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Development and Validation of U V Spectrophotometric Method for Determination of Trifluoperazine Hydrochloride in bulk and Pharmaceutical Dosage Form

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Abstract- A new simple, rapid, accurate, sensitive and precise spectrophotometric method for the determination of trifluoperazine hydrochloride in bulk and capsule dosage form .pharmaceutical formulations is described. The method is based on the simple solubility of trifluoperazine hydrochloride in methanol. The absorbance maximum of trifluoperazine hydrochloride measured at wave length 265 nm. The drug obeys Beer's Law in the concentration range 2-45 µg/ml employed for this method. Accuracy and reproducibility of the proposed method was statistically validated by recovery studies. The method is easily be employed in the laboratory for the routine estimation of drug and it's extended to the analysis of trifluoperazine hydrochloride in pharmaceutical formulations.

Index Terms- UV spectrophotometry, trifluoperazine hydrochloride, methanol, Beer's law, capsule analysis . Least square regression analysis

I. INTRODUCTION

The primary application of trifluoperazine is for schizophrenia .Other official indications may vary country by country, but generally it is also indicated for use in agitation and patients with behavioural problems, severe nausea and vomiting as well as severe anxiety. Its use in many parts of the world has declined because of highly frequent and severe early and late tardive dyskinesia, a type of extrapyramidal symptom. The annual development rate of tardive dyskinesia may be as high as 4%. A 2006 study suggested that trifluoperazine may be able to reverse addiction to opioids.[1]A multi-year UK study by the Alzheimer's Research Trust suggested that this and other antipsychotic drugs commonly given to Alzheimer's patients with mild behavioural problems often make their condition worse.[2] Pharmacology OF Trifluoperazine has central antiadrenergic,[3] antidopaminergic,[4][5] and minimal anticholinergic effects.[6] It is believed to work by blockading dopamine D1 and D2 receptors in the mesocortical and mesolimbic pathways, relieving or minimizing such symptoms of schizophrenia as hallucinations, delusions, and disorganized thought and speech.[7]Side effectsA 2004 meta-analysis of the studies on trifluoperazine found that it is more likely than placebo to cause extrapyramidal side effects such as akathisia, dystonia, and Parkinsonism.[7] It is also more likely to cause somnolence and anticholinergic side effects such as blurred vision and xerostomia (dry mouth).[7] All phenothiazines can cause the rare and sometimes fatal neuroleptic malignant syndrome.[8] Trifluoperazine can lower the seizure threshold.[9] The antimuscarinic action of trifluoperazine can cause excessive dilation of the pupils (mydriasis), which increases the chances of patients with hyperopia developing glaucoma.[10] Chemically Trifluoperazine hydrochloride is (2-trifluoromethyl-10-[3-(4-methyl-1-piperazinyl)propyl]phenothazine). It has a highly selective mode of action, and produces a tranquil state of mind without excessive sedation [11,12]it is synthesized in the manner described already for prochlorperazine, alkylation is performed using 2-trifluoromethylphenothazin-4-methyl-1-piperazinylpropylchloride as the substrate.Many patients that have not responded to other therapies have benefited from this versatile psychotropic.agent. Recently, TFPH has been the focus of extensive research, both for the development of rapid and simple methods for its detection and because of its structural properties as a reagent for chemical analysis [13]. Various methods have been employed for the determination of TFPH including titrimetry [14], potentiometry [15],spectrophotometry [16–19] fluorimetry [20], flow injection analysis [21,22], sequential injection analysis [23],high-performance liquid chromatography [24,25] and liquid chromatography electrospray ionization mass spectrometry (LC–ESI–MS) [26]. chromatography, and LC–ESI–MS, all require highly sophisticated instruments that are relatively expensive and not available in all laboratories. By contrast, ultraviolet–visible (UV–Vis) spectroscopy is simple with sufficient precision and sensitivity, but it does suffer from strong interferences from drug excipients and diluents [27]. Therefore, an attempt was made to develop a simple spectrophotometric method for the estimation of the present drug in bulk and pharmaceutical formulations

II. EXPERIMENTAL

2.1. *Materials*

Pharmaceutical gradeTFPH was obtained from south Africa and india .methanol was used in all stages. Two brands of capsules STELAZINE SPANSULE CAPSULE (smithkline beecham pharmaceuticals pvt ltd, johannesberg, SA) and

ARBPAN-2 (arbro pharmaceuticals, New delhi, India) were procured from the local market.

2.2. *Apparatus*

Spectrophotometric analysis was performed on a Shimadzu UV-1800 spectrophotometer, with a 1.00 cm quartz cells. The instrument settings were optimized to produce a spectrum with about 80% full-scale deflection and acceptable noise level. Each spectrum was recorded in triplicate. For each replicate measurement the cell was refilled with fresh solution

Methods

2.3.1. *Preparation of TFPH standard solutions*

A stock solution containing 100 µg mL-1 of TFPH was prepared by dissolving 10mg of TFPH in methanol, then transferring into a 100 mL volumetric flask and diluted up to the mark with methanol. All measurements were made at room temperature.The standard solutions were prepared by the proper dilutions of the stock standard solution with methanol to reach concentration range of 2-45 µg mL-1. The determination was conducted in triplicate.

2.3.2. *Preparation of sample solution*

A powder (55.10 mg) equivalent to 10 mg of TFPH was transferred to 100 mL volumetric flask. The content was mixed with 50 mL of methanol. The mixture was sonicated for 20 min. This solution was filtered through the whatman filter paper No.41 and the filtrate and washings were combined and diluted to the 100 mL with methanol to get solution having TFPH 100 µg mL-1.

2.3.3 *Preparation of Calibration Curve*

The standard stock solution of TFPH was scanned in the wavelength range of 200 nm to 400 nm against methanol as a blank. A calibration curve was constructed over a concentration range 2-45 µg mL-1. Absorbance of each solution was measured at the wavelength of 265 nm. Calibration curve was constructed for TFPH by plotting absorbance versus concentration at 265 nm wavelength. The determination was conducted in triplicate. (Fig **: 2)**

2.4. *Validation of Method*

The method was validated with respect to linearity, accuracy, precision, limit of detection (LOD) and limit of quantitation (LOQ) [31,32,33]

2.4.1 *Linearity*

To establish linearity of the proposed method, ten separate series of solutions of TFPH (2–45 µg mL-1 in methanol) were prepared from the stock solutions and analyzed. Least square regression analysis was performed on the obtained data

2.4.2 *Accuracy*

The accuracy of the method is the closeness of the measured value to the true value for the sample. To determine the accuracy of the proposed method, different levels of drug concentrations lower concentration (LC, 80%), intermediate concentration (IC, 100%) and higher concentration (HC, 120%) were prepared from independent stock solutions and analyzed (n = 10). Accuracy was assessed as the percentage relative error and mean % recovery (Table 1). To provide an additional support to the accuracy of the developed assay method, a standard addition method was employed, which involved the addition of different concentrations of pure drug (10, 20 and 30 µg mL-1) to a known preanalyzed formulation sample and the total concentration was determined using the proposed method (n = 10). The% recovery of the added pure drug was calculated as % recovery = [(Ct–Cs)/Ca] x 100, where Ct is the total drug concentration measured after standard addition; Cs, drug concentration in the formulation sample; Ca, drug concentration added to formulation (Table 1).

2.4.3 *Precision*

Repeatability was determined by using different levels of drug concentrations (same concentration levels taken in accuracy study), prepared from independent stock solutions and analyzed (n=10) (Table 2). Inter-day and intra-day variations were studied to determine intermediate precision of the proposed analytical method. Different levels of drug concentrations in triplicates were prepared three different times in a day and studied for intraday variation. The same procedure was followed for three different days to study inter-day variation (n = 10). The percent relative standard deviation (% R.S.D.) of the predicted concentrations from the regression equation was taken as precision (Table 2). Precision studies were also carried out using the real samples of TFPH capsule in a similar way to standard solution to prove the usefulness of the method

2.4.4 Limit of Detection (LOD) and Limit of Quantitation (LOQ)

LOD (k=3.3) and LOQ (k=10) of the method was established according to ICH definitions (C1=k*So/S, where C1 is LOD or LOQ, So is the mean standard deviation of blank determination, S is the slope of standard curve and k is the constant related to confidence interval). LOD and LOQ of method are reported in Table 2.

III. Results and Discussion

The development of spectrophotometry methods for the determination of drugs has increased considerable in recent years because of their importance in pharmaceutical analysis.Based on the experimental data the standard calibration curve was plotted (Fig.2) The absorbance range was found to be 0.107-1.879 The content of drug was calculated from the equation y = 0.041x + 0.028. These solutions obeyed Beer-Lambert's law in concentration range of 2-45 µg mL-1 with R2 value of 0.999. The assays were validated by means of ANOVA (Analysis of variance), as described in official literature [34]. This developed method presented no parallelism deviation and no linearity deviation (P < 0.05).The reproducibility of the proposed method was determined by performing capsule assay at different time intervals on same day (Intra-day assay precision) and on three different days(Inter-day precision). Result of intra-day and inter-day precision is expressed in % RSD.Percent RSD for Intraday assay precision was found to be 0.0504. Inter-day assay precision was found to be 0.0905. According to the equation, the LOD and LOQ were found to be 0.27and 0.82 µg/mL, respectively. This data shows that this method is sensitive for the determination of TFPH. To ascertain the accuracy of proposed methods, recovery studies were carried out by standard addition method at three different levels (80%, 100% and120%). Percent recovery for TFPH by the proposed method was found in the range of 99.33 % to 100.14 %. Repeatability is based on the results of the method operating over short time interval under same conditions. The low RSD values of intra-day precision (Table 2),recovery (Table 1), and pharmaceutical preparations (Table 3) showed high repeatability.

IV. Conclusion

UV spectrophotometric method developed for trifluoperazine hydrochloride is simple ,accurate, sensitive, rapid and economic and it can be conveniently employed for the routine analysis and the quality control of trifluoperazine hydrochloride in pharmaceutical dosage forms.The method was suitable to determine concentrations in the range 2-45 µg mL-1.The limits of detection and quantitation for trifluoperazine hydrochloride with a lower concentration were 0.27 and 0.82 µg mL-1 respectively, which are under the lowest expected concentrations in the sample. The sample recovery from the formulation was in good agreement with its respective label claim.

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Fig : 1 The chemical l Structure of trifluperazine hydrochloride (TEPH)

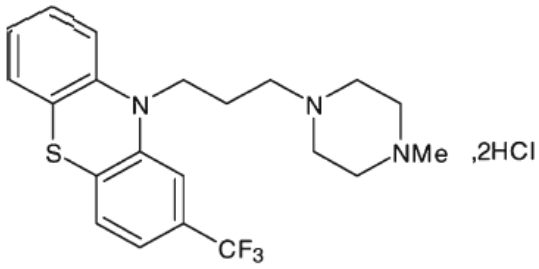

Fig : 2 Calibration Curve of trifluperazine hydrochloride in methanol

Y=0.041X+.028
R^2 =0.999

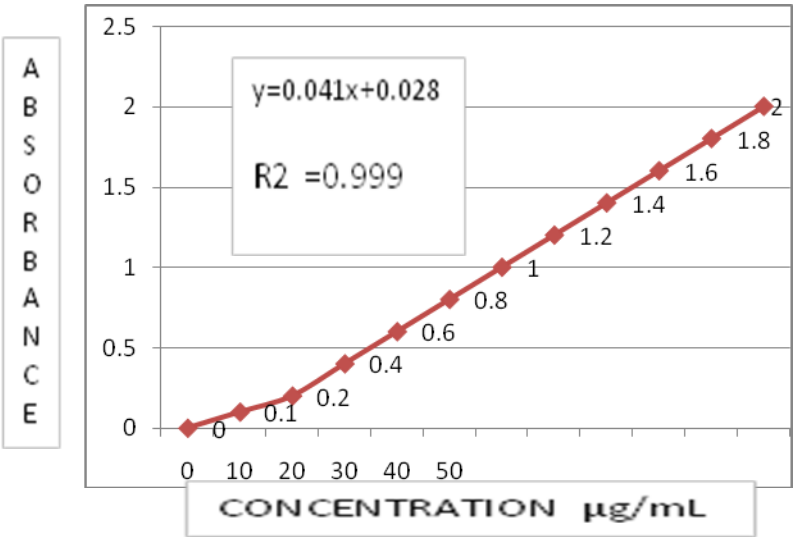

Table 1: Recovery Studies of TFPH (n=3)

Label claim (mg)	Amount added (%)	Recovery (%) ± S.D	% R.S.D
2	80	99.33± 0.0033	0.0032
	100	99.96 ± 0.0081	0.0080
	120	100.14 ± 0.0068	0.0068

Table 2: Optical Characteristics of TFPH

Parameter	values
λmax, nm	265
Beer's law limit, µg mL^{-1}	2-45
Molar absorptivity, L/mol×cm	2.049×10^4
Regression equation	y = 0.041x + 0.028
Slope ± S.D	0.041 ± 0.00014
Intercept ± S.D	0.028 ± 0.0027
Correlation coefficient (r2)	0.999
Limit of Detection (LOD), µg mL^{-1}	0.27
Limit of Quantitation (LOQ), µg mL^{-1}	0.82
Intra day precision (% R.S.D)	0.0504
Inter day precision (% R.S.D)	0.0905

Table 3: Analysis of TFPH capsule

Brand Name	Label Claim(mg)	% Assay± S,D	% R,S,D
Stelazine	2	99.76 ± O.0087	0.0087
Arbpan	2	99.54 ± 0.0079	0.0080