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Keywords: Haloperidol; HPLC; SLNs; Validation

Introduction

Haloperidol is an orally administered dopamine inverse agonist of the typical antipsychotic class of medications that chemically belongs to butyrophenone group [1]. Its mechanism of action is mediated by blockade of D2 dopamine receptors in brain [2]. It is used to treat certain psychiatric conditions including schizophrenia, manic states, medicament induced psychosis and neurological disorders with hyperkinesias [3]. Haloperidol is chemically 4-[4-(4-chlorophenyl)-4-hydroxypiperidino] 4'-uorobutyrophenone (Figure 1). The molecular formula of haloperidol is $C_{21}H_{23}ClFNO_2$ and molecular weight is 375.86 g/mol.

Author developed the solid lipid nano particles of haloperidol for nose to brain delivery (data are not given here and already published by same author). SLNs have the several advantages including targeted drug delivery and controlled release delivery, and increase bioavailability so as to reduce the dose and adverse effects associated with drug. SLNs offer an improvement to traditional nose-to-brain drug delivery since they are able to protect the encapsulated drug from biological and/or chemical degradation and may also increase nasal retention time due to an occlusive effect, good application properties, and adhesion of the SLNs to mucous membranes [4].

Various analytical techniques have been used for determination of haloperidol in pharmaceutical formulations. These include high performance thin-layer chromatography (HPTLC) [5], ¹⁹F NMR spectroscopy [6], square-wave adsorptive stripping voltammetry at a mercury electrode [7], square-wave and cyclic voltammetry at hanging mercury drop electrode [8], cyclic voltammetry at multi-walled carbon nanotubes-modified glassy carbon electrode [9]. Non-aqueous titrimetric method has also been developed for haloperidol determination [10]. Several UV spectrophotometric assay procedures have been developed and described in official compendia [11-13]. The literature survey revealed that haloperidol was quantitatively assayed in pharmaceutical preparations as well as in biological fluids either individually or in the presence of other drugs using HPLC (Table

1) but procedures related to detection of haloperidol in solid lipid nanoparticles are not published in literature till today. Author tried to reproduce the previously reported methods (Table 1) but due to unknown reasons none of them could be reproduced. Hence, the aim of the research work was to develop and validate a new HPLC method for haloperidol. The different analytical performance parameters such as linearity, precision, accuracy, reproducibility, limit of detection (LOD) and limit of quantification (LOQ) were determined as per ICH Q2 (R1) guidelines [14,15]. The significance and relevance of this method was in vitro dissolution assessment of solid lipid nanoparticles. It showed sustained drug release with maximum value of $95.52 \pm 5.21\%$ in found to be 2.31 years at 4°C.

New Delhi, India. All reagents were of HPLC grade. Milli-Q grade (Millipore, Molsheim, France) water was used to prepare solutions wherever required and it was filtered before use through a 0.22 µm membrane filter.

Instruments

Bath sonicator (Multitech Pvt. Ltd., N. Delhi), Balance (AUX 220, Shimadzu Corporation, Kyoto, Japan), Magnetic stirrer (Remi Instruments Pvt. Ltd., Mumbai, India), pH meter (Hicon Scientific Instruments, Delhi, India), Centrifuge (Remi Instruments Pvt. Ltd., India), Micro pipettes (Labnet, USA), Vortex mixture (S.M. Scientific instruments, Pvt. Ltd, Delhi) were used for the study.

Method validation

HPLC method for Haloperidol was validated as per the ICH guidelines Q2 (R1) for linearity, precision, accuracy, repeatability, LOD and LOQ.

Linearity: r = 0.999

HPLC System

The HPLC system consisted of a pump (Jasco PU 2080 plus), UV/VIS detector (Jasco UV 2075 plus) and the software used was Jasco Borwin version (1.5, LC-Net II/ADC system). The column used was C-18 Cosmosil packed column (5 µm, MS-II, 250 mm × 4.6 mm with particle size of 5.0 µm). Syringe filters (Syringe-driven filters of 0.22 µm, HiMedia Laboratories Pvt. Ltd., Mumbai, India) were used to filter sample.

Preformulation studies of haloperidol: Haloperidol was received as a gift sample from Vamsi Labs Ltd Solapur, Maharashtra, India. Authenticity of haloperidol was verified by conducting the preformulation studies like organoleptic properties, melting point, FTIR spectral analysis, DSC study, partition coefficient determination.

HPLC method

Several methods have been developed for the determination of haloperidol in pharmaceutical preparation.

The concentration of haloperidol had been earlier determined by HPLC method using methanol-water (63:37, v/v) containing 0.2 M ammonium acetate as mobile phase and diphenylamine as internal standard [21]. The method used here was slightly modified in the terms of mobile phase. The mobile phase consisted of 100 mM/L potassium dihydrogen phosphate-acetonitrile-TEA (10:90:0.1, v/v/v) and the pH was adjusted with o-phosphoric acid to 3.5. It was sonicated for 15 min and filtered through 0.22 µm membrane filter. Flow rate of mobile phase was maintained at 2 mL/min and eluents were monitored at 230 nm. The samples were injected using a 20 µL HPLC injector. All determinations were performed at ambient temperature for a run time of 5 min [22].

Preparation of calibration curve: The 1.0 mg/mL stock solution of haloperidol was prepared in mobile phase. An appropriate volume of stock solution was further diluted with mobile phase to obtain a standard solution having a final concentration 100 µg/mL. Different

Application of HPLC Method in *in-vitro* drug release study

Haloperidol loaded solid lipid nanoparticles (HP-SLNs) were prepared by modified solvent emulsification–diffusion technique and optimised by Boxbehken design [13].

The *in vitro* dissolution studies were carried out to evaluate the release of drug from optimized HP-SLN formulation and comparing it with the pure drug.

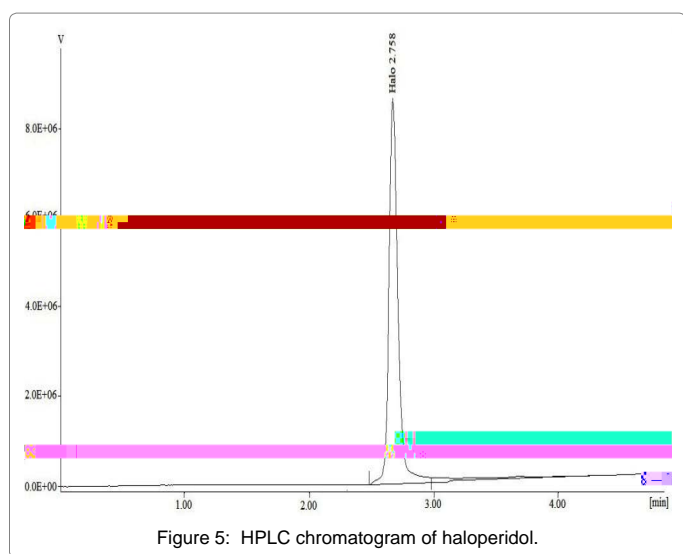
An accurate amount of freeze dried HP-SLNs and aqueous haloperidol suspension, each containing drug equivalent to 10 mg was transferred to a dialysis bag and sealed at both ends. The sealed bag was then suspended in a beaker containing 100 mL of dissolution medium (Phosphate buffer pH 7.4, corresponding to cerebrospinal fluid pH) and stirred at a constant speed at $37 \pm 0.5^\circ\text{C}$. Aliquots were withdrawn at predetermined time intervals up to 24 h from receiver compartment (beaker) and replaced with an equal volume of fresh medium to maintain sink condition. The samples were analyzed by HPLC using the selected mobile phase. Comparison between release profile of drug suspension and optimized HP-SLN formulation was made by student's *t* test at $p < 0.05$.

Analysis of haloperidol by HPLC method for the determination of shelf life

The shelf life of optimized HP-SLNs was determined by

% of standard spiked to the sample	Concentration			% drug recovered	% RSD	SE

Conc. ($\mu\text{g/ mL}$)	Repeatability (intraday precision)			M p 6 ĩ			



References

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