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Research Article

REVERSE PHASE HIGH PERFORMANCE LIQUID CHROMATOGRAPHIC METHOD FOR THE ANALYSIS OF BUPROPION HYDROCHLORIDE IN PHARMACEUTICAL DOSAGE FORM

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ABSTRACT

A rapid and sensitive reverse phase HPLC method is depicted for the qualitative and quantitative assay of Bupropion hydrochloride in pharmaceutical dosage form. Bupropion hydrochloride was chromatographed on a reverse phase C_{18} column with a mobile phase consisting of methanol: phosphate buffer (pH-6) in the ratio of 80:20% v/v.The mobile phase was pumped at a flow rate of 1ml/min. Aceclofenac was used as an internal standard and the eluents were monitored at 223 nm. The retention time of the drug was 5.7 min.With this method, linearity is observed between area under curve (AUC, expressed in mV.min) and concentration of Bupropion hydrochloride in the injected solution, in the range of 10-200 µg/ml.The method was found to be applicable for analysis of drug in tablets. The results of the analysis were validated statistically.

Key words: Bupropion hydrochloride, Reverse phase HPLC, Tablets. Validation

INTRODUCTION

Bupropion[1, 2] chemically (±)-2-(*tert*-butylamino)-1-(3-chlorophenyl) propan-1-one,previously known as amfebutanone is an atypical antidepressant and smoking cessation aid it acts as a nor epinephrine and dopamine reuptake inhibitor as well as α_3 β_4 nicotinic receptor antagonist.Bupropion hydrochloride belongs to the chemical class aminoketone and is similar in structure to stimulate cathinone and diethylepropion and phenethyleamines in general, Effectiveness of bupropion hydrochloride in long-term use (more than 6 weeks) has not been systematically evaluated in controlled trials. Therefore, the physician who elects to use Bupropion hydrochloride sustained release tablets for extended periods should periodically reevaluate the long-term usefulness of the drug for the individual patient. The drug is available in tablet form (100mg, 150mg and 450mg) and is official in any pharmacopoeia. A few methods of analysis of Bupropion hydrochloride have been reported using different techniques such as validated liquid chromatographic method for the quantization of Bupropion hydrochloride in human plasma using liquid-liquid extraction[3], solid-phase extraction-liquid chromatography-mass spectrometry[4,5],spectrophotometric estimation of Bupropion hydrochloride in bulk drug and dosages forms. Most of these methods are considered tedious. The HPLC methods using the most commonly available columns and detector like UV are preferred. The present study describes the determination of Bupropion hydrochloride in pharmaceutical dosage forms by using RP-C_{18} column with UV detectors. Owing to the widespread use of HPLC in routine analysis, it is important that well validated[6, 7, 8] HPLC methods for the estimation of Bupropion hydrochloride in different pharmaceutical dosage forms.

Material and method

The pure Bupropion hydrochloride we used for the development of analytical method was gifted by Lupins pharmaceutical and Aceclofenac (internal standard) was supplied from sun pharma. Methanol and water were of HPLC grade (Merck). All other regents were of AR grade. An isocratic HPLC (Waters India, USA) with a single Waters 510 pump, Waters 486 tunable absorbance detector and RP-C_{18} column (Bondapak C_{18} 250×4.6mm, packed with 5µm particle size) was used. The HPLC system was equipped with ***Millennium^{32}software.*** Commercial Bupropion hydrochloride tablets were purchased from local market. Nicotex and Bupron a product by Eli Lilly & Co. (New Delhi, India) contained 150mg tablets.

Chromatographic conditions

The composition of the mobile phase is methanol and phosphate buffer at ph 6 in the ratio of 80:20 % v/v. The mobile phase was filtered before use through a 0.45µm membrane filter and degassed for 30 min.

The components of the mobile phase were pumped from the solvent reservoir to the column at a flow rate of 1ml/min that produced column back pressure 140-150 kg/cm^2. Ambient column temp was maintained. The eluents were monitored at 223nm.

Drug and internal standard solution:

A pure sample of Aceclofenac procured from sun pharmaceutical was used as reference standard in the study. About 50mg of Bupropion hydrochloride was weighed accurately and transferred into a 50ml volumetric flask and dissolved in 25ml of the mobile phase. Then the volume was made up with a further quantity of mobile phase to get 1mg/ml solution. Following this the solution was sonicated for 30min to degas it. Subsiquent dilution of this solution ranging from 10-200µg/ml were made in 10ml

volumetric flask after addition of 1ml Aceclofenac solution (100µg/ml) as an internal standard to each dilution.20µl of the solution was injected each time into the stream of mobile system at a flow rate of 1ml/min.Each of the dilution was injected six times into the column and the corresponding chromatograms were obtained. From these chromatograms, the area under the peaks of the drug and internal standard were noted. Using these values, the mean ratio of peak area of the drug to that of the internal standard for each dilution was calculated. The regression of the drug concentration was computed. This regression equation was used to estimate the amount of Bupropion hydrochloride in the pharmaceutical dosage form. Solutions containing 50-70ug/ml of Bupropion hydrochloride were subjected to the proposed HPLC analysis to check the intra-day and inter-day variation of the method. The recovery studies were carried out by adding known amounts of Bupropion hydrochloride to the preanalyzed sample and then analyzing them by the proposed HPLC method.

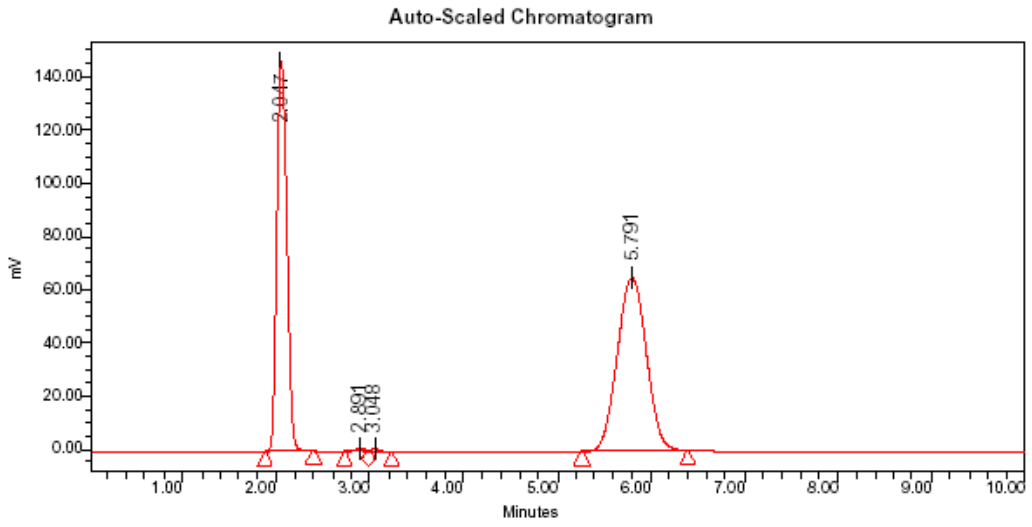
Estimation of Bupropion hydrochloride in the Tablet dosage form:

Two commercial brands of tablets (Nicotex and Bupron a product by Eli Lilly & Co) were chosen for testing suitability of the proposed method to estimate Bupropion hydrochloride in tablet formulation. Twenty tablets were weighed and powdered. An accurately weighed portion of this powder equivalent to 50mg of Bupropion hydrochloride was transferred to a 50ml volumetric flask containing 25ml of mobile phase. The content of the flask were allowed to stand for 6 hours with intermittent sonication to ensure complete suitability of the drug and then filtered through 0.45µ membrane filter. From the filtrate different aliquots were taken in separate 10ml volumetric flask. These solution were spiked with suitable volume of internal standard solution, such that the concentration of each solution was 100ug/ml. The contents of the flask was made up to the volume with the mobile phase and mixed well. Each of this solution (20µL) was then injected 6 times into the column. The mean peak area ratio of

the drug to the internal standard of 6 such determinations was calculated and the drug content in tablets was quantified using the regression equation obtained for the pure sample.

RESULT AND DISCUSSION

To achieve precise component peaks with good resolution under isocratic conditions mixtures of methanol and phosphate buffer in different combination were tested as mobile phase on a C_{18} stationary phase. A binary mixture of methanol and phosphate buffer (pH-6) in 80:20%v/v proportion was proved to be the most suitable of all combination since the chromatographic peaks were better defined and resolved and almost tailing with this system. Though the structure of Aceclofenac is not too similar to Bupropion hydrochloride, it was chosen as an internal standard, because it showed better peak shape and peak location compared to other potential internal standard. Under the above mentioned chromatographic condition the retention time obtained for Bupropion hydrochloride and internal standard were 5.7 and 2.0 min respectively. A model chromatogram was shown in the figure below.



Each of the samples was injected six times and same retention times were observed in all cases. The ratio of peak area of Bupropion hydrochloride to peak area of internal standard for different concentration set up as above were calculated and the average values for six such determination are

shown in Table-1.The peak area of both the drug and internal standard were reproducible as indicated by low coefficient of variance (1.01%).A good linear relationship (r=0.9996) was observed between the concentration of Bupropion hydrochloride and the respective ratio of peak areas.

The drug content in the tablets was quantified using the proposed analytical method. The mean amount of Bupropion hydrochloride in two different brands of tablet dosage form is shown in Table-4.The absence of additional peaks in the chromatogram indicates non-interference of the common exipients used in the tablets. It can be concluded that the proposed HPLC method is sufficiently sensitive and reproducible for the analysis of Bupropion hydrochloride in pharmaceutical dosage forms within a short analysis time. The method was duly validated by the required Para Development of new analytical methods for the determination of drugs in pharmaceutical dosage forms is more important in pharmacokinetic, toxicological and biological studies.

Table-1 Calibration of the proposed method

SL. NO	CONC (µg/ml)	Mean Peak Area Ratio (n = 6)	% R. S. D
1.	10	0.279101302	0.13
2.	20	0.536527078	0.03
3.	30	0.733388637	0.12
4.	40	1.00317242	0.32
5.	50	1.286038653	0.07
6.	60	1.4796	0.15
7.	70	1.716630968	0.73
8.	80	1.970037647	0.51
9.	90	2.26921383	0.09
10.	100	2.419993374	0.41
11.	150	3.705465536	0.31
12.	200	4.898005953	0.13

Table-2 Precision of the proposed method

CONC OF BUPROPION HYDROCHLORIDE (µg/ml)	OBSERVED CONC OF BUPROPION HYDROCHLORIDE (µg/ml)			
	INTRA DAY (MEAN) n=6	% R. S. D	INTER DAY (MEAN) n=6	% R. S. D
50	50.25	0.13	50.13	0.17
60	59.48	0.17	59.73	0.29
70	69.23	0.62	69.58	0.78

Table-3 Recovery Data of Bupropion hydrochloride

Amount of drug added(µg)to solution of pure drug /tablet formation	Recovery from drug solution		Recovery from tablet formulation	
	Mean(±S.D) Amount(µg) Found(n=6)	Mean(±S.D) %recovery(n=6)	Mean(±S.D) Amount(µg) Found (n=6)	Mean(±S.D) %recovery(n=6)
16	15.27±0.21	96.01±0.19	15.56±0.07	97.31±0.27
20	19.55±0.81	97.81±0.78	19.19±0.58	96.02±0.87
24	23.92±0.06	99.68±0.09	23.86±0.11	99.31±0.18

Table-4 Assay of Bupropion hydrochloride dosage form

Brand name of the tablet	Labeled amount of drug (mg)	Mean (±S.D) amount (mg) found by the proposed method	% Mean (± S.D) of labeled amount (n=6)
Nicotex	150	150.79±0.38	100.19±0.14
Bupron	150	149.98±0.47	99.0±0.21

CONCLUSION

Development of new analytical methods for the determination of drugs in pharmaceutical dosage forms is more important in pharmacokinetic, toxicological and biological studies.

The aim of the present study was to develop simple, fast and sensitive HPLC method for the determinations of Bupropion hydrochloride

The method described herein is simple validated assay procedures that can readily be used in any laboratory for the quantitative determination of Bupropion hydrochloride. Analytical figures of merit demonstrated during the method validation protocol compare well with those of known methods for the determination of Bupropion hydrochloride. The assay procedure was simple with satisfactory precision (less than 3.5) and accuracy in terms of relative error (less than ± 2.0). We believe that this method fulfill experimental and clinical requirements for determining the drugs and can be applied for any study. Concentration of buffers used in this method was very low and pH used also reliable and doesn't produce any troubleshoot to column and instrument. Practically, low cost of single analysis are central features of routine laboratory tests, and the herein described RP-HPLC method, because it uses low cost solvents and is easily affordable by clinical laboratories equipped with standard high-performance liquid chromatography systems. So the developed method was very useful in the determination of Bupropion hydrochloride.

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REFERENCES

1. Budavari,S O.Neil M J Smith A Heckleman PE eds ,The Merk index, an Enclyopedia of chemicals, drugs and biologicals 11th edition,new jersey merk and co Inc 1989, Page –1523.
2. United State Pharmacopoeia 29 (NF24), Rockville MD: United State Pharmacopeal Convention Inc,2006.
3. Journal of liquid chromatography and related Technologies,Taylor and Francis,Volumn29,Number 1/2006, Pg:123-135.
4. Elen Katz, Quantitative analysis using chromatographic techniques, John Wiley & sons, (1987) 55-59.
5. Sane RT ,Francies M, khedkar S.Menezrs.A, Moghe A and Patil P"Gas chromatographic determination of bupropion hydrochloride from its phramceutical formulation"Indian drugs,40(4) April 2003 Page 231.
6. Guideline for submitting samples and analytical data for methods validation, (1987).
7. Text on validation of Analytical Procedures, international conference on Harmonization.
8. C.Arelano,C.Phillibert,C,vacoux,J.woodley,G.houin "Validation of liquid chromatography and mass spectroscopy method to asses the metabolism of Bupropion hydrochloride in rat everted gut sac ,"J chromatography B-829; 50-55 (2005).

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