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STABILITY INDICATING METHOD DEVELOPMENT AND VALIDATION FOR THE SIMULTANEOUS ESTIMATION OF LOBEGLITAZONE SULFATE AND GLIMEPIRIDE IN BULK AND TABLET DOSAGE BY UV SPECTROPHOTOMETRIC METHOD

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ABSTRACT
UV-spectrophotometric methods have been developed and validated for the Simultaneous estimation of Lobeglitazone sulfate (LOBE) and Glimepiride (GLIM) in combined dosage form. In the simultaneous equation method LOBE and GLIM have absorbance maxima at 251nm and 228 nm respectively in methanol. The method involves First order derivative spectroscopy using 265 nm and 241 nm as zero crossing points for LOBE and GLIM respectively. Linearity was observed in the concentration range of LOBE is 2-10 µg/ml and GLIM is 4-20 µg/ml for Simultaneous equation method and first order derivative method the concentration range of LOBE is 3-15 µg/ml and GLIM is 6-30 µg/ml The correlation coefficient was found to be 0.9998 for LOBE and 0.9997 for GLIM for Simultaneous equation method and 0.9998 for LOBE and 0.9998 for GLIM for First order derivative method, respectively. The precision (intraday, inter day) of methods was found within limits (RSD < 2%). Method showed good reproducibility and recovery with % RSD less than 2%. The samples of LOBE and GLIM were subjected to stress conditions like acidic, alkaline, oxidation, photolysis degradation. It could be concluded from the results obtained in the present investigation that the methods for simultaneous estimation of LOBE and GLIM in bulk and tablet dosage form are simple, rapid, accurate, precise and economical and can be used, successfully, in the quality control of tablet formulations and other routine laboratory analysis and the stability studies were successfully applied as per ICH guidelines.

KEYWORDS: Lobeglitazone sulfate, Glimepiride, Simultaneous equation method, first order derivative method, Forced degradation studies.

I - INTRODUCTION
Lobeglitazone sulfate is chemically 5 - [[4 - [2- [[6- (4-methoxyphenoxy) pyrimidin – 4 - yl]-methylamino] ethoxy] phenyl] methyl] - 1, 3 – thiazolidine - 2, 4 - dione; sulfuric acid. Glimepiride is chemically 3 – Ethyl – 4 – methyl – N - [2 - (4 - {[(Trans – 4 – methylcyclohexyl) carbomoyl] sulfamoyl} phenyl) ethyl] - 2 – oxo - 2, 5 – dihydro - 1H –pyyrole – 1 - carboxamide. Lobeglitazone sulfate and Glimepiride are Anti-diabetic drugs. Lobeglitazone is an antidiabetic medication from the thiazolidinedione class of drugs. It primarily functions as an insulin sensitizer by binding and activating Peroxisome Proliferator-Activated Receptors (PPAR) gamma within fat cells. It including glucose and lipid metabolism. The primary mechanism action of glimepiride in lowering blood glucose appears to be dependent on stimulating the release of insulin from functioning pancreatic beta cells. On literature survey, several methods were reported for the estimation of LOBE and GLIM individually and in combination with other drugs. So we have developed a novel, simple, rapid, accurate, precise, economical and highly sensitive UV spectrophotometric methods for LOBE and GLIM in bulk and tablet dosage form and validated according to ICH guidelines.

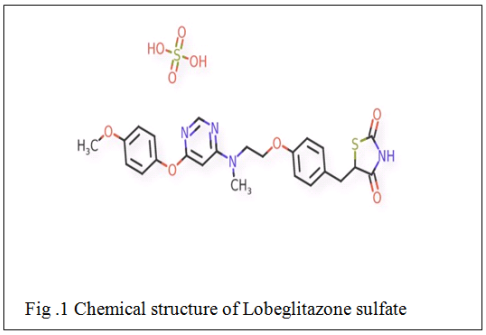

Fig .1 Chemical structure of Lobeglitazone sulfate

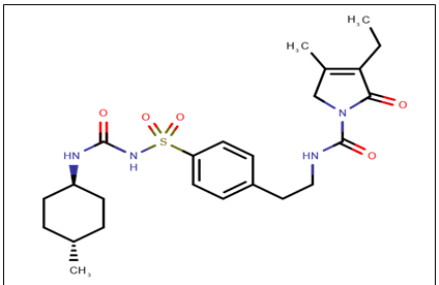

Fig. 2 Chemical structure of Glimepiride

II -MATERIALS AND METHODS

Instrumentation: The instrument used in the present study was Shimadzu double beam UV/Visible spectrophotometer (Model UV1700) with spectral band width of 1nm. All weighing was done on electronic balance (Model Shimadzu AUX - 220).

Reagents and Chemicals: Analytically pure sample of Lobeglitazone sulfate was procured from Akhums drugs and Pharmaceutical Ltd New Delhi Glimepiride was purchased from A to Z Pharmaceuticals PVT Chennai. The pharmaceutical dosage form used in this study was a LOBG-G1 tablet manufactured by Glenmark Pharmaceutical Ltd. Lobeglitazone equivalent to 0.5mg and Glimepiride equivalent to 1mg was purchased from Sayar Pharmaa Distributors, Chennai.

Preparation of stock standard solutions: Stock standard solution of LOBE (500 μg/ml) and GLIM (1000 μg/ml) were prepared by dissolving 50 mg of LOBE and 100 mg of GLIM in 100 ml of Methanol in 100ml volumetric flask, separately with vigorous shaking.

Preparation of Working standard solution: From the stock standard solution, pipetted out 1 ml into 25 ml clean volumetric flask and diluted with Methanol to get the concentration for LOBE (20 μg/ml) and GLIM (40 μg/ml).

METHODS

1**. Simultaneous Equations:** Pure drug sample of LOBE and GLIM were dissolved separately in methanol so as to give several dilutions of standard in the concentration range 2-10µg/ml of LOBE and 4-20µg/ml of GLIM. All dilutions were scanned in the wavelength range of 400-200 nm. Fig-3 represents the overlain spectra of both the drugs. Two wavelengths selected for the formation of simultaneous equations were 251 nm and 228 nm (λ max of both the drugs respectively). The simultaneous equations formed were.

$$CX=A_1ay_2-A_2ay_1/ax_1ay_2-ax_2ay_1$$
$$CY=A_1ax_2-A_2ax_1/ay_1ax_2-ay_2ax_1$$

Where A1 and A2 are the absorbances of sample solution at 251 nm and 228 nm respectively. Cx and CY are the concentration of LOBE and GLIM respectively (µg/ml) in sample solution.

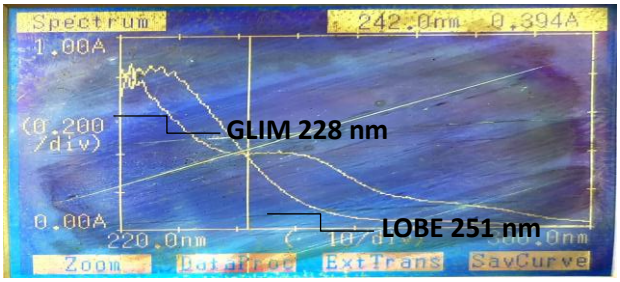

FIGURE 3: UV Overlaid spectrum for Lobeglitazone and Glimepiride.

2. First order derivative method: In this method, solutions of LOBE and GLIM (10µg/ml, each), were prepared separately by appropriate dilution of standard stock solution with methanol and scanned in the spectrum mode from 200 nm to 400 nm. The absorption spectra thus obtained were derivatized for first order. From the overlain spectra of these drugs, the wavelengths selected for quantitation were 226.0 nm for LOBE (Zero cross for LOBE) and 241.0 nm for GLIM (Zero cross for GLIM). The overlain first order derivative spectra of LOBE and GLIM is shown in Fig.No.4

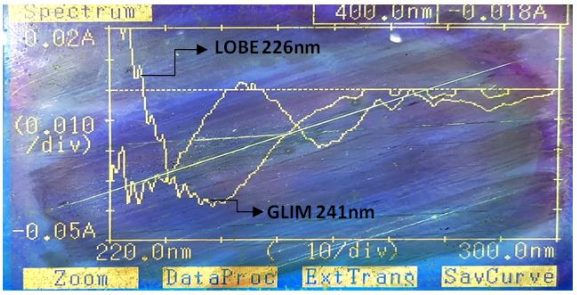

Fig. 4: First order derivative overlain spectra of 10µg/ml of LOBE and GLIM.

ANALYSIS OF TABLET FORMULATION

Twenty tablets were weighed accurately; the average weight was determined and then ground to fine powder. A quantity equivalent to 1 mg of GLIM was transferred to 25ml volumetric flask, sufficient methanol was added sonicated for 15 min and diluted to the mark with same solvent. It was filtered through Whatman Filter paper no: 41, filtrate was suitably diluted to get final concentration for LOBE (4µg/ml) and GLIM (8 µg/ml). Absorbance's were measured at the 251 nm & 228 nm wavelengths in the zero order and at the 226 nm & 241 nm in the first order, and amount present was calculated using simultaneous equation method and first order derivative method. Findings are tabulated in table 1.

Validation: The proposed methods were validated as per ICH guidelines.

Linearity: From the working standard solution of LOBE (1-5 ml) and GLIM (1-5 ml) were transferred into 10 ml volumetric flasks and made up to the volume to get 2-10 µg/ml and 4-20 µg/ml concentrations with methanol. The absorbance of different concentration solutions was measured at 251 nm and 228 nm in the normal spectrum LOBE and GLIM for simultaneous equation method. The calibration curve was plotted at their corresponding wavelengths. All two drugs Lobeglitazone and Glimepiride were linear with the concentration range of 2-10 µg/ml and 4-20 µg/ml respectively.

For first order derivative from the working standard solution of LOBE (1.5-7.5 ml) and GLIM(1.5-7.5 ml) were transferred into 10 ml volumetric flasks and made up to the volume to get 3-15 µg/ml and 6-30 µg/ml concentrations with methanol. The absorbance of different concentration solutions was measured at 226 nm and 241 nm in the normal spectrum LOBE and GLIM for First order derivative.

Accuracy: To the pre analysed sample solutions, a known amount of standard stock solution was added at different levels i.e. 50, 100 and 150 %. The solutions were re analysed by proposed method.

Precision: The reproducibility of the methods was determined by analysing tablets at different time intervals on same day in triplicates (Intra-day assay precision) and on three different days (Inter-day assay precision). And recovery studies were carried out by proposed method.

3. FORCED DEGRADATION STUDIES (STABILITY STUDIES)

PREPARATION OF SAMPLE STOCK SOLUTION:

Twenty tablets were weighed accurately; the average weight was determined and then ground to fine powder. A quantity equivalent to 1 mg of GLIM was transferred to 25ml volumetric flask, sufficient methanol was added sonicated for 15 min and diluted to the mark with same solvent. It was filtered through Whatman Filter paper no: 41, filtrate was suitably diluted to get final concentration for LOBE (4 µg/ml) and GLIM (8 µg/ml).

1. Hydrolytic degradation under acidic condition

Pipetted out 2.0 ml of sample stock solution into a 10 ml clean volumetric flask and added 3 ml of 0.1 N HCl. Then, the volumetric flask was kept at 60ºC for 24 hours then neutralized with 0.1 N NaOH and made up to 10 ml with methanol.

2. Hydrolytic degradation under alkaline condition

Pipetted out 2.0 ml of sample stock solution into a 10 ml clean volumetric flask and added 3 ml of 0.1 N NaOH. Then, the volumetric flask was kept at 60ºC for 24 hours then neutralized with 0.1 N HCl and made up to 10 ml with methanol.

3. Oxidative degradation

Pipetted out 2.0 ml of sample stock solution into a 10 ml clean volumetric flask and added 3 ml of 0.1%w/v of Hydrogen peroxide. Then, the volumetric flask was kept at 60ºC for24 hours then the volume was made up to the mark with methanol.

4. Photo degradation

Pipetted out 2.0 ml of sample stock solution into a 10 ml clean volumetric flask and expose to sunlight for 24 hours and methanol was added and made up to the mark.

4. RESULT AND DISCUSSION

The methods discussed in the present work provide a convenient and reliable way for quantitative determination of LOBE and GLIM in combined dose tablet formulation. For Simultaneous equation method the wavelength was selected 251 nm(λmax of LOBE)and also at 228 nm (λmax of GLIM). The absorption spectra thus obtained were derivatized to get first order. From the overlain spectra of these drugs, wavelength selected for quantitation were 226 nm and 241 nm in first order derivative, respectively. Percent label claim for LOBE and GLIM in tablet analysis was found in the range of 99.46 to 100.74% in Simultaneous method and 99.68 to 100.54% in first order derivative method. Percent recovery for LOBE and GLIM, was found in the range of 99.88 to 100.13% in Simultaneous method and 99.80 to 100.19% in first order derivative method with relative standard deviation well below 2 indicating accuracy of the methods. Intraday and Inter-day precision studies were carried out by analysing tablet formulation, three times on the same day and on three different days, respectively. Relative standard deviation for intra-day and inter-day precision studies was satisfactorily well below 2.0 indicating high degree of precision and reproducibility of the methods. In forced degradation studies shows Acidic, Alkaline and Oxidation conditions of two drugs were degraded and not degraded under Photolytic condition.

Table 1: Results of Analysis of Tablets.

Tablet sample	Label claim (mg/tab)	% Label claim (n = 6)		%RSD	
		Simultaneous method	First order derivative method	Simultaneous method	First order derivative method
Lobeglitazone sulfate	0.5mg	99.685	99.95	1.0989	0.5711
Glimepiride	1mg	100.61	99.96	1.0366	0.7106

Table 2: Results of Recovery Studies (SIMULTANEOUS EQUATION METHOD).

Recovery level	Initial amount (μg/mL)		Concentration of std drug added (μg/mL)		%Recovery (n = 3) Simultaneous method	
	LOBE	GLIM	LOBE	GLIM	LOBE	GLIM
50%	4	8	2	4	99.76	99.25
100%	4	8	4	8	100.30	100.62
150%	4	8	6	12	99.86	99.91
			Mean		**99.97**	**99.93**

Table 3: Results of Recovery Studies (FIRST ORDER DERIVATIVE METHOD).

Recovery level	Initial amount (μg/mL)		Concentration of std drug added (μg/mL)		%Recovery (n = 3) First order derivative method	
	LOBE	GLIM	LOBE	GLIM	LOBE	GLIM
50%	6	12	3	6	99.93	100.33
100%	6	12	6	12	100.16	99.98
150%	6	12	12	18	99.87	100.05
			Mean		**99.98**	**100.12**

TABLE 4: FORCED DEGRADATION STUDY.

STRESS CONDITION	TIME (hr.)	COMMENT
0.1 M HCl	24	Degraded (60%)
0.1 M NaOH	24	Degraded (100%)
0.1% H_2O_2	24	Degraded (100%)
Photolysis	24	Not Degraded

5. CONCLUSION

The proposed UV spectrophotometric methods employed here proved to be simple, economical, rapid, precise and accurate. Thus these can be used for routine simultaneous estimation of Lobeglitazone sulfate and Glimepiride in tablet dosage form instead of processing and analysing each drug separately.

6. ACKNOWLEDGMENT

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REFERENCE

1. Sakala Bhargavi, Gopisetty Suryasagar, UV Spectrophotometric Method for Determination of Glimepiride in Pharmaceutical Dosage forms. Int. J. Pharm. Sci. Rev. Res; 21(2).
2. Dyade GK, Joshi HA, Estimation of metformin hydrochloride and Glimepiride in tablet formulations by UV- visible Spectrophotometry. Indo American Journal of Pharmaceutical Research, 2013.
3. Audumbar Digambar Mali, *ET. al.*, Simultaneous UV spectrophotometric methods for estimation of Metformin Hcl and Glimepiride in bulk and tablet dosage form. International Journal of advances in Pharmaceutics, 2015; 4(6).
4. https://go.drugbank.com/drugs/DB09198.
5. https://pubchem.ncbi.nlm.nih.gov/compound/Lobeglitazone.
6. https://go.drugbank.com/drugs/DB00222.
7. https://www.webmd.com/drugs/2/drug-12271/glimepiride-oral/details.
8. Madhusudhanareddy Induri, Bhagavan raju, Development and Validation of a Spectrophotometric method for Quantification and dissolution studies of Glimepiride in tablets, E-Journal of Chemistry, 2012; 9(2).