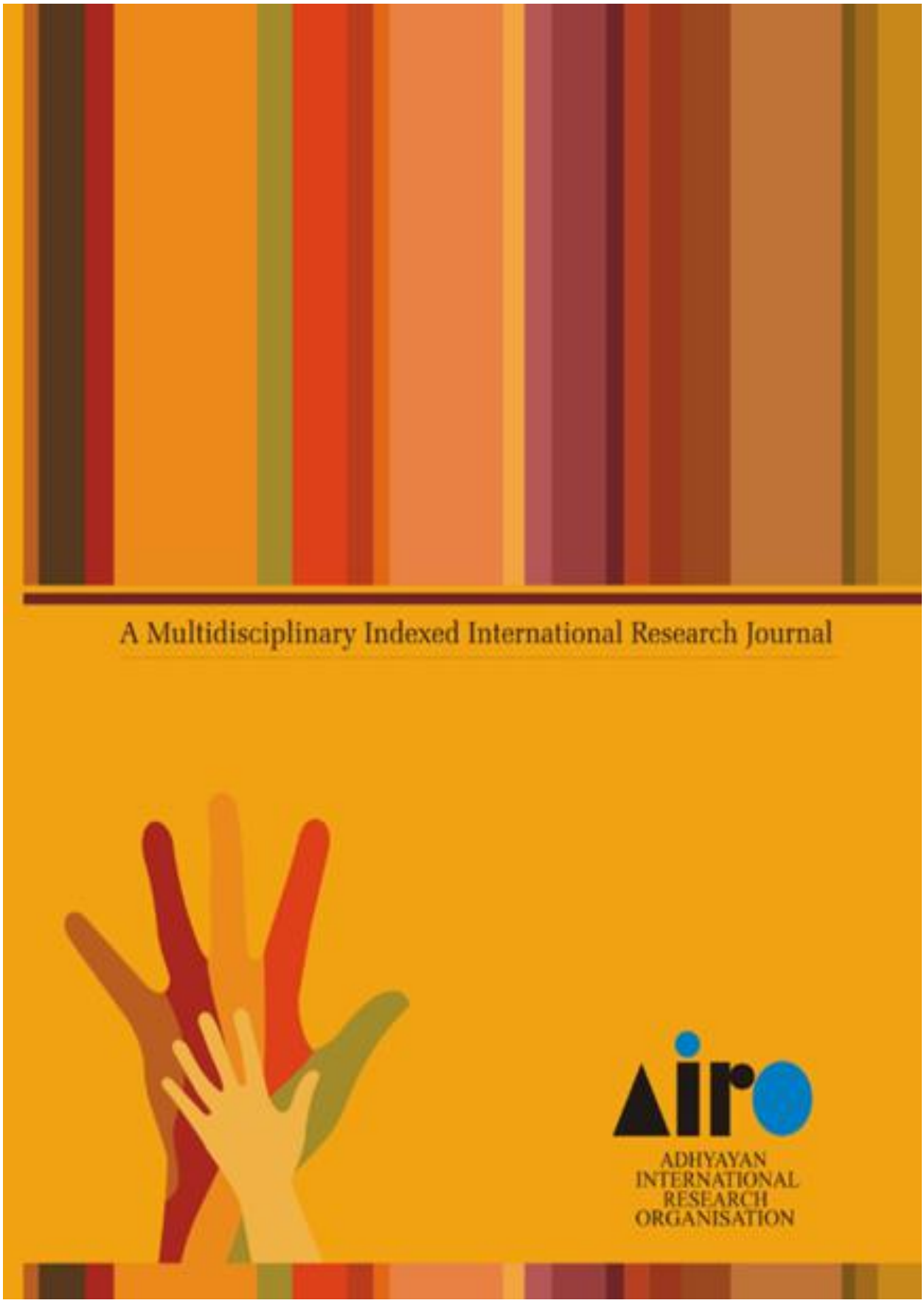
Airo International Research Journal
Volume XV, ISSN: 2320-3714
April, 2018
Impact Factor 0.75 to 3.19
UGC Approval Number 63012



Airo International Research Journal
Volume XV, ISSN: 2320-3714
April, 2018
Impact Factor 0.75 to 3.19
UGC Approval Number 63012

DEVELOPMENT AND VALIDATION OF A RP-HPLC METHOD FOR ESTIMATION OF SERTALINE AND ALPRAZOLAM IN A TABLET& BULK DOSAGE FORM

Debasis Bora

Research Scholar, Department of Chemistry, Sunrise University, Alwar, Rajasthan

Dr. Niraj Saxena

Supervisor, Department of Chemistry, Sunrise University, Alwar, Rajasthan

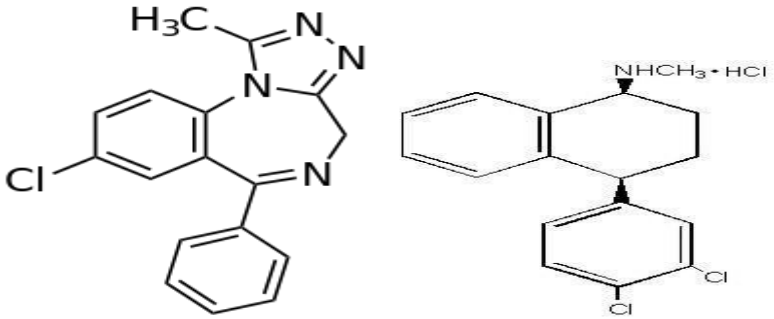
ABSTRACT

The purpose of the study was to develop a simple, sensitive and rapid RP-HPLC method for the determination of Sertaline and Alprazolam in marketed products. Chromatographic determination was performed in a reverse phase C18 column (250 mm × 3.3 mm I.D,5µm particle size) using a mixture of acetonitrile: methanol: 0.065 M ammonium acetate buffer (50:20:30, v/v/v), final pH adjust to 5.5 ± 0.02 with ortho phosphoric acid as mobile phase and delivered at a flow rate of 1 ml/min. The UV detection was set at 225 nm. The calibration range was from 2.0 to 30.0 µg/ml. The method was validated in term of linearity (r2>0.98, RSD= 1.958%), precision (RSD=3.757 %) and accuracy. The limit of quantification was 2 µg/ml and the limit of detection was 0.1 µg/ml. The potency of Sertaline and Alprazolam in marketed products was determined by this method with acceptable precision andreproducibility.

Keywords*: Sertaline and Alprazolam, marketed products, RP-HPLC, development*

INTRODUCTION

Sertaline (1S-cis)-4-(3,4-dichlorophenyl)-1,2,3,4-tetrahydro-N- Methyl-1-napthalenamainehydrochloride Antidepressant: Selectiveserotonin reuptakeinhibitor – Alprazolam Chloro-1-methyl-6-phenyl-4H-s-triazolo [4,3-α][1,4]benzodiazepine Anti-anxiety Solublein methanol or ethanol but which hasno considerable solubilityin water at physiological pH shown in Figures below.



OPTIMISED CHROMATOGRAPHIC CONDITIONS

The analytical method for the simultaneous stimation of Sertraline HCl and Alprazolam will be developed by RP-HPLC method by optimizing the chromatographic conditions. WATERS, software: Empower,2695 separation module,PDAdetector Phosphate buffer(0.05M) pH3.6: ACN (40:60%v/v) Column SymmetryC18 5µm(4.6x250mm) Make; waters with 225 nm.

MobilePhase Optimization

Initially the mobile phase tried was methanol: Ammonium acetate buffer and acetonitrile: phosphate buffer with various combinations of pH as well as varying proportions. Finally, the mobile phase was optimized to potassium dihydrogen phosphate with buffer (pH 3.5), acetonitrile in proportion 40: 60 v/v respectively.

Wavelength selection

UVspectrumof10µg/ml Sertaline HCl and Alprazolam indiluents (mobile phase composition) was recorded by scanning in the range of 200nm to 400nm. From the UV spectrum wavelength selected as 225. At this wave length both the drugs show good absorbance.

OptimizationofColumn

Themethod wasperformed with various columns like C18 column, hypersil column, lichrosorb, and inertsil ODS column. Symmetry C8(4.6x150mm,5µm, Make: XT erra) was found to be ideal a sit gave good peak shapeandresolutionat0.6ml/min flow.

Preparation of buffer and mobile phase

Preparation of Phosphate buffer

Accurately weighed 6.8grams of $KH_2PO_4$was taken in a 1000ml volumetric flask, dissolved and diluted to1000 ml with HPLC water and the volume was adjusted to pH 2.8 with Orthophosphoric

Preparationofmobilephase

Accurately measured 450ml (45%) of above buffer and 550ml of Methanol HPLC(55%) were mixed and degassed in an ultrasonic water bath for10 minutes and then filtered through 0.45µ filter under vacuum filtration.

Diluent Preparation: The Mobile phase was used as the diluents

Methodvalidationsummary

Precision

Preparationofstocksolution

Accurately weigh and transfer 25mg of Sertraline HCl and Alprazolam working standard intoa10mLsanitarywaterlessvolumetricflask, diluents added upto 7ml and sonicate to liquefy it wholly and makeup solvent up to mark.

Intermediateprecision/ruggedness

Accurately weigh and transfer 25 mg of Sertraline HCl and10mg of Alprazolam working standard into a 10mL fresh water less volumetric flask add about 7m Lof mobile phase and sonicate to dissolve it totally and solvent added upto spot with similar solvent.

1ml to 5 ml and0.1mlofstocksolutionshastakenin different10mlofgraduatedvolumetric flasks, dilute up to themark with diluent.

Limit of detection

Limit ofDetection: (ForSertralineHCl) Preparationof300µg/mlsolution and 0.12µg/mlsolution

Accurately weigh and transfer10mg of Sertraline HCl standard transferred into a 10mLfresharidvolumetric flaskaddabout 7mLofmobile phaseand sonicate todissolve it completelyandcomposevolumeup tothe spot with the alike solvent. (Stock solution). Furtherpipette3mlof theabovepreviouslypreparedstocksolutionintoa graduated 10ml volumetric flask and attenuate upto themark with mobilephase.

Limit of quantification

Limit ofQuantification (for SertalineHCl)
Preparationof300µg/mlsolution

Accurately weigh and transfer10mg of Sertraline HCl transferred into a10mL hygienic dried up volumetric flask add about Diluent addedupto7mlsonicateto dissolve it entirelyandcomposevolumeup to thespot with thesimilar solvent.

Fig No: 1 Chromatogram showing for Sertaline HCl and Alprazolam sample Preparation

S.No	Injection	PeakName		Rt		Area	Area
1	Injection-1	SertalineHCl	Alprazolam	2.344	3.281	13005070	122487
2	Injection-2	SertalineHCl	Alprazolam	2.343	3.281	1304520	122626
3	Injection-3	SertalineHCl	Alprazolam	2.345	3.283	1305937	122632
4	Injection-4	SertalineHCl	Alprazolam	2.344	3.278	1306476	122702
5	Injection-5	SertalineHCl	Alprazolam	2.342	3.278	130871	122962
					Mean	1305070.2	122681.8
					Standard Deviation	3061.8	174.8
					%RSD	0.2	0.1

Table no 1: Results ofIntermediateprecision forAlprazolam and Sertaline

Parameters	**SertalineHCl Hcl**	**Alprazolam**
Slope (m)	66574	12529
Intercept (c)	53592	50245
Correlation coefficient (R^2)	0.999	0.999

Table no :2 Analytical performanceparameters ofSertalineHClandAlprazolam

Drug name	**Baselinenoise(µV)**	**Signal obtained**	**S/N ratio**
SertalineHCl	52	522	10.03

Alprazolam	52	524	10.1

Table No: 3 Results of LOQ

	Flow Rate (ml/min)	**SystemSuitabilityResults**	
		USPPlateCount	**USPTailing**
1	0.8	7063.3	1.3
2	1.0	6090.3	1.2
3	1.2	6998.0	1.3

Table No:4 Flowrate(ml/min) data.

CONCLUSION

High performance liquid chromatography is at present one of the most sophisticatedtoooftheanalysis.TheestimationofSertalineHClandAlprazolamwas donebyRP-HPLC.ThePhosphatebufferwaspH2.8andthemobilephasewasoptimized with consists of Methanol: Phosphate buffer mixed in the ratio of 55:45%v/v. AC 18 columnC18(4.6x150mm,5µm, Make: XTerra)or equivalent chemically bonded to porous silica particles was used as stationary phase. The detection was carried out using UV detectorat 225nm. The solutions were chromatographed at a constant flow rate of 1.0ml/min the linearity range of Sertaline HCl and Alprazolam were found to be from 100-500 µg/mlof Sertaline HCl and 1-5µg/ml of Alprazolam. Linear regressioncoefficient was not morethan 0.999.

ACKNOWLEDGEMENT

I Sincerely thank almighty god for his grace and extend support from my guide and each and every one from the research department of Sunrise University, Alwar and the gifted sample provided by Spectrum labs Hyderabad.

REFERENCES

[1] Beckett A. H 0and Stenlake J.B; textbook of pharmaceutical chemistry 4thEdition,-part CBS publishers and Distributors, New Delhi;**1998:**P-278,307

[2] Douglas S k o o g A , James H o l l a r F. and Timothy Nieman, APrinciples of

[3] Instrumental Analysis.5thedition., Thomson Learning Inc. Singapore; **1998**; P-110,300

[4] Sethi,P.D,QuantitativeAnalysis of Drugs in Pharmaceutical Formulation, 3rded., CBS Publishers and Distributors;**1997,** P-1-29 ,50-64

[5] Mendham,R.C.,Denny,J.D.,Barnis,M.andThomas,J.K.,Vogel's TextBookof Quantitative Chemical Analysis.6thed., PearsonEducation**,2003**; P-1, 676

[6] Sharma, B.K., InstrumentalmethodofChemicalAnalysis. 24th edition, GOELPublishingHouse;Meerut,**2005**; P-46, 68.