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Research Article

Development and validation of simple simultaneous analysis for olanzapine and samidorphan by reverse phase high performance liquid chromatography in table dosage form

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ABSTRACT

A simple, Accurate, precise method was developed for the simultaneous estimation of the Olanzapine and Samidorphan in bulk and pharmaceutical dosage form. Chromatogram was run through Std Zorbax 150 x 4.6 mm, 5µm. Mobile phase containing Buffer 0.01N Sodium hydrogen phosphate: Acetonitrile taken in the ratio 60:40 %v/v was pumped through column at a flow rate of 1.0 ml/min. Buffer used in this method was 0.01N Na2hpo4 buffer. Temperature was maintained at 30°C. Optimized wavelength selected was 268.0 nm. Retention time of Olanzapine and Samidorphan were found to be 2.235 min and 2.784 min. %RSD of the Olanzapine and Samidorphan were and found to be 0.4% and 0.7% respectively. %Assay was obtained as 99.19% and 99.81% for Olanzapine and Samidorphan respectively. %Recovery was obtained as 99.49% and 99.49% for Olanzapine and Samidorphan respectively. LOD, LOQ values obtained from regression equations of Olanzapine and Samidorphan were 0.21, 0.63 and 0.09, 0.23 respectively. Regression equation of Samidorphan is y = 31381x + 3743.6, y = 34206x + 4549.1of Olanzapine. Retention times were decreased and that run time was decreased, so the method developed was simple and economical that can be adopted in regular Quality control test in Industries.

Keywords: Samidorphan, Olanzapine, RP-HPLC

INTRODUCTION

Olanzapine and Samidorphan ia s Antipsychotic Drugs. It belongs to Second Generation: Serotonin-Dopamine Activity Modulators; Antimanic Agents. Both The Drugs Are Available in The Brand Name of Lybalvi. It Is Available in The Dosage Form Of 5mg/10mg, 10mg/10mg, 15mg/10mg, 20mg/10mg. A combination of olanzapine and samidorphan administered as a single tablet wa s recently approved by the US food drug administration for the treatment of patient with schizophrenia or bipolar-1 disorder in adults. Schizophrenia is a chronic severe, debilitating mental illness characterized by disorder thoughts, abnormal behaviors and anti-social behaviors. It is a psychotic disorder it means the person with schizophrenia does not identify with reality at

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times. Schizophrenia effects about 1.1% of worlds population. It is most commonly diagnosed between the age of 16 to 25. It can be hereditary. Olanzapine is used to treat certain mental and mood conditions. It may also used in combination with other medications to treat depression. This medication can help to decrease the hallucinations. The primary goal of this work is to provide an efficient, quick and accurate RP-HPLC approach for estimating olanzapine and samidorphan in medical dosage form. According to ICH recommendations, A proven approach was also used to estimate the amounts of olanzapine and samidorphan.

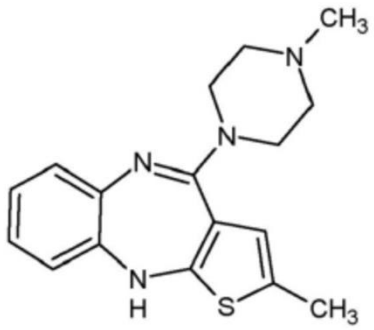


Fig1: Structure of Olanzapine

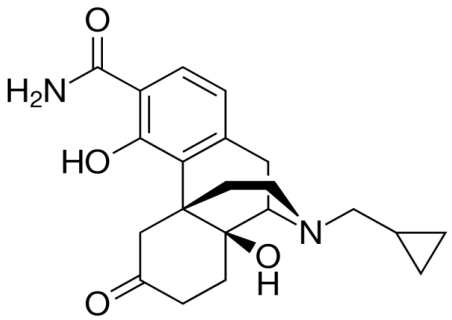


Fig2: Structure of Samidorphan

MATERIALS AND REAGENTS

Olanzapine and Samidorphan pure drugs (API), Combination Olanzapine and Samidorphan (Lybalvi), Distilled water, Acetonitrile, Phosphate buffer, Methanol, Potassium dihydrogen ortho phosphate buffer, Ortho-phosphoric acid.

Instruments:

- Electronics Balance-Denver
- pH meter -BVK enterprises, India
- Ultrasonicator-BVK enterprises
- WATERS HPLC 2695 SYSTEM equipped with quaternary pumps, Photo Diode Array detector and Auto sampler integrated with Empower 2 Software.
- UV-VIS spectrophotometer PG Instruments T60 with special bandwidth of 2 mm and 10mm and matched quartz cells integrated with UV win 6 Software was used for measuring absorbances of Olanzapine and Samidorphan solutions.

Methods:

Diluent: Based up on the solubility of the drugs, diluent was selected, Acetonitrile and Water taken in the ratio of 50:50.

Preparation of Standard stock solutions: Accurately weighed 5 mg of Samidorphan, 10mg of Olanzapine and transferred to 50ml volumetric flasks and 3/4 th of diluents was added to these flask and sonicated for 10 minutes. Flask were made up with diluents and labeled as Standard stock solution. 100µg/ml of Samidorphan and 200µg/ml Olanzapine)

Preparation of Standard working solutions (100% solution): 1ml from each stock solution was pipetted out and taken into a 10ml volumetric flask and made up with diluent. (10µg/ml of Samidorphan and 20µg/ml of Olanzapine)

Preparation of Sample stock solutions: 10 tablets were weighed and equivalent to 1 tablet is weighed and transferred to 100 ml volumetric flask, to this 5 ml of acetonitrile was added and sonicated. Volume was made upto 50ml with diluents and filtered through 0.45 µm or finer porosity membrane filter (100µg/ml of Samidorphan and 200µg/ml of Olanzapine).

Preparation of Sample working solutions (100% solution): 1ml of filtered sample stock solution was transferred to 10ml volumetric flask and made up with diluent. (10µg/ml of Samidorphan and 20µg/ml of Olanzapine)

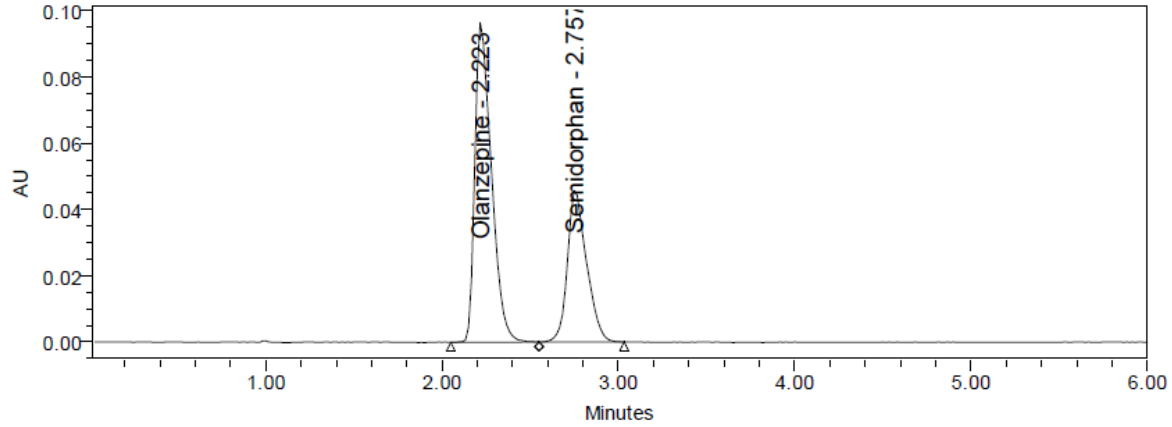
Method Validation:

System suitability parameters: The system suitability parameters were determined by preparing standard solutions of Samidorphan (10ppm) and Olanzapine (20ppm) and the solutions were injected six times and the parameters like peak tailing, resolution and USP plate count were determined. The % RSD for the area of six standard injections results should not be more than 2%.

Specificity: Checking of the interference in the optimized method. We should not find interfering peaks in blank and placebo at retention times of these drugs in this method. So this method was said to be specific.

Table: 1 System suitability parameters for Olanzapine and Samidorphan

S no	Olanzapine			Samidorphan			
Inj	RT (min)	USP Plate Count	Tailing	RT (min)	USP Plate Count	Tailing	RS
1	2.217	2458	1.43	2.737	2738	1.23	2.7
2	2.218	2419	1.41	2.742	2733	1.23	2.6
3	2.218	2412	1.43	2.750	2739	1.21	2.6
4	2.221	2342	1.44	2.753	2755	1.21	2.7
5	2.223	2369	1.43	2.757	2795	1.24	2.7
6	2.226	2431	1.42	2.766	2750	1.23	2.7



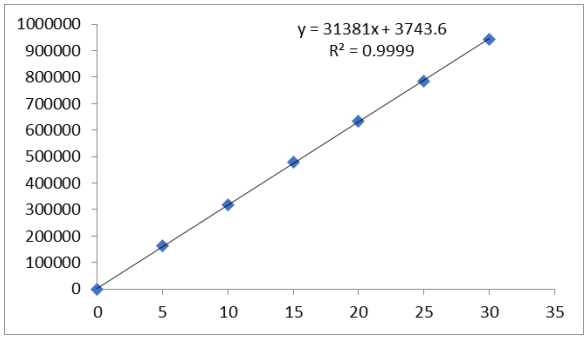
Typical Chromatogram

Discussion: Retention times of Olanzapine and Samidorphan were 2.223 min and 2.752 min respectively. We did not found and interfering peaks in blank and placebo at retention times of these drugs in this method. So this method was said to be specific.

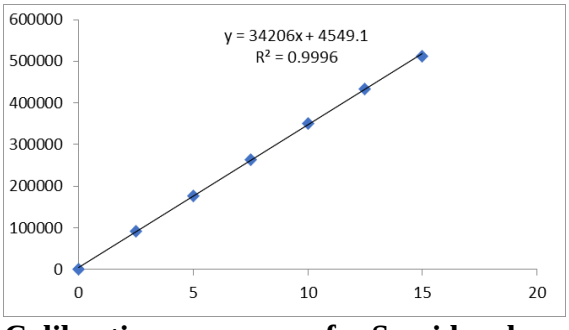
Linearity:

Table 2 : Linearity table for Olanzapine and Samidorphan

Olanzapine		Samidorphan	
Conc (µg/mL)	**Peak area**	**Conc (µg/mL)**	**Peak area**
0	0	0	0
5	163090	2.5	92029
10	316562	5	175574
15	478718	7.5	263323
20	633932	10	351412
25	784529	12.5	433302
30	944352	15	512006



Calibration curve of Olanzapine

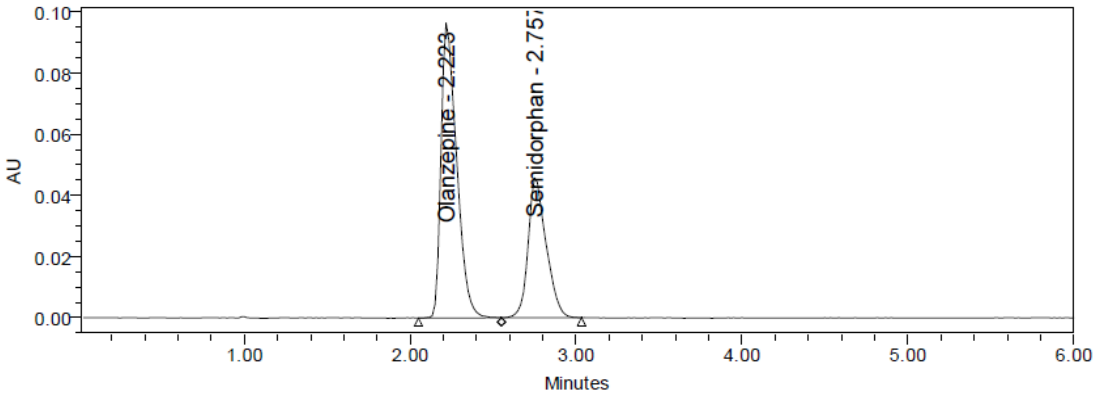


Calibration curve of Samidorphan

System Precision:

Table 3: System precision table of Olanzapine and Samidorphan

S. No	Area of Olanzapine	Area of Samidorphan
1.	629584	345257
2.	630377	345860
3.	630760	340303
4.	624480	344948
5.	629782	345210
6.	629195	346742
Mean	629030	344720
S.D	2298.2	2257.4
%RSD	0.4	0.7

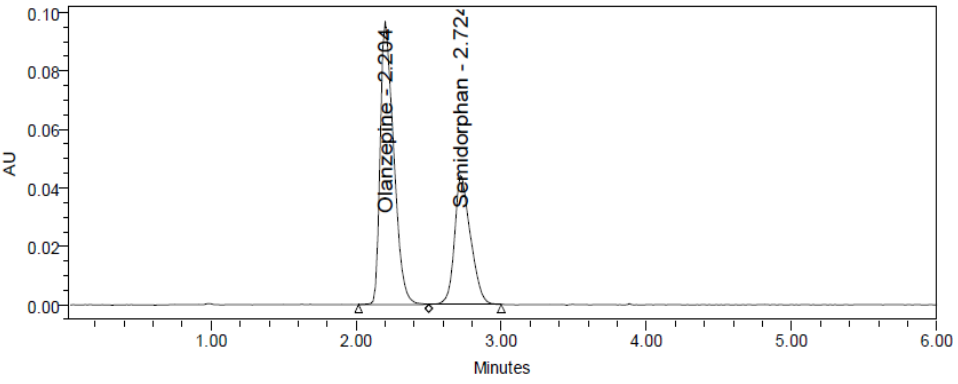


System precision chromatogram

Method precision:

Table 4. Method precision table of Olanzapine and Samidorphan

S. No	Area of Olanzapine	Area of Samidorphan
1.	622966	346117
2.	627174	345529
3.	626289	344495
4.	624321	346224
5.	624021	344685
6.	626180	342132
Mean	625159	344864
S.D	1624.0	1516.2
%RSD	0.3	0.4



Method precision chromatogram

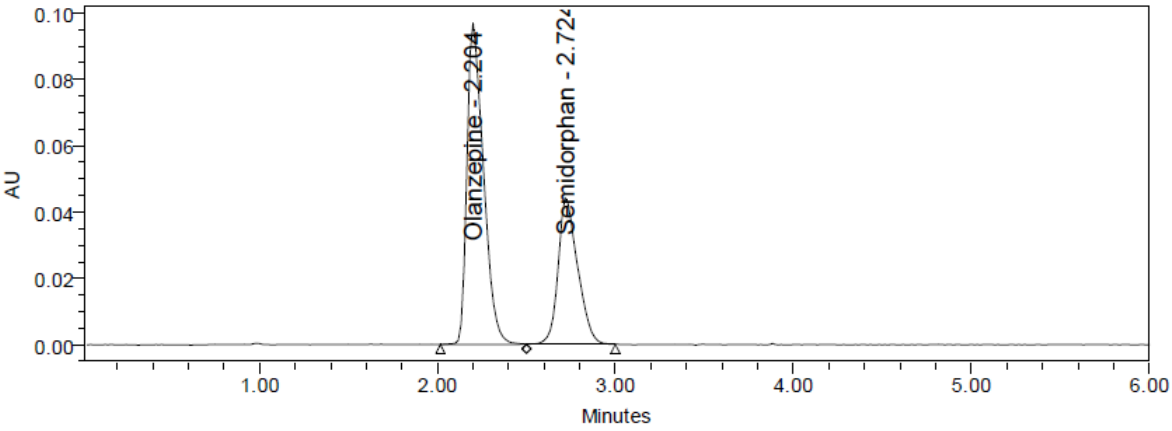
% RSD were calculated for two drugs (Olanzapine and Samidorphan) and obtained as 0.3% and 0.4% respectively. As the limit of Precision was less than "2" the Method precision was passed in this method.

Discussion: Multiple sampling from a sample stock solution was done and six working sample solutions of same concentrations were prepared, each injection from each working sample solution was given and obtained areas were mentioned in the above table. Average area, standard deviation and % RSD were calculated for two drugs and obtained as 0.3% and 0.4% respectively for Olanzapine and Samidorphan. As the limit of Precision was less than "2" the Method precision was passed in this method.

Intermediate precision (Day_ Day Precision):

Table 5: Intermediate precision table of Olanzapine and Samidorphan

S. No	Area of Samidorphan	Area of Olanzapine
1.	626453	340109
2.	619220	340616
3.	629678	343660
4.	629912	343449
5.	618899	343558
6.	621932	341109
Mean	624349	342084
S.D	5012.4	1644.7
%RSD	0.8	0.5



Intermediate precision Chromatogram

Discussion: Multiple sampling from a sample stock solution was done and six working sample solutions of same concentrations were prepared, each injection from each working sample solution was given on the next day of the sample preparation and obtained areas were mentioned in the above table. Average area, standard deviation and % RSD were calculated for two drugs and obtained as 0.8% and 0.5% respectively for Olanzapine and Samidorphan. As the limit of Precision was less than "2" the Intermediate precision was passed in this method.

Accuracy:

Table 6 : Accuracy table of Olanzapine

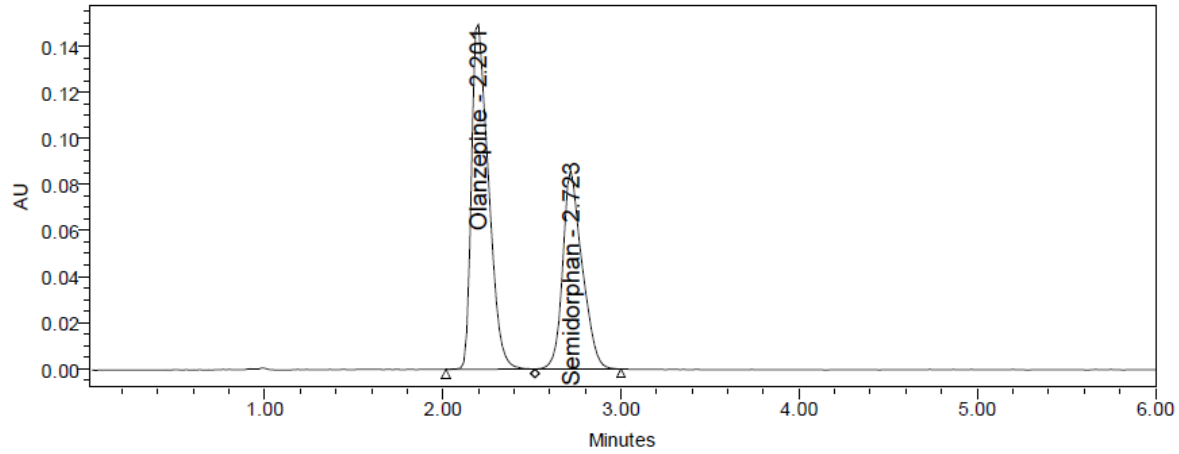
% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean %Recovery
50%	10	9.93	99.27	99.49%
	10	9.97	99.65	
	10	9.98	99.81	
100%	20	20.19	100.93	
	20	19.70	98.50	
	20	19.88	99.40	

150%	30	29.8	99.4	
	30	29.5	98.3	
	30	30.0	100.1	

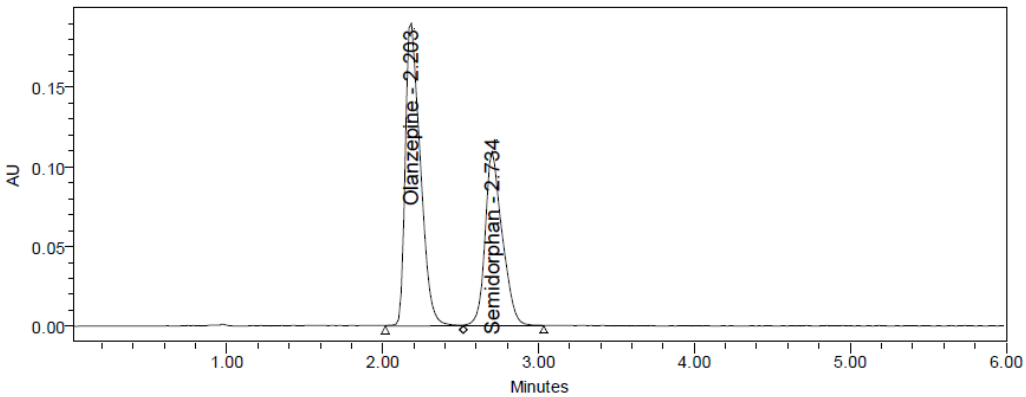
Table 7: Accuracy table of Samidorphan

% Level	Amount Spiked (µg/mL)	Amount recovered (µg/mL)	% Recovery	Mean %Recovery
50%	5	5.01	100.25	99.49%
	5	4.95	98.94	
	5	4.98	99.55	
100%	10	9.96	99.58	
	10	9.90	99.00	
	10	9.88	98.83	
150%	15	15.09	100.62	
	15	14.87	99.14	
	15	14.93	99.54	

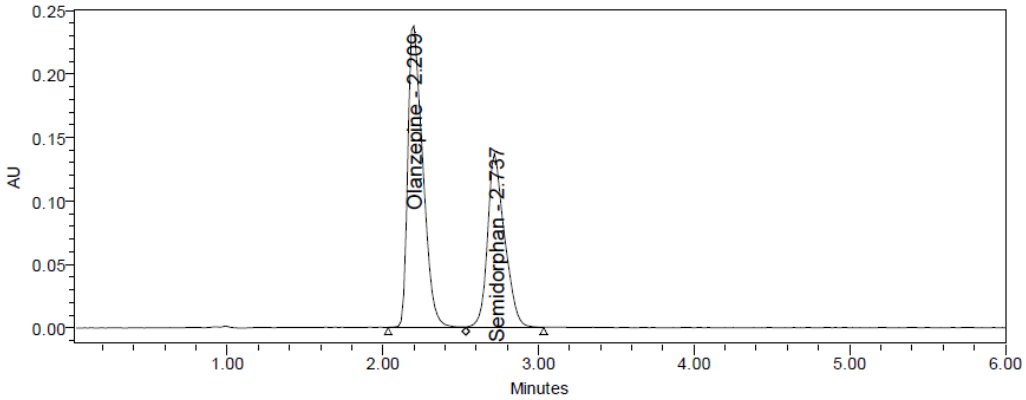
Discussion: Three levels of Accuracy samples were prepared by standard addition method. Triplicate injections were given for each level of accuracy and mean %Recovery was obtained as 99.49% and 99.49% for Olanzapine and Samidorphan respectively.



Accuracy 50% Chromatogram of Olanzapine and Samidorphan



Accuracy 100% Chromatogram of Olanzapine and Samidorphan



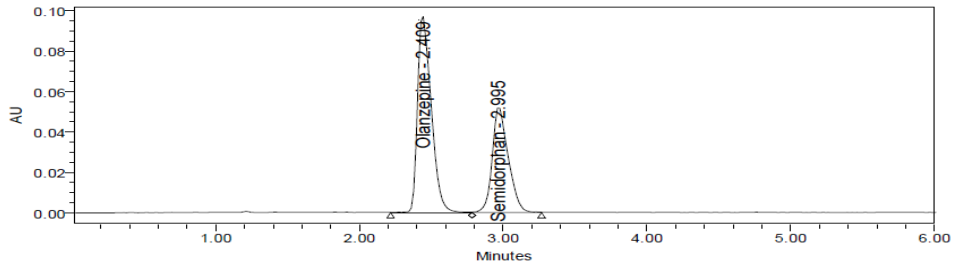
Accuracy 150% Chromatogram of Olanzapine and Samidorphan

Robustness:

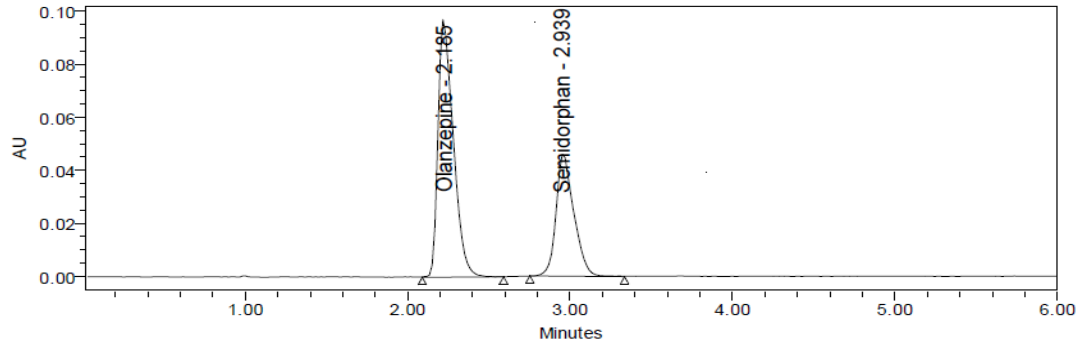
Table 8 : Robustness data for Olanzapine and Samidorphan.

S.no	Condition	%RSD of Olanzapine	%RSD of Samidorphan
1	Flow rate (-) 0.9ml/min	0.2	1.3
2	Flow rate (+) 1.1ml/min	0.3	0.7
3	Mobile phase (-) 65B:35A	0.6	0.8
4	Mobile phase (+) 55B:45A	0.4	1.0
5	Temperature (-) 27°C	0.6	0.8
6	Temperature (+) 33°C	0.3	0.4

Discussion: Robustness conditions like Flow minus (0.9ml/min), Flow plus (1.1ml/min), mobile phase minus (65B:35A), mobile phase plus (55B:45A), temperature minus (27°C) and temperature plus(33°C) was maintained and samples were injected in duplicate manner. System suitability parameters were not much affected and all the parameters were passed. %RSD was within the limit.



Flow minus Chromatogram of Olanzapine and Samidorphan



Flow plus Chromatogram of Olanzapine and Samidorphan

ASSAY: (Lybalvi) Bearing the label claims Samidorphan 10mg, Olanzapine 20mg. Assay was performed with the above formulation. Average % Assay for Olanzapine and Samidorphan obtained was 99.95%and 100.75% respectively.

Table 9: Assay Data of Olanzapine

S.no	Standard Area	Sample area	% Assay
1	629584	622966	98.84
2	630377	627174	99.51
3	630760	626289	99.37
4	624480	624321	99.05
5	629782	624021	99.01
6	629195	626180	99.35
Avg	629030	625159	99.19
Stdev	2298.2	1624.0	0.26
%RSD	0.4	0.3	0.3

Table 10: Assay Data of Samidorphan

S.no	Standard Area	Sample area	% Assay
1	345257	346117	100.00
2	345860	345529	100.03
3	340303	344495	99.73
4	344948	346224	100.24
5	345210	344685	99.79
6	346742	342132	99.05
Avg	344720	344864	99.81
Stdev	2257.4	1516.2	0.4
%RSD	0.7	0.4	0.4

Degradation Studies: Degradation studies were performed with the formulation and the degraded samples were injected. Assay of the injected samples was calculated and all the samples passed the limits of degradation.

Table 11 : Degradation Data of Olanzapine

S.NO	Degradation Condition	Area	% Drug Degraded	% Drug Undegraded
1	Acid	590567	93.70	6.30
2	Alkali	602911	95.66	4.34
3	Oxidation	604107	95.85	4.15
4	Thermal	612402	97.16	2.84
5	UV	619477	98.28	1.72
6	Water	625366	99.22	0.78

Table 12 : Degradation Data of Samidorphan

S.NO	Degradation Condition	Area	% Drug Degraded	% Drug Undegraded
1	Acid	324822	94.04	5.96
2	Alkali	329145	95.29	4.71
3	Oxidation	330537	95.69	4.31
4	Thermal	337278	97.65	2.35
5	UV	339904	98.41	1.59
6	Water	342656	99.20	0.80

CONCLUSION

A new stability indicating RP-HPLC techinique was developed and validated for the simultaneous estimation of olanzapine and samidorphan in bulk and tablet dosage form. The developed method was set to be simple presize accurate with high resolution, shorter retention times with separated degradants and economical. Hence this method was used for the in-process evaluation in pharmaceutical manufacturing firms and routine quality control of drug testing laboratories.

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REFERENCES

1. B.k Sharma, Instrumental methods of chemical analysis, Introduction to analytical chemistry, 23rd Edition Goel publication , Meerut, (2007)
2. Lindholm.J, Development and Validation of HPLC Method for Analytical and Preparative purpose. Acta Universitatis Upsaliensis, pg . 13-14, (2004)
3. Rashmin, An introduction to analytical Method Development for Pharmaceutical formulations. Indoglobal Journal of Pharmaceutical Sciences , Vol.2 , Issue 2, Pg 191-196 (2012).
4. Malvia R, Bansal V , Pal O.P and Sharma P.K. A Review of High Performance Liquid Chromatography. Journal of Global Pharma technology (2010)
5. Douglas A Skoog, F. James Holler, Timothy A. Niemen, Principles of Instrumental Analysis Pg 725-760.
6. Dr.S. Ravi Shankar, Text book of Pharmaceutical analysis, Fourth edition, Pg 13.1-13.2
7. David G.Watson. Pharmaceutical Analysis, A text book for Pharmacy students and Pharmaceutical Chemists. Harcourt Publishers Limited; 2nd Ed., Pg 221-232.
8. Remingtonn's The Sciences and Practise of Pharmacy, 20th Edition (2000)
9. Connors Ka. A Textbook of Pharmaceutical Analysis, Wiley intersciences Inc; Delhi, 3rd Ed, Pg 373-421, (1994)
10. Gurdeep R.Chatwal , Sham K .Anand, Instrumental Methods of Chemical Analysis , Pg 2.566-2.638 (2007)
11. David G. Watson Pharmaceutical Analysis, A text book for pharmacy students and Pharmaceutical Chemists. Harcourt Publishers Limited; 2nd Ed.,Pg- 267-311
12. Nasal.A, Siluk.D, and Kaliszan.R. Chromatographic Retention Parameters in Medicinal Chemistry and Pharmacology, Pubmed, Vol.10, Issue 5 Pg no-381-426, March (2003)
13. Ashok Kumar, Lalith Kishore, navpreet Kaur , Anroop Nair. Method Development and Validation for Pharmaceutical Analysis. International Pharmaceutica Sciencia, Vol 2, Issue 3, Jul-Sep (2012)
14. Kaushal.C, Srivatsava.B, A Process of Method Development: A Chromatographic Approach. J Chem Pharm Res, Vol.2, Issue 2, 519-545, (2010)
15. Vibha Gupta, Ajay Deep Kumar Jain, N.S.Gill, Kapil, Development and Validation of HPLC method. International Research Journal of Pharmaeutical and Applied Sciences, Vol 2, Issue 4, Jul-Aug (2012)
16. Hokanson GC. A life cycle approach to the validation of analytical methods during Pharmaceutical Product Development. Part 1: The Initial Validation Process. Pharm Tech (1994) 92-100