

ISSN : 2320-1304



International Journal Of Pharmamedix India.

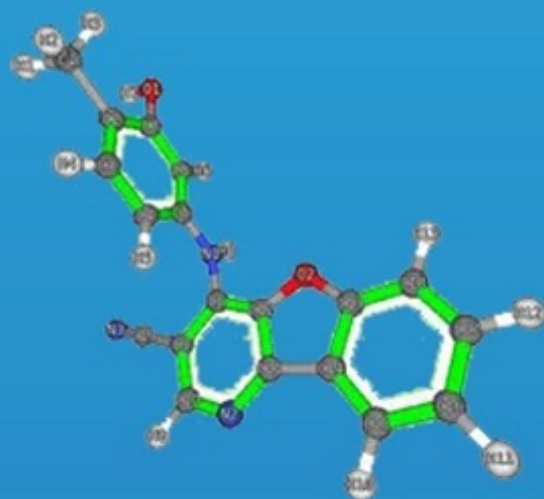
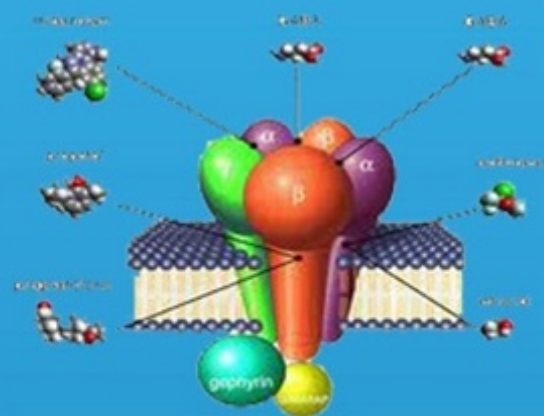
January 2013. Vol. -I Issue - I.

Research, Review, Survey.

Published by : Pharmamedix.India

www.pharmamedix.in

mail: info@pharmamedix.in



Validate the sensitivity of detector of HPLC by linearity study on Desloratadine drug.

Patel Hemant¹, Rawat Deepak, Dr. Gupta M.K., Mr. Raman Gupta, Mansoori Ahamed Noor²

Department of pharmaceutical chemistry, Department of Pharmacology,

1.Kota college of Pharmacy, SP-1,Ranpur road, Kota

2. Jaipur College of Pharmacy, Sitapura, Jaipur

Corresponding Author - Patel Hemant

E-mail- Hemupatel444@gmail.com

Contact no- 08000025811,09782045526.

Abstract:

Validation study generally to ensure the process, that detector is validate for the further process. Validation its named as specification or verification process for to confirm the process or design preactical. Desloratadine drug act as anti- histaminic, its use for the allergic condition, rhinitis, pluritis etc. Its metabolite of loratadine. By the validation study, I would like to confirm the sensitivity of the detector used in HPLC for the force degradation process. By the force degradation process, to confirm the stability of Desloratadine drug at various stress. To demonstrate the linearity of detector response for Desloratadine, prepare solutions of Desloratadine sample to achieve concentrations ranging from 80% to 120% of working test concentration. Injected all concentration solutions in duplicate into the liquid chromatograph & recorded the chromatograms. Calculated the Y-intercept, slope of regression line, correlation coefficient and R^2 value.

Introduction:

Verification and validation are independent procedures that are used together for checking that a product,

service, or system meets requirements and specifications and that it fulfills its intended purpose. These are critical components of a quality management

system such as ISO 9000. The words "verification" and "validation" are sometimes preceded with "Independent" indicating that the verification and validation is to be performed by a disinterested third-party¹.

The purpose of method validation is to demonstrate that the established method is fit for the purpose. This means that the method, as used by the laboratory generating the data, will provide data that meets the criteria set in the planning phase. The analyst's primary objective is to select an approach that will demonstrate a true validation while working in a situation with defined limitations, such as cost and time.

The method must 'fit the purpose' as agreed upon between the client and the analyst. In the case of trace analysis, the following criteria are typically evaluated as part of the method development process:

- Specificity
- Accuracy
- Repeatability
- Limit of Detection (LOD)
- Sensitivity
- Limit of Quantitation
- Linearity or Range

Robustness: Robustness testing deals with the critical operational parameters and the

tolerances for their control. These are all examples of parameters that could be easily altered, either intentionally or unintentionally, that could significantly affect the reliability of the determination. These are all examples of parameters that could be easily altered, either intentionally or unintentionally, that could significantly affect the reliability of the determination^{2,3,4}.

TYPES OF PROCESS VALIDATION

Prospective Validation

Prospective validation is conducted before a new product is released for distribution or, where the revisions may affect the product's characteristics, before a product made under a revised manufacturing process is released for distribution.

Retrospective Validation

Retrospective validation is the validation of a process based on accumulated historical production, testing, control, and other information for a product already in production and distribution. This type of validation makes use of historical data and information which may be found in batch records, production log books, lot records, control charts, test and inspection results, customer complaints or lack of complaints, field failure reports, service reports, and audit reports. Historical data must contain

enough information to provide an in-depth picture of how the process has been operating and whether the product has consistently met its specifications. Retrospective validation may not be feasible if all the appropriate data was not collected, or appropriate data was not collected in a manner which allows adequate analysis.^{5 to 9}

Force Degradation Study Forced degradation studies are used to identify reactions which may occur to degrade a processed product. Usually conducted before final formulation, forced degradation uses external stresses to rapidly screen material stabilities.

Common Stresses

There are a number of common stresses which are used to

- pH (acid/base)

Chemical processes are often catalyzed by the presence of acids and bases. The exposure of materials to these can therefore accelerate degradation reactions.

- Temperature

In accordance to arrhenius kinetics, increasing temperature increases the rate of any degradation process. Temperature is

often used in conjunction with other stresses to increase reaction rates.

- Oxidation
- Concentration
- Light

Forced degradation studies are designed to generate product-related variants and develop analytical methods to determine the degradation products formed during accelerated pharmaceutical studies and long-term stability studies. Any significant degradation product should be evaluated for potential hazard and the need for characterization and quantization.

Testing of stressed samples is required to demonstrate the following abilities of analytical techniques employed in stability studies:

- To evaluate stability of DS and DP in solution
- To determine structural transformations of the drug substance and drug product
- To detect low concentrations of potential degradation products
- To detect unrelated impurities in the presence of the desired product and product-related degradants
- To separate product-related degradants from those derived

from excipients and intact placebo¹⁰⁻¹⁸.

Instrumentation: High performance liquid chromatography is a powerful tool in analysis. High performance liquid chromatography is basically a highly improved form of column chromatography. Instead of a solvent being allowed to drip through a column under gravity, it is forced through under high pressures of up to 400 atmospheres. That makes it much faster. High-performance liquid chromatography (HPLC) is a form of liquid chromatography to separate compounds that are dissolved in solution. HPLC instruments consist of a reservoir of mobile phase, a pump, an injector, a separation column, and a detector. Compounds are separated by injecting a plug of the sample mixture onto the column. The different components in the mixture pass through the column at different rates due to differences in their partitioning behavior between the mobile liquid phase and the stationary phase.¹⁹

Literature reviews:-

- M. I. Walash, et.al (2011) have been reported that Stability-indicating micelle-enhanced spectrofluorimetric method for determination of loratadine and

desloratadine in dosage forms which describe A highly sensitive and simple spectrofluorimetric method was developed for the determination of loratadine (LRT) and desloratadine (DSL) in their pharmaceutical formulations²⁰.

- Dantu Durga Rao, et.al (2010) have been reported that A validated stability-indicating UPLC method for desloratadine and its impurities in pharmaceutical dosage forms which describe A novel stability-indicating gradient reverse phase ultra-performance liquid chromatographic (RP-UPLC) method was developed for the determination of purity of desloratadine in presence of its impurities and forced degradation products²¹.
- Rudy Bonfilio, et.al (2012) have been reported that Analytical Validation of Quantitative High-Performance Liquid Chromatographic Methods in Pharmaceutical Analysis: A Practical Approach which describe about Analytical validation is an essential component in allowing a laboratory to ensure routine

acceptable performance of analytical methods²².

- Cochran David J. , et.al (2010) have been reported that An Analysis of Grasp Force Degradation with Commercially Available Gloves which describe about Excessive force has been indicated as a causal factor in repetitive motion related occupational diseases such as carpal tunnel syndrome, tendinitis, and tenosynovitis²³.
- Nalini Kanta Sahoo, et.al (2010) have been reported that Stability Indicating Assay of Niflumic acid Using RP-HPLC which describe on study describes with the development and subsequent validation of a stability indicating reverse phase HPLC (RP-HPLC) method for the determination of Niflumic acid API (Active Pharmaceutical Ingredient)²⁴.

S N o.	Instruments	Instrument ID
1	HPLC–Dionex with DAD-Ultimate 3000	C-ARL/HPLC-017
2	HPLC–Dionex with DAD-UVD340U	C-ARL/HPLC-008
3	Balance- Mettler Toledo AX105 series, Readability: 0.01mg	C-ARL/MET-BAL-02
4	pH meter-Lab India PHAN	C-ARL/pHM-02

Method:

Table 1 :-Instrument used:

Table 2: Chemicals used:

S. N.	Chemicals	Make
1	Purified water	Milli-Q water
2	Acetonitrile (HPLC grade)	Merck
3	Sodium dihydrogen orthophosphate monohydrate (AR Grade)	Merck
4	1-Pentane Sulphonic Acid Na Salt monohydrate (HPLC Grade)	Spectrochem
5	Triethyl Amine (HPLC Grade)	Merck
6	Orthophosphoric Acid (HPLC Grade)	Merck

Table 3:- Column used:

S N	Columns	Make
1	Zorbax SBC8 (150 x 4.6mm), 5µm Column ID: ARL/CSBU/C8/0038	Agilent

2	Zorbax SBC8 (150 x 4.6mm), 5µm Column ID: ARL/CSBU/C8/0044	Agilent
---	---	---------

Desloratadine Standard Preparation:

Accurately weighed and transferred 50.4mg of Desloratadine working standard in to 50.0mL volumetric flask, added about 20mL of diluent and sonicated to dissolve the solids and made the volume up to the mark with diluent, mix well. Further diluted to this solution 10.0mL to 50.0mL with diluent and mixed well.

Linearity stock solution-1:

Accurately weighed and transferred 50.3mg of test sample in to a 50.0mL volumetric flask, added about 20mL of diluent and sonicated to dissolve the solids and made the volume up to the mark with diluent, mix well. Further diluted to this solution 10.0mL to 50.0mL with diluent and mixed well.

Linearity solution preparation:

The linearity solutions at various levels are prepared from stock solution-1 using diluent as follows:

Table: 4 linearity solution preparation table

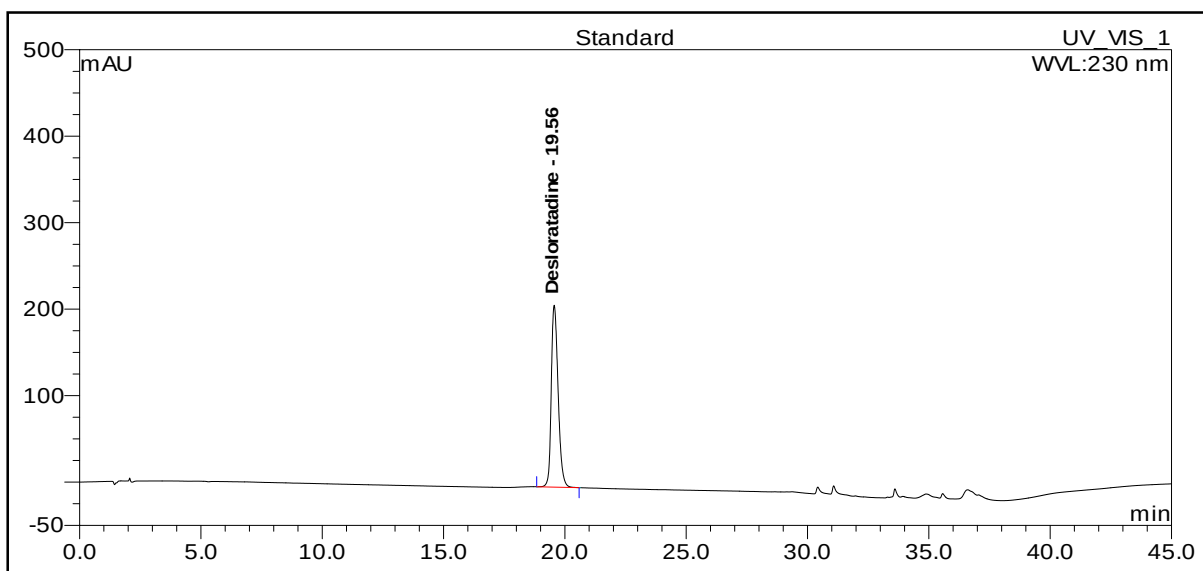
Linearity level	Stock solution-1 taken in mL	Final volume in diluent (mL)	Final concentration of Desloratadine in mcg/mL
80%	8.0	50.0	160.155
90%	9.0	50.0	180.175
100%	10.0	50.0	200.194
110%	11.0	50.0	220.213
120%	12.0	50.0	240.233

Observation:-For Linearity Study

Table: 5 Standard Preparation Solution-for Linearity study:

No. of Inj.	Desloratadine	
	R.T.	Peak Area
1	19.56	68.292
2	19.56	68.617
3	19.56	68.080
4	19.57	68.552
5	19.56	67.568
6	19.57	68.293
Mean	19.56	68.234
SD	0.00516	0.37998
%RSD	0.03	0.56

Acceptance Criteria: %RSD of peak area due to Desloratadine peak in to the standard preparation should not be more than 2.00.



No.	Ret.Time	Peak Name	Area	Rel.Area
	Min		mAU*min	%
1	19.56	Desloratadine	68.617	100.00
Total:			68.617	100.000

[Fig: -1 Schematic diagram shows HPLC graph for Linearity study]

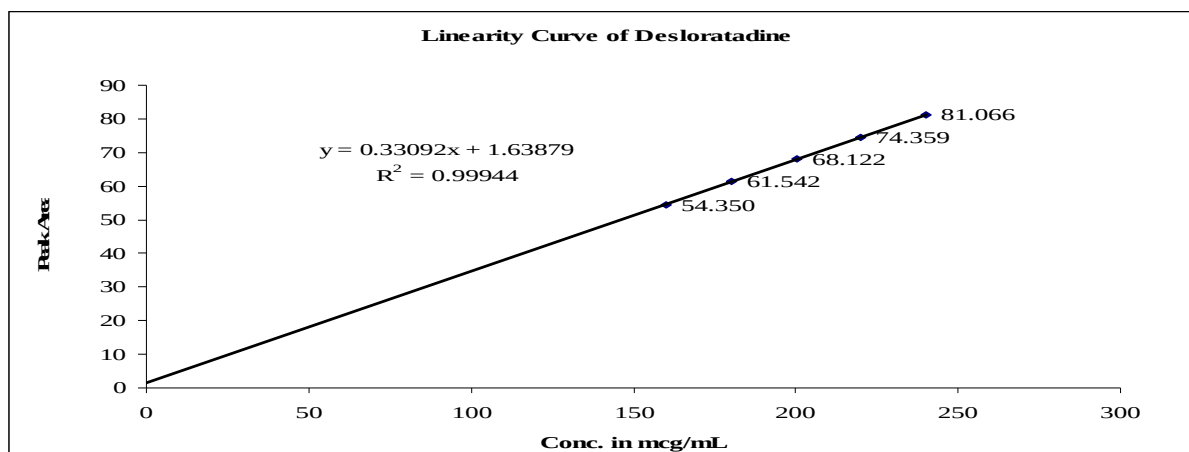
Linearity Observation:

The linear response for Desloratadine of different concentration were determined and found to be linear in the concentration range shown in following table

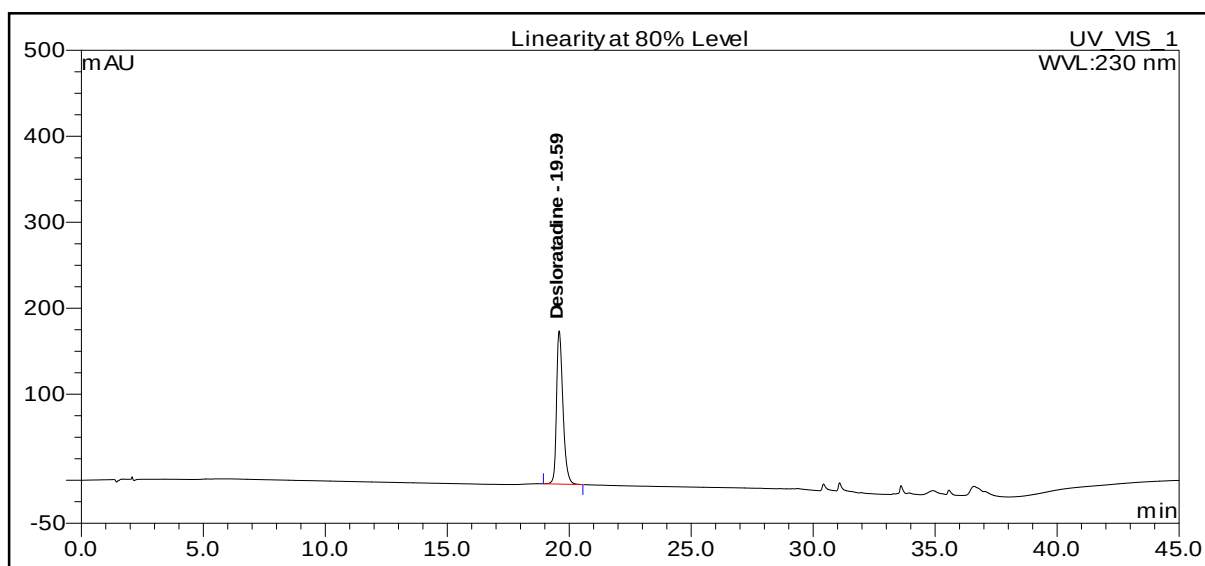
Table:6 Standard Preparation Solution at different concentration-for Linearity study

Linearity Level	Concentration in mcg/mL	Peak area		
		Rep-1	Rep-2	Mean
80%	160.155	54.189	54.510	54.350
90%	180.175	61.392	61.691	61.542
100%	200.194	68.172	68.072	68.122
110%	220.213	74.771	73.946	74.359
120%	240.233	81.714	80.417	81.066
y-Intercept				1.63879
Slope				0.33092
R ²				0.99944
Correlation coefficient				0.99972

Acceptance criteria: The correlation coefficient should not be less than 0.999

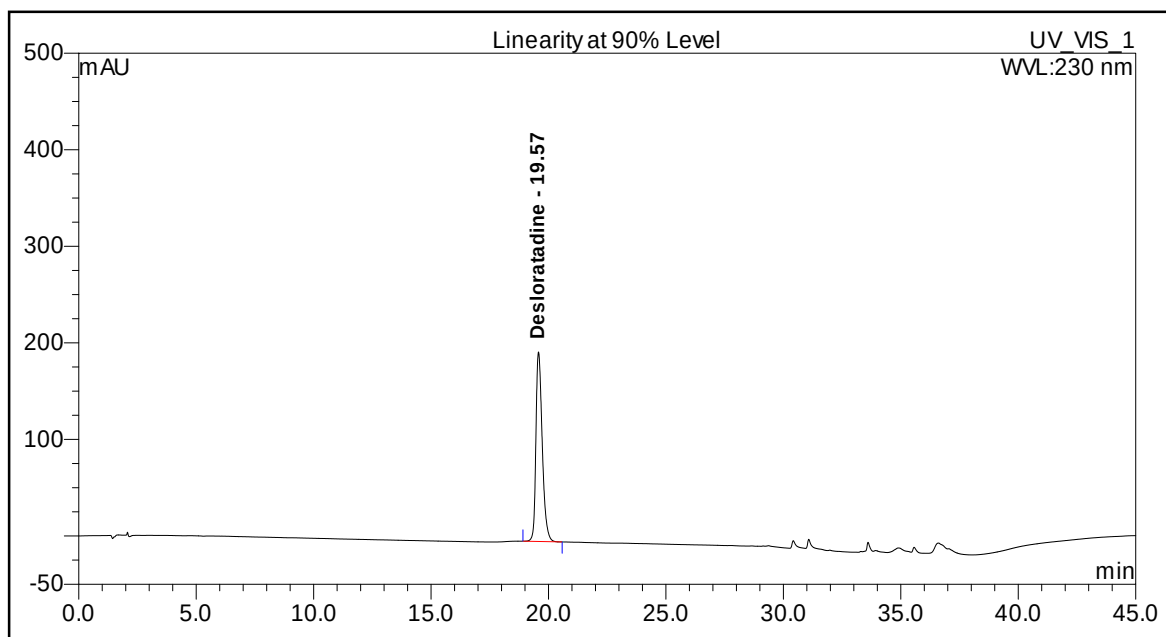


[Fig: - 2 Schematic diagram shows Linearity Curve of Desloratadine for Linearity study]



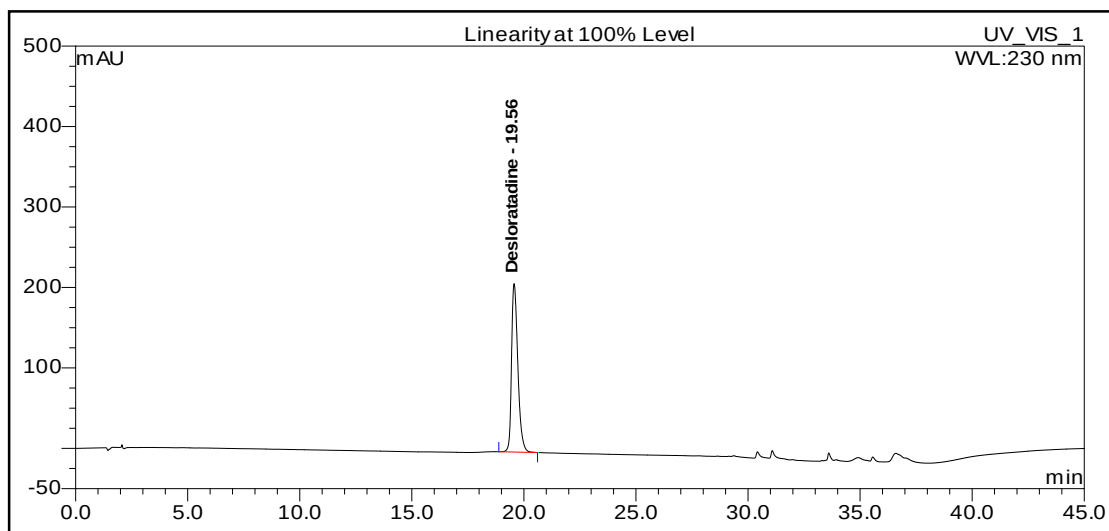
No.	Ret.Time Min	Peak Name	Area mAU*min	Rel.Area %
1	19.59	Desloratadine	54.189	100.00
Total:			54.189	100.000

[Fig: -3 Schematic diagram shows HPLC graph for Linearity study at linearity 80% concentration level]



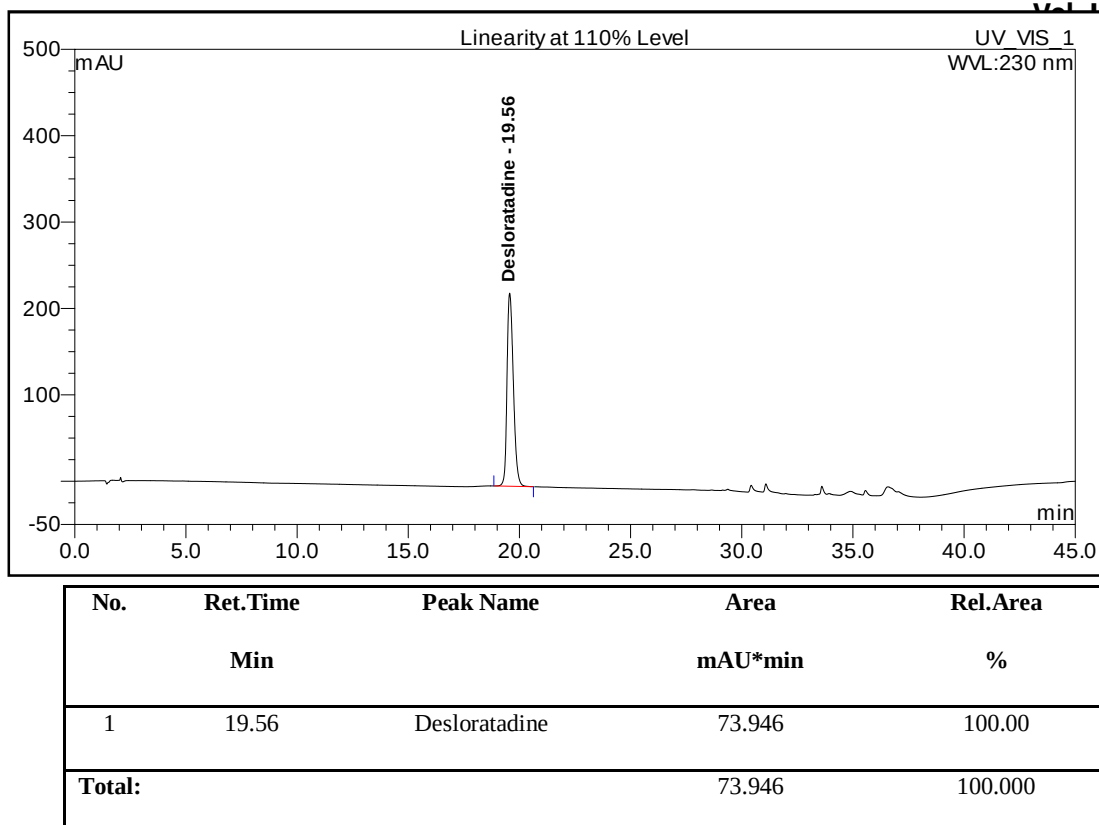
No.	Ret.Time	Peak Name	Area	Rel.Area
	Min		mAU*min	%
1	19.57	Desloratadine	61.392	100.00
Total:			61.392	100.000

[Fig: 4- Schematic diagram shows HPLC graph for Linearity study at linearity 90% concentration level]

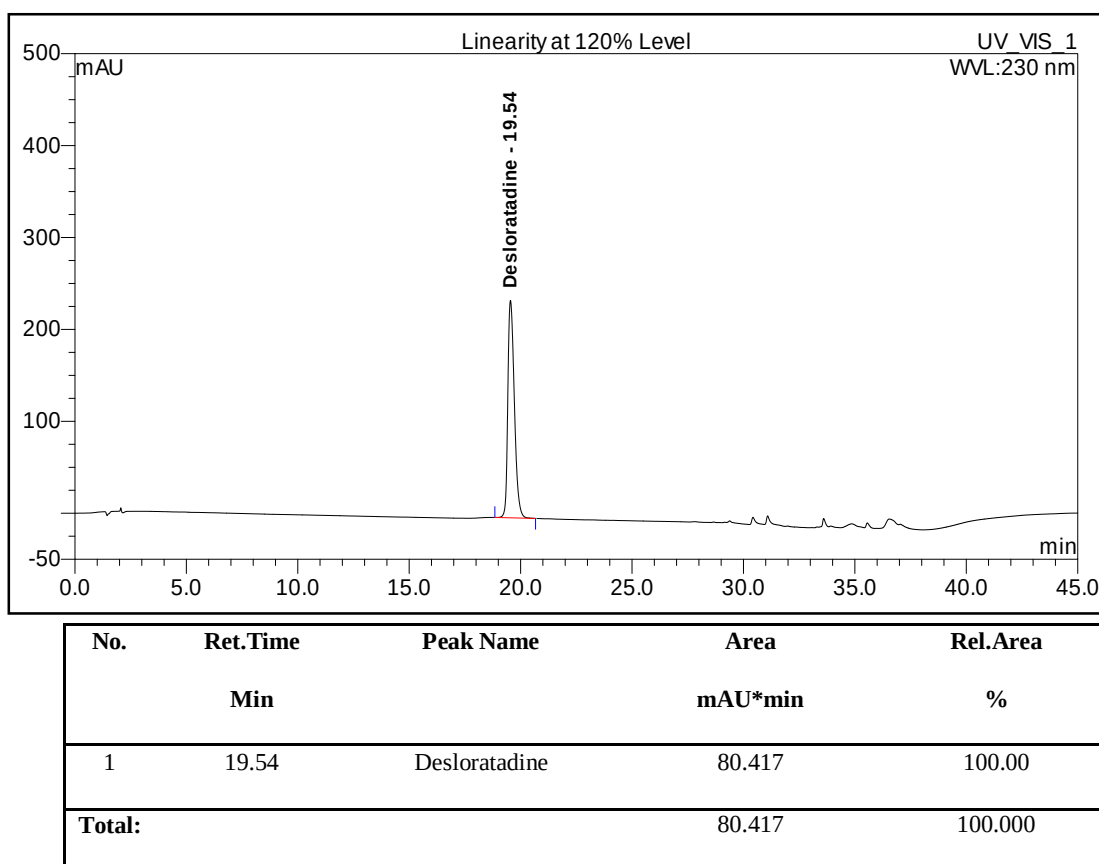


No.	Ret.Time	Peak Name	Area	Rel.Area
	Min		mAU*min	%
1	19.56	Desloratadine	68.072	100.00
Total:			68.072	100.000

[Fig: 5- Schematic diagram shows HPLC graph for Linearity study at linearity 100% concentration level]



[Fig: 6- Schematic diagram shows HPLC graph for Linearity study at linearity 110% concentration level]



[Fig: 7- Schematic diagram shows HPLC graph for Linearity study at linearity 120% concentration level]

Result & Summary:

- The correlation coefficient for Desloratadine is 0.99972.
- From results it is observed that the Y-intercept, slope of regression line and R² value for Desloratadine are 1.63879, 0.33092 and 0.99944 respectively.

From data it is concluded that method for Desloratadine is linear between the ranges of 80% to 120% level.

Conclusion:

This analytical performance parameter is checked as per the approved validation protocol and found to be well within specified acceptance criteria. Hence it is concluded that this method is capable to produce the accurate and precise result during routine analysis.

References:

1. Global Harmonization Task Force - Quality Management Systems - Process Validation Guidance (GHTF/SG3/N99-10:2004 (Edition 2) page 3.
2. Guideline on General Principles of Process Validation, May 1987, FDA, CDRH/CDER
3. Journal of Validation Technology, Vol. 1, No. 4, August 1995
4. <http://www.esslab.com/iv/tech/IV-reliable-measurements/part17.html>
5. "Guideline on general principles of process validation". U.S. Food and Drug Administration. Retrieved 12 July 2008.
6. "Prospective validation". Groupe Novasep. Retrieved 24 September 2008.
7. Quinn, James et al.; McDermott, D; Stiell, I; Kohn, M; Wells, G (2006). "Prospective Validation of the San Francisco Syncope Rule to Predict Patients With Serious Outcomes". *Annals of Emergency Medicine (Elsevier)* **47** (5): 448–454. doi:10.1016/j.annemergmed.2005.11.019. PMID 16631985.
8. Sangiovanni, A. et al.; Manini, M; Iavarone, M; Fraquelli, M; Forzenigo, L; Romeo, R; Ronchi, G; Colombo, M (2007). "Prospective validation of AASLD guidelines for the early diagnosis of hepatocellular carcinoma in cirrhotic patients". *Digestive and Liver Disease (Elsevier)* **40** (5): A22–A23. doi:10.1016/j.dld.2007.12.064.
9. Germing, U. et al.; Strupp, C; Kuendgen, A; Isa, S; Knipp, S; Hildebrandt, B; Giagounidis, A; Aul, C et al. (2006). "Prospective validation of the WHO proposals for the classification of myelodysplastic syndromes". *Haematologica* **91** (12): 1596–1604. PMID 17145595. Retrieved 24 September 2008.
10. http://en.wikipedia.org/wiki/Forced_degradation

11. ICH. Guidance for Industry, Q6B. "Specifications: Test Procedures and Acceptance Criteria for Biotechnological/Biological Products." ICH-Q6B. 1999 August 18.
12. ICH. Guidance for Industry, Q5C. "Quality of Biotechnological Products: Stability Testing of Biotechnological/Biological Products." ICH-Q5C. 1996 July 10.
13. FDA. Guidance for Industry. "INDs for Phase 2 and 3 Studies of Drugs, Including Specified Therapeutic Biotechnology-Derived Products." Draft Guidance, 1999 February.
14. ICH. Guidance for Industry. "Q1A Stability Testing of New Drug Substances and Products" ICH-Q1A. 2001 August.
15. ICH. "Final Guidance on Stability Testing of Biotechnological/Biological Products; Availability." Federal Register 61FR p. 36466-9. 1996.
16. FDA. Guidance for Industry. "Analytical Procedures and Methods Validation: Chemistry, Manufacturing, and Controls Documentation", Draft Guidance, 2000 August.
17. ICH. Guidance for Industry, Q3A "Impurities in New Drug Substances." ICH-Q3A. 2003 February 11.
18. ICH. Guidance for Industry, Q3B(R). "Impurities in New Drug Products." ICH-Q3B. 2003 November 14.
19. "HPLC instrumentation"<http://www.chemguide.co.uk/analysis/chromatography/hplc.html>
20. M. I. Walash, F. Belal, N. El-Enany, M. Eid, and R. N. El-Shaheny, "Stability-indicating micelle-enhanced spectrofluorimetric method for determination of loratadine and desloratadine in dosage forms" Luminescence, Volume 26, Issue 6, November/December 2011, pages 670–679
21. Dantu Durga Rao, N.V. Satyanarayana, A. Malleswara Reddy, Shakil S. Sait, Dinesh Chakole, and K. Mukkanti, "A validated stability-indicating UPLC method for desloratadine and its impurities in pharmaceutical dosage forms", Journal of Pharmaceutical and Biomedical Analysis, Volume 51, Issue 3, 5 February 2010, Pages 736–742
22. Rudy Bonfilio, Edith Cristina Laignier Cazedey, Magali Benjamim de Araújo & Hérica Regina Nunes Salgado, "Analytical Validation of Quantitative High-Performance Liquid-Chromatographic Methods in Pharmaceutical Analysis: A Practical Approach" Critical Reviews in Analytical Chemistry, Volume(1), 2012, pages 87-100
23. David J. Cochran, Thomas J. Albin, Ram R. Bishu and Michael W. Riley, "An Analysis of Grasp Force Degradation with

Commercially Available Gloves” ,Vol 3,
No 12 (2010).

24. Nalini Kanta Sahoo, Madhusmita Sahu,
Siba Prasad pradhan and Alok Kumar
Moharana, “Stability Indicating Assay of
Niflumic acid Using RP-HPLC”, Journal
of Pharmacy Research, Vol 3, No 12
(2010)