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Development and Validation of Stability Indicating Assay Method of Haloperidol in Oral Solution

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ABSTRACT:

The present research work aims to develop a simple, precise, accurate, rapid, reproducible and economical method for the estimation of Haloperidol by RP-HPLC method. An absorbance maximum for Haloperidol was found to be at 254 nm using methanol as a solvent. The chromatography was performed on a Restek Pinnacle II C18 (250 mm x 4.6 mm i.d., 5 µm particle size) column with mobile phase containing Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45 v/v). The flow rate was 1 ml/min and the eluent was monitored at 254 nm. The selected chromatographic conditions were found effectively to separate Haloperidol at 7.707 min, Methyl Hydroxyl Benzoate (MHB) at 5.323 min and Propyl Hydroxy Benzoate (PHB) at 12.492 min. Linearity was found in the range of 20-200 µg/ml for Haloperidol, 20-200 µg/ml for MHB and 20-200 µg/ml for PHB. The obtained correlation coefficient was 0.999. The accuracy was evaluated by recovery study and recovery result was obtained between 98.8% to 100.8% and the relative standard deviation below 2% was achieved. The value obtained for LOD was 0.90 µg/ml, 1.18 µg/ ml, 0.08 µg/ml and LOQ was 2.75 µg/ml, 3.58 µg/ml, 2.62 µg/ml for Haloperidol, MHB and PHB simultaneously. The proposed method was found to be fast, accurate, precise, simple, sensitive and reproducible for analysis of Haloperidol in Oral Solution. Haloperidol was also subjected to stress conditions of acid hydrolysis, base hydrolysis, oxidation, photolysis and thermal degradation under same chromatographic condition. From all stability results, it is concluded that stability study of Haloperidol can be apply to oral solution sample of haloperidol.

Keywords: Haloperidol, MHB, PHB, RP-HPLC, Validation.

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INTRODUCTION:

Haloperidol (antipsychotic drug) Antipsychotics(also known as **neuroleptics** or **major tranquilizers**) are a class of psychiatric medication primarily used to manage psychosis (including delusions, hallucinations, or disordered thought), particularly in schizophrenia and bipolar disorder, and is increasingly being used in the management of non-psychotic disorders.

Classification : It is divided in two types. (1) Typical Anti-psychoticsAtypical and (2) Anti-psychotics

Basic Information

Schizophrenia: Schizophrenia is a debilitating disorder of the central nervous system. This disorder reduces the ability of the individual to interact with the society.Its symptoms have been divided into two classes: (1) Positive symptoms including hallucinations, delusions and conceptual disorganization. (2) Negative symptoms including social withdrawal, blunted affect, and poverty of speech.

Antipsychotics are most frequently used for the following conditions: Schizophrenia, Bipolar disorder and Psychotic depression.

Materials and Methods

Methanol, HPLC Grade/ AR Grade was obtained from Merck specialties pvt. Ltd., Mumbai and Tetrabutyl ammonium hydrogen sulphate, HPLC Grade from Merck specialties pvt. Ltd., Mumbai. Water, HPLC Grade obtained from Rankem Pvt Ltd.

RP HPLC METHOD DEVELOPMENT

Establishment of the optimum conditions of the HPLC method

Selection of column

Satisfactory results was obtained with Restek Pinnacle II C18 (250 × 4.6mm, 5µ) column. It was observed that better resolution, peak shape and Rt were obtained using this column.

Selection of wavelength for detection

Standard solution of Haloperidol, MHB and PHB were scanned and three different λ_{max} (230, 247 and 254) were obtained but at 254 nm responses were in good agreement compared to 230 nm and 247 nm. So, 254 nm was selected as λ_{max} of Method development.

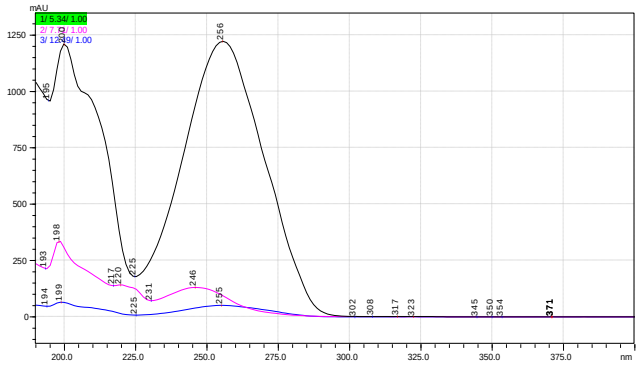


Figure 1 Spectra of Haloperidol, MHB and PHB.

Table 1 λmax in n.m.

Sr. no.	Drug	λmax (nm)
1.	Haloperidol	246
2.	Methyl Hydroxy Benzoate	256
3.	Propyl Hydroxy Benzoate	255
4.	Selected Wavelength	254

Selection of Mobile phase

On the basis of various trials, the mixture of METHANOL: TETRABUTYL AMMONIUM HYDROGEN SULPHATE (55: 45 v/v) at 1 ml/min flow rate, proved to be better than the other mixtures in terms of resolution, peak shape and asymmetry.

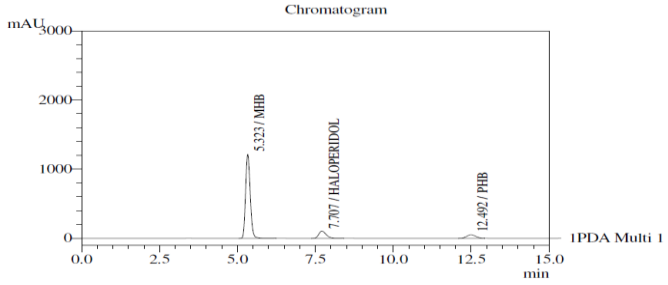


Figure 2 Chromatogram of Haloperidol, MHB, PHB

Table 2 Rt values in minut

Sr. No	Drug	Rt Value (Min)
1.	Haloperidol	7.707
2.	Methyl Hydroxy Benzoate	5.323
3.	Propyl Hydroxy Benzoate	12.492

Optimization of flow rate

1.0 ml/min flow rate, proved to be better than the other in terms of resolution, peak shape and shorter retention time.

Preparation of Mobile Phase

Mobile phase was prepared by mixing 55 ml Methanol, 45 ml Tetrabutyl ammonium hydrogen sulphate. The mobile phase was sonicated for 5 min and then it was filtered through 0.45 µm membrane filter paper.

Preparation of stock solution

Take 200 mg of Haloperidol reference standard and dilute it up to 100 ml with Diluent Mobile phase, take 400 mg of Methyl Hydroxy Benzoate reference standard and dilute it up to 100 ml with Diluent Mobile phase, take 20 mg of Propyl Hydroxy Benzoate reference standard and dilute it up to 50 ml with Diluent Mobile phase, Then take 25 ml from each in a flask and dilute it up to 100 ml with diluent and further take 2 ml from it and dilute it up to 10 ml and sonicate it for 15 minute. (100µg/ml)

Preparation of Sample solution

Take 2 ml from sample and dissolve it in diluent mobile phase up to 20 ml and filter it with nylon filter. (100µg/ml)

Table 3 RP-HPLC optimized chromatographic conditions

Parameters	Chromatographic condition
Mode of elution	Isocratic
Mobile Phase	Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45 v/v)
Column	RESTEK Pinnacle II C18 (4.6 x 250 mm); 5 µm
Column Oven Temperature	25 ºC
Flow rate	1 ml/min
Run time	15 min
Injection volume	10 µl
Detection wavelength	254 nm

METHOD VALIDATION

Linearity and range

The linearity of an analytical method is its ability to elicit test results that are directly or by a well-defined mathematical transformation proportional to the concentration of analyte in samples within a given range. Result should be expressed in terms of correlation co-efficient.

Preparation of Calibration curve

Haloperidol

A calibration curve was plotted over a concentration range of 20-200 µg/ml for Haloperidol. Accurately measured working standard stock solution of Haloperidol (0.4, 1.0, 1.5, 2.0, 3.0 and 4.0 ml) were transferred to a series of 10 ml volumetric flasks and the volume in each flask made up to the mark with Mobile Phase to get concentration range of 20-200 µg/ml for Haloperidol . The resulting solution was injected into the column and the peak area obtained at retention time 7.707 minute at flow rate 1 ml/min were measured at 254 nm for Haloperidol. Calibration curve was constructed by plotting peak area versus concentration at 254 nm. A representative chromatogram has been shown in Figure. Better Shape of the peak with clear base line separation was found.

Methyl Hydroxy Benzoate

A calibration curve was plotted over a concentration range of 40-400 µg/ml for Methyl Hydroxy Benzoate. Accurately measured working standard stock solution of Methyl Hydroxy Benzoate (0.4, 1.0, 1.5, 2.0, 3.0 and 4.0 ml) were transferred to a series of 10 ml volumetric flasks and the volume in each flask made up to the mark with Mobile Phase to get concentration range of 40-400 µg/ml for Methyl Hydroxy Benzoate. The resulting solution was injected into the column and the peak area obtained at retention time 5.323 minute at flow rate 1 ml/min were measured at 254 nm for Methyl Hydroxy Benzoate. Calibration curve was constructed by plotting peak area versus concentration at 254 nm. A representative chromatogram has been shown in Figure. Better Shape of the peak with clear base line separation was found.

Proyl Hydroxy Benzoate

A calibration curve was plotted over a concentration range of 4-40 µg/ml for Proyl Hydroxy Benzoate. Accurately measured working standard stock solution of Proyl Hydroxy Benzoate (0.4, 1.0, 1.5, 2.0, 3.0 and 4.0 ml) were transferred to a series of 10 ml volumetric flasks and the volume in each flask made up to the mark with Mobile Phase to get concentration range of 4-40 µg/ml for Proyl Hydroxy Benzoate. The resulting solution was injected into the column and the peak area obtained at retention time 12.492 minute at flow rate 1 ml/min were measured at 254 nm for Proyl Hydroxy Benzoate. Calibration curve was constructed by plotting peak area versus concentration at 254 nm. A representative chromatogram has been shown in Figure. Better Shape of the peak with clear base line separation was found.

Precision

The precision is measure of either the degree of reproducibility or repeatability of analytical method.

A. Repeatability (Precision on replication)

It is a precision under a same condition (Same analyst, same apparatus, short interval of time and identical reagents) using same sample.

B. Interday and Intraday precision

It expresses within laboratory variations as on different days analysis or equipment within the laboratory. Variation of results within same day is called Intra-day precision and variation of results amongst days called Inter-day precision.

Accuracy (% Recovery)

Accuracy of an analysis is determined by systemic error involved. It is defined as closeness of agreement between the actual (true) value and analytical value and obtained by applying test method for a number of times. Accuracy may often be expressed as % Recovery by the assay of known, added amount of analyte. It is measure of the exactness of the analytical method.

Limit of Detection

It is the lowest amount of analyte in a sample that can be

detected but not necessarily quantitated under the stated experimental conditions. Limit of detection can be calculated using following equation as per ICH guidelines.

LOD = 3.3 × N/S

Where, N is the standard deviation of the peak areas of the drug and S is the slope of the corresponding calibration curve.

Limit of Quantification

It is the lowest concentration of analyte in a sample that can be determined with the acceptable precision and accuracy under stated experimental conditions. Limit of quantification can be calculated using following equation as per ICH guidelines.

LOQ = 10 × N/S

Where, N is the standard deviation of the peak areas of the drug and S is the slope of the corresponding calibration curve.

Robustness

Robustness was carried by varying experimental parameters of proposed method. In case of liquid chromatography typical variations are the pH of the mobile phase, the mobile phase composition, and flow rate. No significant change was observed.

System suitability

System suitability parameter is establishes to ensure that the validity of the analytical method is maintained whenever used.

RESULTS AND DISCUSSION

Various mobile phase compositions were tried and the best results were obtained with mobile phase was Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45) and the flow rate was 1 ml/min, detection wavelength was 254 nm and the retention time was 7.707 min for Haloperidol, Methyl Hydroxy Benzoate was 5.323 min, Proyl Hydroxy Benzoate was 12.492 min.

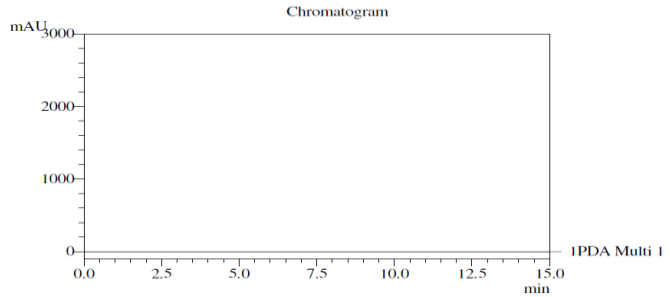


Figure 3 HPLC chromatogram of Blank Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45)

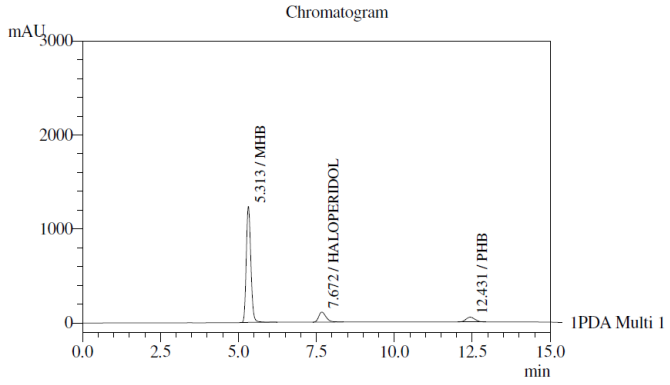


Figure 4 HPLC chromatogram of standard solution

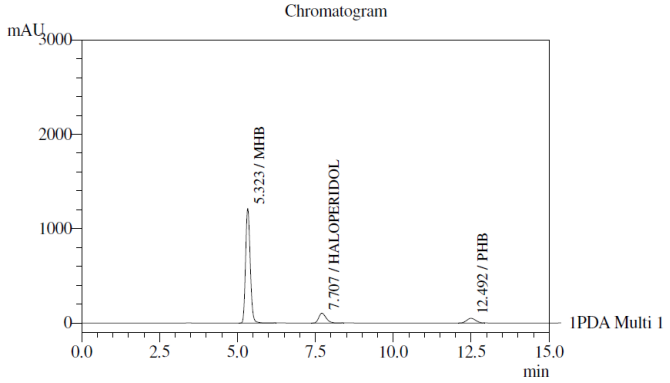


Figure 5 HPLC chromatogram of test solution

Validation

Linearity and Range

The linear response of Haloperidol was determined by analyzing 5 independent levels of the calibration curve in the range of 20 to 200 µg/ml for Haloperidol.

The linear response of Methyl Hydroxy Benzoate was determined by analyzing 5 independent levels of the calibration curve in the range of 40 to 400 µg/ml for Methyl Hydroxy Benzoate.

The linear response of Proyl Hydroxy Benzoate was determined by analyzing 5 independent levels of the calibration curve in the range of 4 to 40 µg/ml for Proyl Hydroxy Benzoate.

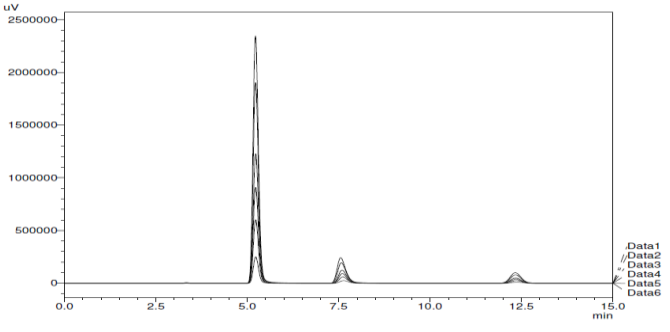


Figure 6 Chromatogram of Linearity of Haloperidol, MHB, PHB.

Linearity curve of Haloperidol

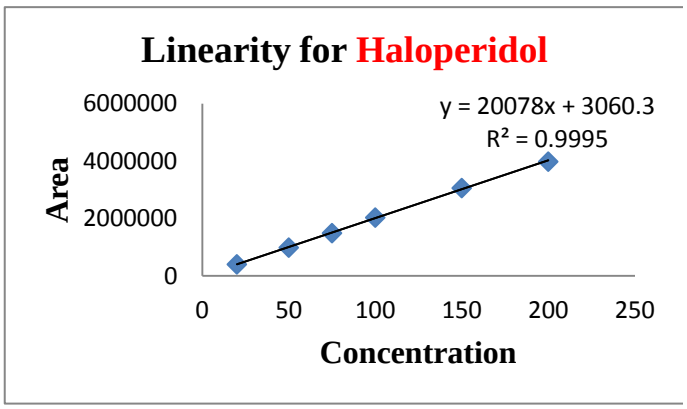


Figure 7 Calibration Graph for Haloperidol (Area vs Concentration)

Linearity curve of MHB

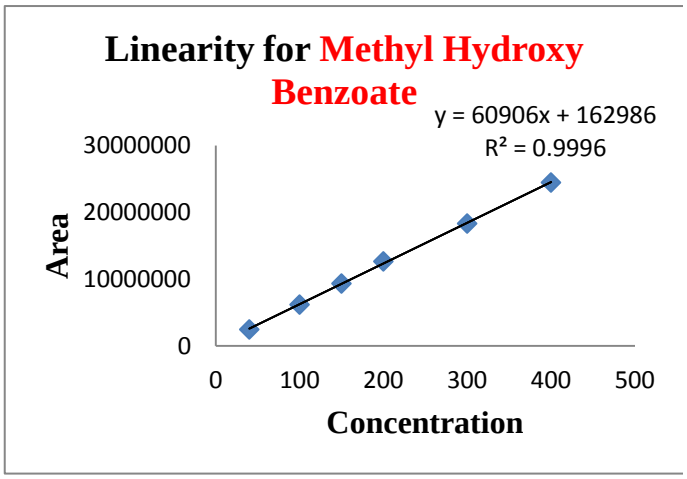


Figure 8 Calibration Graph for Methyl Hydroxy Benzoate (Area vs Concentration)

Linearity curve of PHB

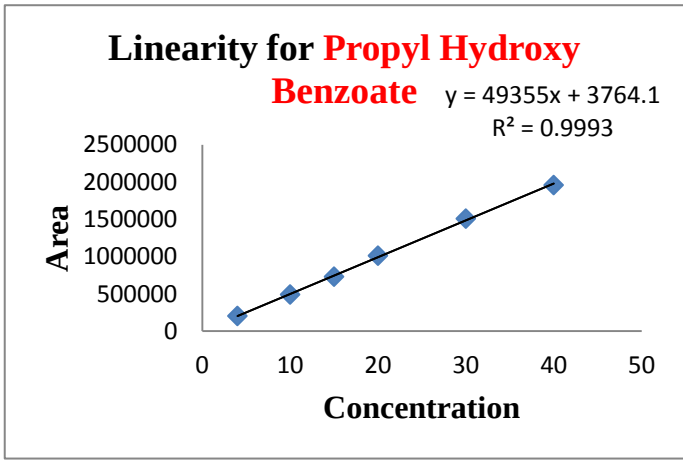


Figure 9 Calibration Graph for Proyl Hydroxy Benzoate (Area vs Concentration)

Precision

The intra-day and inter-day variation for determination of Haloperidol was carried out five times in the same day and five consecutive days. % RSD were calculated. The method was found to be precise due to low values of the %RSD (< 2 %).

Table 4 Method precision study of Haloperidol

Sr. No.	Concentration (µg/ml)	Peak area
1	100	1939436.5
2	100	1940453.5
3	100	1938990.5
4	100	1933094
5	100	1942100.5
6	100	1947492.5
Mean		1940261.25
S.D.		4677.07
%R.S.D.		0.24

Table 5 Method precision study of MHB

Sr. No.	Concentration (µg/ml)	Peak area
1	100	12053347
2	100	12043168
3	100	12032767.5
4	100	12053454
5	100	12040807
6	100	12022407.5
Mean		12040991.83
S.D.		12047.76
%R.S.D.		0.1

Table 7 Method precision study of PHB

Sr. No.	Concentration (µg/ml)	Peak area
1	100	1941497
2	100	1935107.5
3	100	1939619
4	100	1934006.5
5	100	1933736.5
6	100	1934613.5
Mean		1936430
S.D.		3286.98
%R.S.D.		0.17

Table 8 Intermediate precision study of Haloperidol

Concentration (µg/ml)	Intra-Day Precision		Inter-Day Precision	
	Mean ± S.D (n=5)	%RSD	Mean ± S.D (n=5)	%RSD
20	405401.5 ± 5513.46	1.36	407416 ± 5581.599	1.37
50	988120.5 ± 6126.34	0.62	988731 ± 6921.117	0.70
75	1492794.5 ± 25228.22	1.69	1495747 ± 19893.44	1.33
100	2034052 ± 5898.75	0.29	2029756 ± 12787.46	0.63
150	3062241 ± 22660.58	0.74	3037729 ± 29769.74	0.98
200	3982058.5 ± 47386.49	1.19	3949398 ± 61215.67	1.55

Table 9 Intermediate precision study of MHB

Concentration (µg/ml)	Intra-Day Precision		Inter-Day Precision	
	Mean ± S.D (n=5)	%RSD	Mean ± S.D (n=5)	%RSD
40	2475062.5 ± 21780.55	0.88	2457487 ± 12041.69	0.49
100	6191536 ± 58819.59	0.95	6150778 ± 55357	0.90
150	9346551 ± 166368.6	1.78	9456486 ± 156032	1.65
200	12655698.5 ± 70871.91	0.56	14012387 ± 124710.2	0.89
300	18325707.5 ± 82465.68	0.45	19725808 ± 116382.3	0.59
400	24461479 ± 369368.3	1.51	25032597 ± 430560.7	1.72

Accuracy (% Recovery)

Accuracy of the method was determined by % recovery method. Recovery studies were carried out by addition of standard drug solution at the level of 50%, 100% and 150% to the pre analyzed sample. In this method the known concentration standard drug was added to the assay sample and total amount was found. The average % recoveries for Haloperidol, MHB, PHB are shown in Tables.

Table 10 Intermediate precision study of PHB

Concentration (µg/ml)	Intra-Day Precision		Inter-Day Precision	
	Mean ± S.D (n=5)	%RSD	Mean ± S.D (n=5)	%RSD
4	201233.5 ± 1187.2	0.59	2047498 ± 11056.49	0.54
10	489189 ± 4255.9	0.87	4850658 ± 41230.59	0.85
15	728732 ± 8088.9	1.11	7255569 ± 42807.86	0.59
20	1010522 ± 9903.1	0.98	10014348 ± 123176.5	1.23
30	1507662 ± 11307.4	0.75	15895756 ± 106501.6	0.67
40	1958470.5 ± 16647	0.85	1907535 ± 22890.42	1.20

Table 11 Recovery study of Haloperidol

Drug	Amount taken (µg/ml)	Amount added (µg/ml)	Recovered std. concentration (µg/ml)	%Recovery± S.D (n=3)
HALOPERIDOL	100	50	50.43	100.8 ± 0.7999
	100	100	98.76	98.8 ± 0.5669
	100	150	149.57	100 ± 0.6704

Table 12 Recovery study of MHB

Drug	Amount taken (µg/ml)	Amount added (µg/ml)	Recovered std. concentration (µg/ml)	%Recovery± S.D (n=3)
MHB	200	100	101.3	100.5 ± 0.5879
	200	200	200.5	100.4 ± 0.3452
	200	300	300.5	100.5 ± 0.5221

Table 13 Recovery study of PHB

Drug	Amount taken (µg/ml)	Amount added (µg/ml)	Recovered std. concentration (µg/ml)	%Recovery± S.D (n=3)
PHB	20	10	10.4	101.5 ± 0.5556
	20	20	21.2	101.4 ± 0.4432
	20	30	31.3	101.5 ± 0.5578

LOD

Limit of detection for Haloperidol was found to be **0.9062 µg/ml.**

Limit of detection for MHB was found to be **1.1801 µg/ml**.

Limit of detection for PHB was found to be **0.0794 µg/ml**.

LOQ

Limit of quantitation for Haloperidol was found to be 2.7460 **µg/ml**.

Limit of quantitation for MHB was found to be 3.5761 **µg/ml**.

Limit of quantitation for PHB was found to be 2.6195 **µg/ml**.

Robustness

In present study robustness was carried out by small changes of flow rate, wavelength and mobile phase composition and % RSD was calculated. Which indicates method is precise.

Table 14 Robustness of Haloperidol

Factor	Value	Area
Flow Rate	**0.8**	2457343.5
	1.2	1655068
	MEAN ± S.D	2056205.75 ± 567294.4
	%RSD	0.27
Temperature	**23°C**	1898248
	27°C	1952345.5
	MEAN ± S.D	1283531.26 ± 38252.71
	%RSD	0.29
Mobile phase (-5%, +5%)	**52.25:45.75**	1711090
Methanol: Tetrabutyl ammonium hydrogen sulphate(55:45 v/v)	**57.75:42.25**	1715453.5
	MEAN ± S.D	1142181.26 ± 3085.46044
	%RSD	0.27

Table 15 Robustness of MHB

Factor	Value	Area
Flow Rate	**0.8**	15128176
	1.2	10095673.5
	MEAN ± S.D	1211924.75± 3558516.644
	%RSD	0.28
Temperature	**23**	11946905.5
	27	11984505.5
	MEAN ± S.D	11965706±38252.71
	%RSD	0.22
Mobile phase (-5%, +5%)	**52.25:45.75**	12595017
Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45 v/v)	**57.75:42.25**	12592132
	MEAN ± S.D	12593575± 2040.003
	%RSD	0.01

Table 16 Robustness of MHB

Factor	Value	Area
Flow Rate	**0.8**	**1272130.5**
	1.2	**849215.5**
	MEAN ± S.D	**1060673± 299046.1**
	%RSD	**0.28**
Temperature	**23**	**984353.5**
	27	**983447**
	MEAN ± S.D	**983900.3±640.9923**
	%RSD	**0.06**
Mobile phase (-5%, +5%)	**52.25:45.75**	**1012531**
Methanol: Tetrabutyl ammonium hydrogen sulphate (55:45 v/v)	**57.75:42.25**	**1023840**
	MEAN ± S.D	**1018186±7996.671**
	%RSD	**0.79**

System suitability testing

Parameters are shown in below table. No 17

Table 17 System suitability parameter

Parameters	Haloperidol	MHB	PHB
Retention Time	7.707 min	5.323	12.492
Theoretical plate	4946.007	5794.69	7496.981
Tailing factor	1.324	1.175	1.079

Conclusion

All these factors lead to the conclusion that the proposed method is accurate, precise, simple and sensitive and can be applied successfully for the estimation of Haloperidol in oral solution without interference.

STABILITY STUDY

Determination of wavelength for maximum absorbance

Standard solution of Haloperidol (10, 20, 30 µg/ml) were scanned (Figure 5.3) and three different λ_{max} (230, 247 and 254) were obtained but at 254 nm responses were in good agreement compaired to 230 nm and 247 nm. So, 254 nm was selected as $\lambda_{max}$of Haloperidol.

Chromatographic condition

Chromatographic separation was carried out on Shimadzu HPLC (2010 HT) System using Restek Pinnacle II C18 (250 mm x 4.6 mm i.d., 5 µm particle size) column as stationary phase and mobile phase containing Methanol: Tetrabutyl ammonium hydrogen sulphate: Isopropyl alcohol (55:35:10) at flow rate of 1 ml/min using UV detection at 254 nm.

Preparation of Mobile Phase

Mobile phase was prepared by mixing 55 ml Methanol, 35 ml Tetrabutyl ammonium hydrogen sulphate, 10 ml Isopropyl alcohol. The mobile phase was sonicated for 5 min and then it was filtered through 0.45 µm membrane filter paper.

Preparation of stock solution (100 µg/ml)

Take 200 mg of Haloperidol reference standard and dilute it up to 100 ml with Diluent Mobile phase, take 400 mg of Methyl Hydroxy Benzoate reference standard and dilute it up to 100 ml with Diluent Mobile phase, take 20 mg of Propyl Hydroxy Benzoate reference standard and dilute it up to 50 ml with Diluent Mobile phase, Then take 25 ml from each in a flask and dilute it up to 100 ml with diluent and further take 2 ml from it and dilute it up to 10 ml and sonicate it for 15 minute. (100µg/ml)

Preparation of hydrochloric acid solution (0.1 N)

Concentrated hydrochloric acid (0.85 ml) was transferred in 100 ml volumetric flask and diluted up to the mark with water.

Preparation of sodium hydroxide solution (0.1 N)

Accurately weighed 0.4 gm of sodium hydroxide was transferred in 100 ml volumetric flask and diluted up to mark with water.

Preparation of hydrogen peroxide solution (3% w/v)

Hydrogen peroxide (10 ml, 30%) was transferred in 100 ml volumetric flask and diluted up to the mark with water.

Acid hydrolysis

Accurately measured 1 ml of Haloperidol Standard Stock Solution (100 µg/ml) was transferred in to10 ml volumetric flask and volume was made up to the mark with 0.1N HCl to get 10 µg/ml of Haloperidol Standard Stock Solution and solution was heated for 2 hours at 80° C for acid hydrolysis. Then neutralize with 0.1N NaOH and Filtered through 0.45 µm membrane filter paper and injected in to HPLC system.

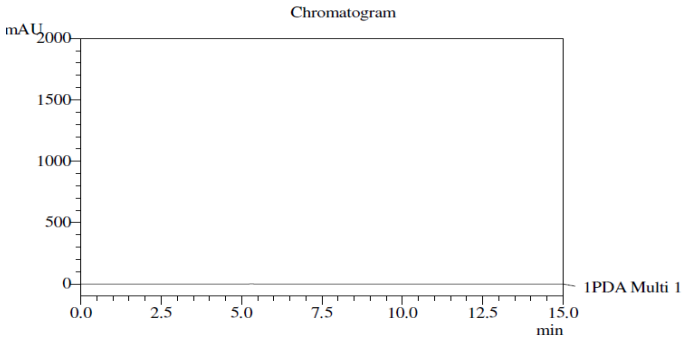


Figure 10 Chromatogram of Diluent under Acidic condition

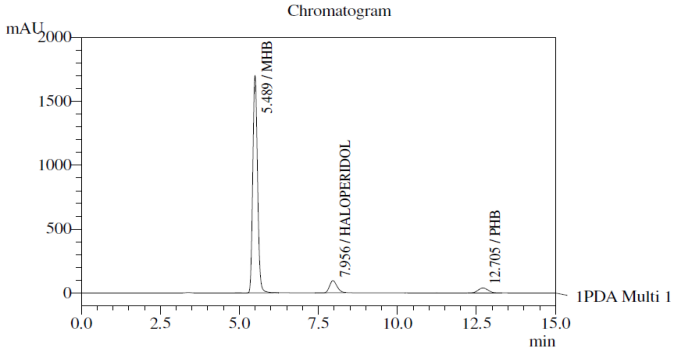


Figure 11 Chromatogram of Haloperidol, MHB, PHB under Acidic condition

Base hydrolysis

Accurately measured 1 ml of Haloperidol Standard Stock Solution (100 µg/ml) was transferred in to10 ml volumetric flask and volume was made up to the mark with 0.1N NaOH to get 10 µg/ml of Haloperidol Standard Stock Solution and solution was heated for 2 hours at 80° C for base hydrolysis. Then neutralize with 0.1N HCL and Filtered through 0.45 µm membrane filter paper and injected in to HPLC system.

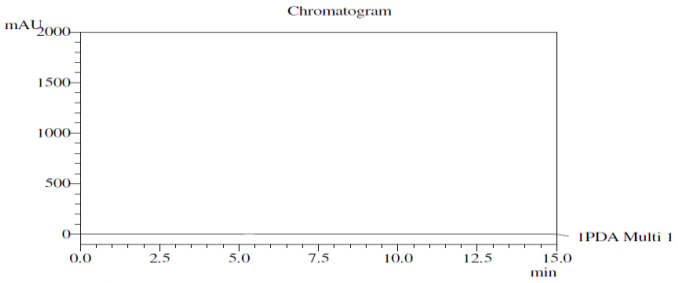


Figure 12 Chromatogram of Diluent under Basic condition

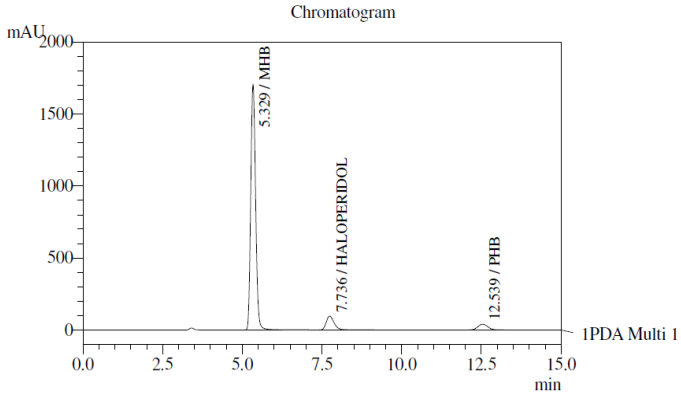


Figure 13 Chromatogram of Haloperidol, MHB, PHB under Basic condition

Oxidative hydrolysis

Accurately measured 1 ml of Haloperidol Standard Stock Solution (100 µg/ml) was transferred in to10 ml volumetric flask and volume was made up to the mark with 3% H_2O_2 to get 10 µg/ml of Haloperidol Standard Stock Solution and solution was heated for 2 hours at 80° C for oxidative hydrolysis. Filtered through 0.45 µm membrane filter paper and injected into HPLC system.

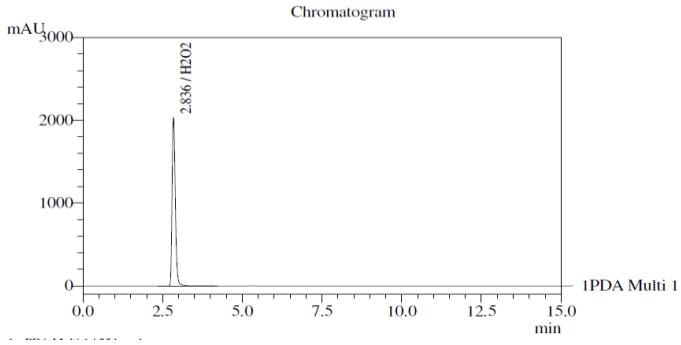


Figure 14 Chromatogram of Diluent under Oxidative

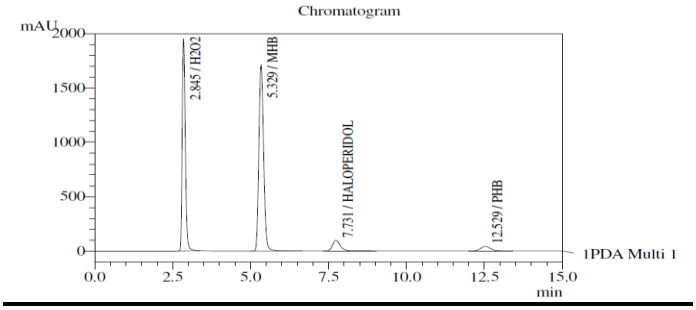


Figure 15 Chromatogram of Haloperidol, MHB, PHB under Oxidative condition

Thermal Degradation

For dry heat degradation study, powder of Haloperidol, Methyl Hydroxy Benzoate, Propyl Hydroxy Benzoate were spread over petri dishes and exposed to dry heat (80˚C) for 12 hour in an oven then from that powder an accurately weighed quantity of Haloperidol (200 mg), Methyl Hydroxy Benzoate (400 mg), Propyl Hydroxy Benzoate (20 mg) were transferred to 100 ml, 100ml and 50 ml volumetric flask simultaneously and dissolved in mobile phase and make up the volume. Take 25 ml from each in a 100 ml volumetric flask and make up the volume with mobile phase. Then take 2 ml from it in 10 ml volumetric flask and make up the volume with mobile phase to get 100 µg/ml of Haloperidol standard stock solution. Further dilution was carried out by diluting 1 ml of solution in 10 ml volumetric flask with mobile phase to get 10 µg/ml of Haloperidol standard stock solution.

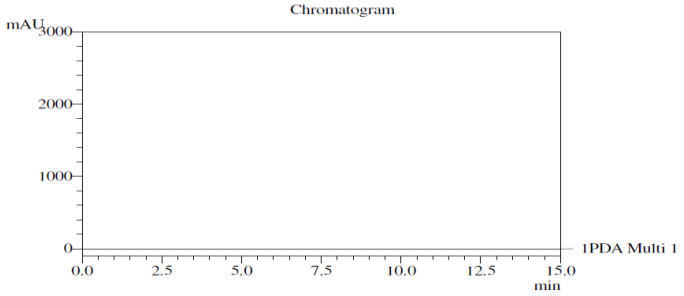


Figure 16 Chromatogram of Diluent under Thermal Degradation

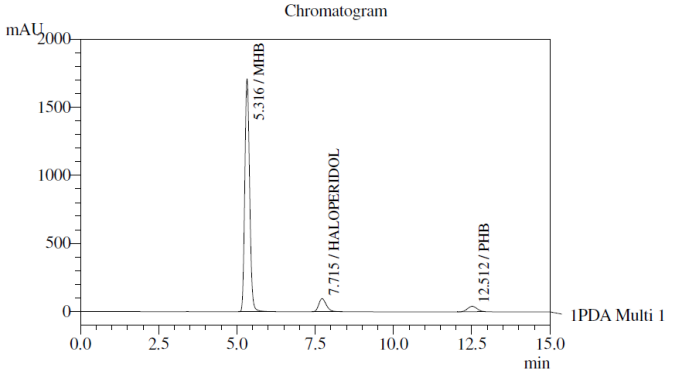


Figure 17 Chromatogram of Haloperidol, MHB, PHB under Thermal Degradation

Photolytic Degradation

For photolytic degradation, powder of Haloperidol, Methyl Hydroxy Benzoate, Propyl Hydroxy Benzoate were put in UV radiation chamber for 2 days. Accurately weighed quantity of Haloperidol (200 mg), Methyl Hydroxy Benzoate (400 mg), Propyl Hydroxy Benzoate (20 mg) were transferred to 100 ml, 100ml and 50 ml volumetric flask simultaneously and dissolved in mobile phase and make up the volume. Take 25 ml from each in a 100 ml volumetric flask and make up the volume with mobile phase. Then take 2 ml from it in 10 ml volumetric flask and make up the volume with mobile phase to get 100 µg/ml of Haloperidol standard stock solution. Further dilution was carried out by diluting 1 ml of solution in 10 ml volumetric flask with mobile phase to get 10 µg/ml of Haloperidol standard stock solution.

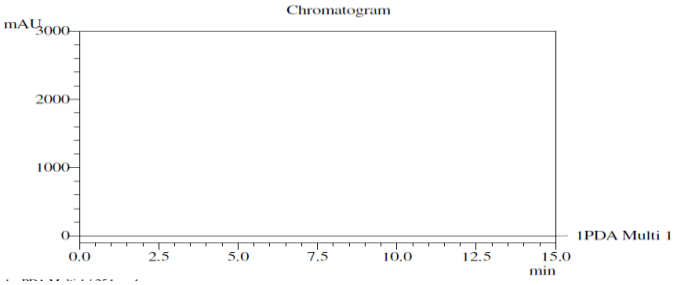
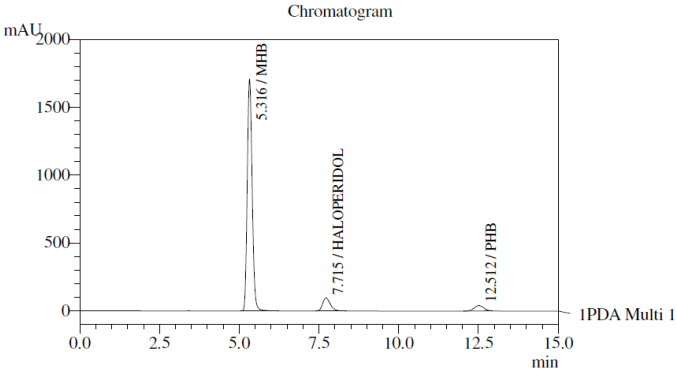


Figure 18 Chromatogram of Diluent under Photolytic Degradation



. Figure 19Chromatogram of Haloperidol, MHB, PHB under Photolytic Degradation

Table Result of Stability study

Condition of forced degradation	%Degradation Haloperidol
0.1N HCL, 2 hr, 80° C	12.96
0.1N NaOH, 2 hr, 80° C	16.14
3% H_2O_2, 2 hr, 80° C	18.83
Thermal,80° C 12 hours	10.95
UV Light, 2 days	10.02

RESULT AND DISCUSSION

Major degradation was not observed in Haloperidol, Methyl Hydroxy Benzoate, Propyl Hydroxy Benzoate which was subjected to light, heat and acid hydrolysis. Result of Stability study is shown in table 6.2

CONCLUSION

From all above results, it is concluded that stability study of Haloperidol can be pply to oral solution of Haloperidol.

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