin L, James SK, Jernberg T. Outcomes in patients treated with ticagrelor or clopidogrel after acute myocardial infarction: experiences from SWEDEHEART registry. European heart journal. 2016 Jul 19;37(44):3335-42.
6. Itkonen MK, Tornio A, Neuvonen M, Neuvonen PJ, Niemi M, Backman JT. Clopidogrel markedly increases plasma concentrations of CYP2C8 substrate pioglitazone. Drug Metabolism and Disposition. 2016 Aug 1;44(8):1364-71.
7. Bays H, Gaudet D, Weiss R, Ruiz JL, Watts GF, Gouni-Berthold I, Robinson J, Zhao J, Hanotin C, Donahue S. Alirocumab as add-on to atorvastatin versus other lipid treatment strategies: ODYSSEY OPTIONS I randomized trial. The Journal of Clinical Endocrinology & Metabolism. 2015 Aug 1;100(8):3140-8.
8. Billings FT, Hendricks PA, Schildcrout JS, Shi Y, Petracek MR, Byrne JG, Brown NJ. High-dose perioperative atorvastatin and acute kidney injury following cardiac surgery: a randomized clinical trial. Jama. 2016 Mar 1;315(9):877-88.

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9. Dalli J, Chiang N, Serhan CN. Elucidation of novel 13-series resolvins that increase with atorvastatin and clear infections. Nature medicine. 2015 Sep;21(9):1071.
10. Watts GF, Chan DC, Dent R, Somaratne R, Wasserman SM, Scott R, Burrows S, R. Barrett PH. Factorial effects of evolocumab and atorvastatin on lipoprotein metabolism. Circulation. 2017 Jan 24;135(4):338-51.
11. Livingstone SJ, Looker HC, Akbar T, Betteridge DJ, Durrington PN, Hitman GA, Neil HA, Fuller JH, Colhoun HM. Effect of atorvastatin on glycaemia progression in patients with diabetes: an analysis from the Collaborative Atorvastatin in Diabetes Trial (CARDS). Diabetologia. 2016 Feb 1;59(2):299-306.
12. Koh KK. Letter by Koh Regarding Article,"Factorial Effects of Evolocumab and Atorvastatin on Lipoprotein Metabolism". Circulation. 2017 Jul 4;136(1):118-9.
13. Masud AA, Begum I. Development and validation of a RP HPLC method for simultaneous estimation of aspirine and clopidogrel in combined tablet dosage form. Int J Pharm Sci Res. 2016 Nov 1;7(11):4443-8.
14. Ramalingam Sathiyasundar and Kannappan Valliappan, Chemometric Assisted HPLC Method for the Simultaneous Estimation of Aspirin, Atorvastatin and Clopidogrel in Biological Matrix, Journal of Advances in Biology & Biotechnology 4(3): 1-10, 2015
15. Elkady EF, Tammam MH, El maaty AA. Stability Indicating HPLC-UV vs. UPLC-DAD for Estimation of Atorvastatin Simultaneously with Aspirin, Clopidogrel and their Related Impurities in Bulk and Capsules. Analytical Chemistry Letters. 2017 Sep 3;7(5):596-610.
16. Porwal PK, Ahmad RA A, Chhajed SS, Chatpalliwar VA. Liquid chromatographic method for simultaneous quantitation of clopidogrel, aspirin and atorvastatin in rat plasma and its application to the pharmacokinetic study. Journal of chromatographic science. 2015 Jan 21;53(7):1155-62.
17. Spiridon AM, Neamtu J, Belu I, Turcu-Stiolica TS, Croitoru O. Simultaneous analysis of Clopidogrel Bisulfate, Acetylsalicylic Acid and Atorvastatin calcium in tablets by HPLC method. Current health sciences journal. 2015 Apr;41(2):172.
18. Turner RM, Fontana V, Bayliss M, Whalley S, Castelazo AS, Pirmohamed M. Development, validation and application of a novel HPLC-MS/MS method for the quantification of atorvastatin, bisoprolol and clopidogrel in a large cardiovascular patient cohort. Journal of pharmaceutical and biomedical analysis. 2018 Sep 10;159:272-81.
19. R. Margret Chandira, P. Palanisamy, B. Jaykar, A. Pasupathi, B. S. Venkateshwarlu, Formulation, Development And Evaluation Of Atorvastatin, Aspirin And Clopidogrel Tablets In Capsules Form, World Journal of Pharmaceutical Research, Volume 4, Issue 10, 791-836.
20. Octavian Croitoru, Adela-Maria Spiridon, Ionela Belu, Adina Turcu-Ftiolics, and Johny Neamuu, Development and Validation of an HPLC Method for Simultaneous Quantification of Clopidogrel Bisulfate, Its Carboxylic Acid Metabolite, and Atorvastatin in Human Plasma: Application to a Pharmacokinetic Study, Journal of Analytical Methods in Chemistry, 2015, 1-12.

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