

Development and Validation of UV Spectrophotometric Methods for the Simultaneous Estimation of Lobeglitazone Sulfate and Glimepiride in Pharmaceutical Dosage Form.

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ABSTRACT: This research aimed to develop simple, specific, accurate, precise, reproducible, and robust spectrophotometric methods for the simultaneous estimation of Lobeglitazone Sulfate and Glimepiride and validate the developed methods. For First Order Derivative Method, the zero order spectra data was processed to obtain first order derivative spectrum in the range of 400-200 nm. In first order spectra Zero Crossing Point for LOB

239.5 nm and 282.6 nm for GLM. Wavelength selected for quantitation were 239.5 nm for GLM (ZCP of LOB) and 282.6 nm for LOB (ZCP of GLM). For Area Under Curve Method, area between the wavelengths 245-255 nm and 225-235 nm was selected for estimation of Lobeglitazone Sulfate and Glimepiride. All UV spectrophotometric method was found to be linear over the concentration range of 2-6 µg/ml and 4-12 µg/ml for Lobeglitazone Sulfate and Glimepiride respectively. All the methods were validated for linearity, precision, accuracy, LOD and LOQ according to ICH guideline.

KEYWORDS: Lobeglitazone Sulfate, Glimepiride, First Order Derivative Method, Area Under Curve Method,

I. INTRODUCTION

Diabetes mellitus is characterized by high levels of sugar (glucose) in the blood. After consuming a

meal, when glucose levels rise, the pancreas secretes insulin, prompting muscle and fat cells to absorb glucose from the blood while encouraging the liver to metabolize it, thereby restoring blood sugar to normal levels. However, individuals with diabetes experience persistently high blood sugar levels due to either inadequate insulin production, insufficient insulin secretion, or reduced insulin effectiveness.^[1,2]

Lobeglitazone Sulfate (LOB):

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. By activating PPAR-gamma and promoting the binding of insulin at fat cells, lobeglitazone thereby has been shown to reduce blood sugar levels, lower haemoglobin A1C (HbA1C) levels, and improve lipid and liver profiles.^[4,5]

Glimepiride (GLM):

Glimepiride blocks the ATP-sensitive potassium channel by binding non-specifically to the B sites of both sulfonylurea receptor-1 (SUR1) and sulfonylurea receptor-2A (SUR2A) subunits as well as the A site of SUR1 subunit of the channel to promote insulin secretion from the beta cell.^[6,7]

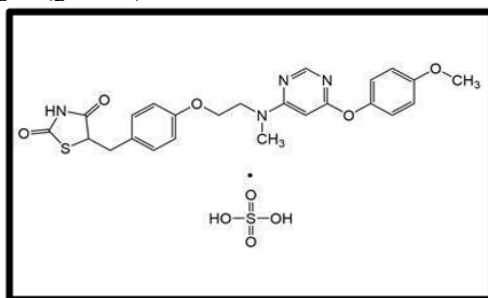


Fig No. 1 Chemical Structure of LOB

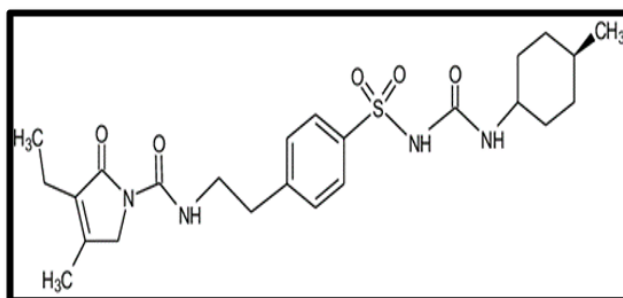


Fig No. 2 Chemical Structure of GLM

UV SPECTROSCOPIC METHOD

❖ FIRST ORDER DERIVATIVE METHOD APPARATUS AND INSTRUMENT

UV-Visible Spectrophotometer (Double Beam) (Shimadzu-1900i, Software- Lab- solution) having matched Quartz cells of light path 1 cm, Weighing balance, Ultra-Sonicator, Volumetric Flask – 10 ml, Pipettes- 1,5 ml.

REAGENTS AND MATERIALS

Lobeglitazone sulfate (Gift sample, Akums Pharmaceutical, Haridwar.) Glimepiride (Gift sample, Exemed Pharmaceutical, Vapi, Gujarat.)

SELECTION OF SOLVENT

The solubility analysis indicated that methanol was the common solvent for both drugs. Methanol was therefore used as the solvent for UV techniques. Drugs (Lobeglitazone sulfate and Glimepiride) provide linear spectra in methanol at their measured wavelength. Therefore, the recommended solvent is methanol.

PREPARATION OF STANDARD SOLUTION

1. Preparation of LOB standard stock solution (1000 µg/ml):

10 mg of LOB was weighed and transferred to 10 ml volumetric flask. It was dissolved in methanol and volume was made upto the mark with methanol to give a solution containing 1000 µg/ml.

2. Preparation of LOB working stock solution (100 µg/ml):

Aliquot of 1 ml from above standard stock solution was pipetted out into 10 ml of volumetric flask and volume was made upto the mark with methanol to give a solution containing 100 µg/ml.

3. Preparation of GLM standard stock solution (1000 µg/ml):

10 mg of GLM was weighed and transferred to 10 ml volumetric flask. It was dissolved in methanol and volume was made upto the mark with methanol to give a solution containing 1000 µg/ml.

4. Preparation of GLM working stock solution (100 µg/ml):

Aliquot of 1 ml from above standard stock solution was pipetted out into 10 ml of volumetric flask and volume was made upto the mark with methanol to give a solution containing 100 µg/ml.

FIRST ORDER DERIVATIVE SPECTROSCOPIC METHOD SELECTION OF WAVELENGTH

Standard solutions of LOB (2 µg/ml) and GLM (4 µg/ml) were prepared by diluting their respective working stocks with methanol. UV spectra were

recorded from 200–400 nm using methanol as blank and converted to first-order derivative spectra. The zero crossing points were identified at 239.5 nm for LOB and 282.6 nm for GLM. These wavelengths were selected for simultaneous quantification, measuring GLM at 239.5 nm (LOB ZCP) and LOB at 282.6 nm (GLM ZCP).

PREPARATION OF CALIBRATION CURVE

Calibration curve for LOB

A calibration curve for LOB was prepared using five concentrations (2–6 µg/ml) obtained by diluting a 100 µg/ml working stock solution with methanol. UV spectra were recorded against a methanol blank and converted to first-order derivative spectra. The derivative absorbance at 239.5 nm was measured for each concentration, and a calibration graph of $dA/d\lambda$ versus concentration was plotted to obtain the regression equation.

2. Calibration curve for GLM

A calibration curve for GLM was developed using five concentrations (4–12 µg/ml) prepared by diluting a 100 µg/ml working stock solution with methanol. UV spectra were recorded against a methanol blank and converted to first-order derivative spectra. The derivative absorbance at 282.6 nm was measured, and a calibration plot of $dA/d\lambda$ versus concentration was constructed to obtain the regression equation.

❖ VALIDATION OF PROPOSED METHOD

Parameters to be considered for the validation of method are:

1. Linearity (n=5)

Linearity was evaluated using five calibration levels over concentration ranges of 2–6 µg/ml for LOB and 4–12 µg/ml for GLM. Calibration plots of $dA/d\lambda$ absorbance versus concentration were constructed, and the regression equations along with correlation coefficients were determined for both drugs.

2. Precision

1. Repeatability (n=6)

Standard solutions containing 4 µg/ml of LOB and 8 µg/ml of GLM were prepared by diluting their respective working stock solutions with methanol. The solutions were analyzed six times ($n = 6$), and the percentage relative standard deviation (%RSD) was calculated to assess precision.

2. Intraday Precision (n=3)

Intra-day precision was evaluated by preparing mixed standard solutions of LOB (3, 4, and 5 µg/ml) and GLM (6, 8, and 10 µg/ml) from their working

stock solutions using methanol. Each concentration was analyzed three times on the same day ($n = 3$), and the %RSD was calculated.

3. Interday Precision ($n=3$)

Inter-day precision was assessed by preparing mixed standard solutions of LOB (3, 4, and 5 $\mu\text{g/ml}$) and GLM (6, 8, and 10 $\mu\text{g/ml}$) using methanol. Each solution was analyzed three times ($n = 3$) on three different days, and the %RSD was calculated.

3. Accuracy ($n=3$)

Accuracy of the method was assessed by standard addition method at different concentration levels i.e. 80%, 100% and 120%) and % recoveries were computed.

$$\% \text{ Recovery} = \frac{A - B}{C} \times 100$$

Where, A = Total amount of drug estimated

B = Amount of drug found on pre analysed basis

C = Amount of Pure drug added

Each solution was scanned from 200-400 nm against methanol as a blank. Absorbance of solution was measured at selected wavelengths for LOB and GLM. The amount of LOB and GLM was calculated at each level (80%, 100% and 120%) and % recoveries were computed.

4. LOD and LOQ

Detection limit and quantitation limit were determined based on the standard deviation of y-intercepts of calibration curves and average slope of calibration curves.

$$\text{LOD} = 3.3 \times \text{S.D./Slope}$$

$$\text{LOQ} = 10 \times \text{S.D./Slope}$$

5. SIMULTANEOUS ESTIMATION OF LOBEGLITAZONE SULFATE AND GLIMEPIRIDE IN MARKETED FORMULATION (TABLET) BY FIRST ORDER DERIVATIVE METHOD

The applicability of the method was evaluated by assay of tablet formulation ($n = 5$). Tablet powder

equivalent to 0.5 mg of LOB and 1 mg of GLM was extracted with methanol, sonicated, filtered, and diluted to obtain stock solutions of 50 $\mu\text{g/ml}$ LOB and 100 $\mu\text{g/ml}$ GLM. Further dilution produced a test solution containing 4 $\mu\text{g/ml}$ LOB and 8 $\mu\text{g/ml}$ GLM.

Absorbance was measured using first-order derivative spectra at 282.6 nm for LOB and 239.5 nm for GLM, and drug concentrations were calculated using the respective regression equations.

❖ AREA UNDER CURVE METHOD SELECTION OF WAVELENGTH

Appropriate dilutions were prepared for drugs from the working stock solution were scanned in the wavelength range of 200-400 nm. The absorption spectra of Area Under Curve (AUC) in absorption spectra of LOB were measured between the wavelength range 235-245 nm and for GLM were measured between the wavelength range 225-235 nm.

SIMULTANEOUS ESTIMATION OF LOBEGLITAZONE SULFATE AND GLIMEPIRIDE IN MARKETED FORMULATION (TABLET) BY AREA UNDER CURVE (AUC) METHOD

The applicability of the method was assessed by assay of tablets ($n = 5$). Tablet powder equivalent to 0.5 mg of LOB and 1 mg of GLM was extracted with methanol, sonicated, filtered, and diluted to obtain solutions of 50 $\mu\text{g/ml}$ LOB and 100 $\mu\text{g/ml}$ GLM. Further dilution produced a test solution containing 4 $\mu\text{g/ml}$ LOB and 8 $\mu\text{g/ml}$ GLM. The area under the curve (AUC) was measured at 245–255 nm and 225–235 nm against methanol. Concentrations of Lobe-glitazone Sulfate and Glimepiride were calculated using simultaneous equations based on mean absorptivity coefficients and solved by Cramer's rule.

II. RESULT AND DISCUSSION

FIRST ORDER DERIVATIVE METHOD

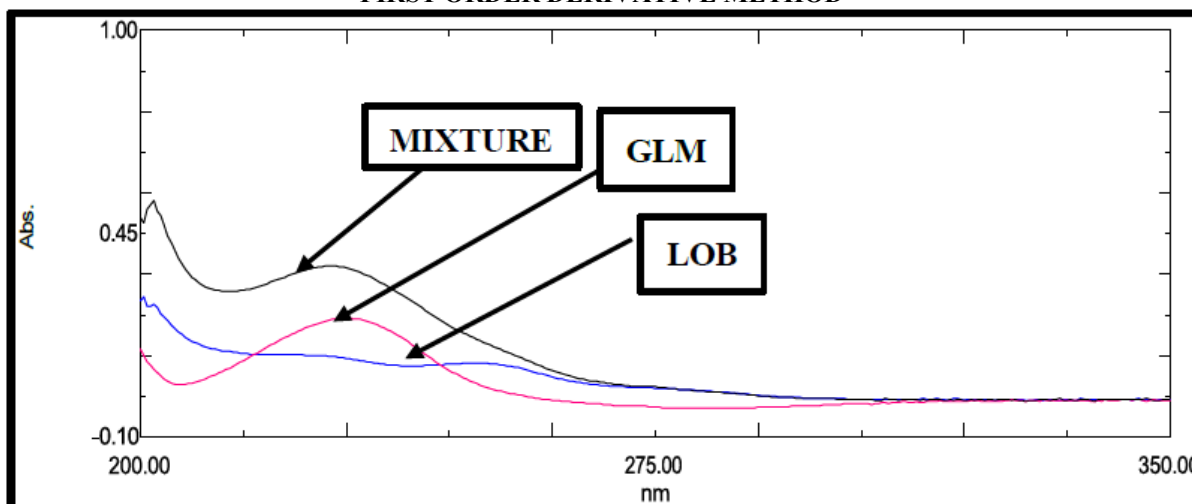


Fig.3. Overlain Spectra of LOB (2 µg/ml), GLM (4 µg/ml) and MIXTURE (2+4 µg/ml)

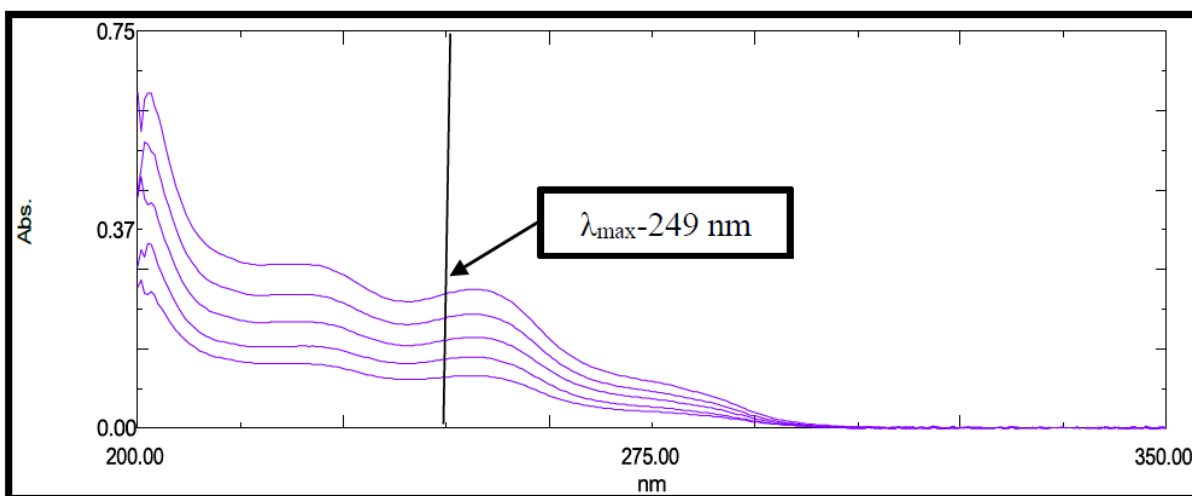


Fig.4. Zero Order Spectra of LOB (2-6 µg/ml).

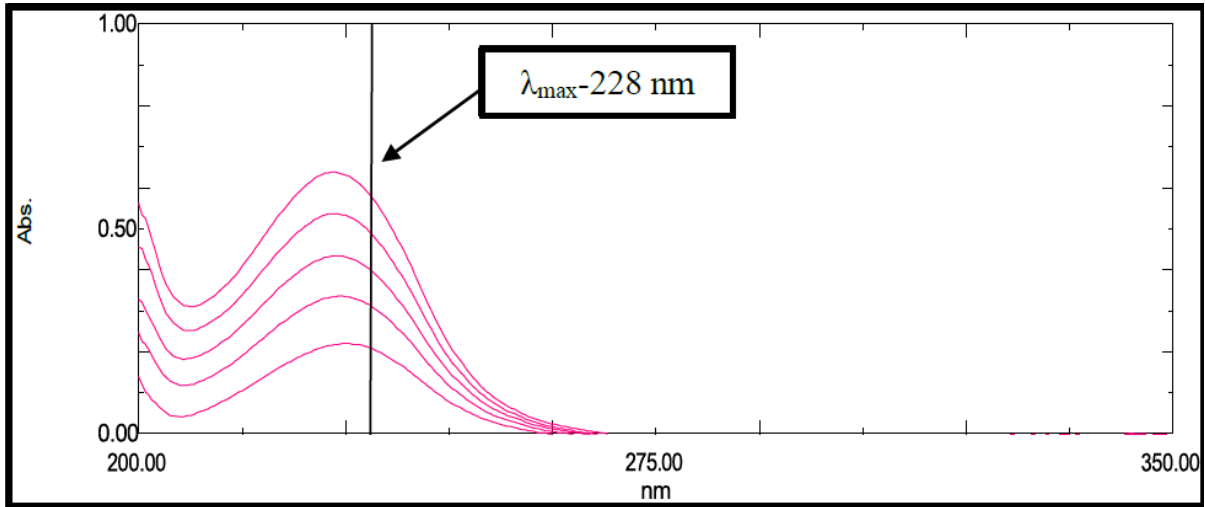


Fig.5. Zero Order Spectra of GLM (4-12 µg/ml)

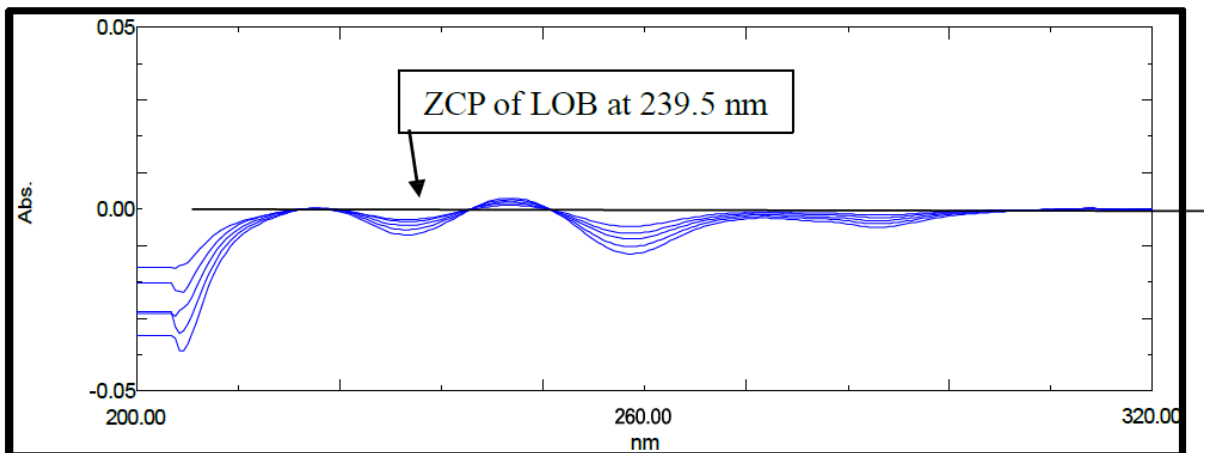


Fig.6. ZCP of LOB at 239.5 nm

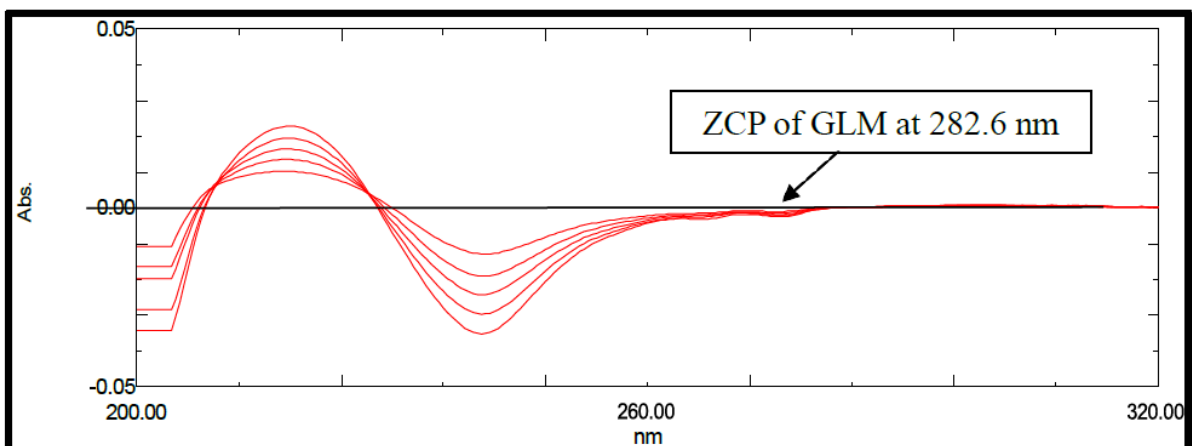


Fig.7. ZCP of GLM at 282.6 nm

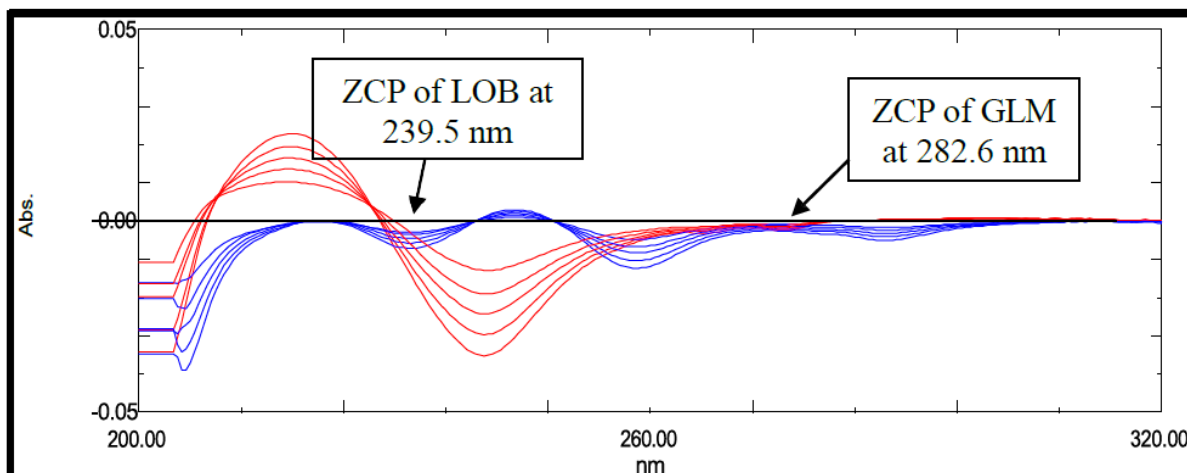


Fig.8. Overlain Spectra of LOB (2-6 µg/ml) and GLM (4-12) µg/ml

Validation of proposed UV method

1. Linearity:

The linearity range for LOB and GLM was found to be in the range of 2-6 µg/ml and 4-12 µg/ml respectively. Linearity data for LOB at 282.6 nm and GLM at 239.5 nm.

Table no. 1 Linearity of LOB at 282.6 nm (ZCP of GLM)

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=5)	%R.S.D
1.	2	0.0013 ± 0.000010	0.7629
2.	3	0.0018 ± 0.000013	0.7222
3.	4	0.0023 ± 0.000015	0.6521
4.	5	0.0029 ± 0.000015	0.5172
5.	6	0.0034 ± 0.000016	0.4705

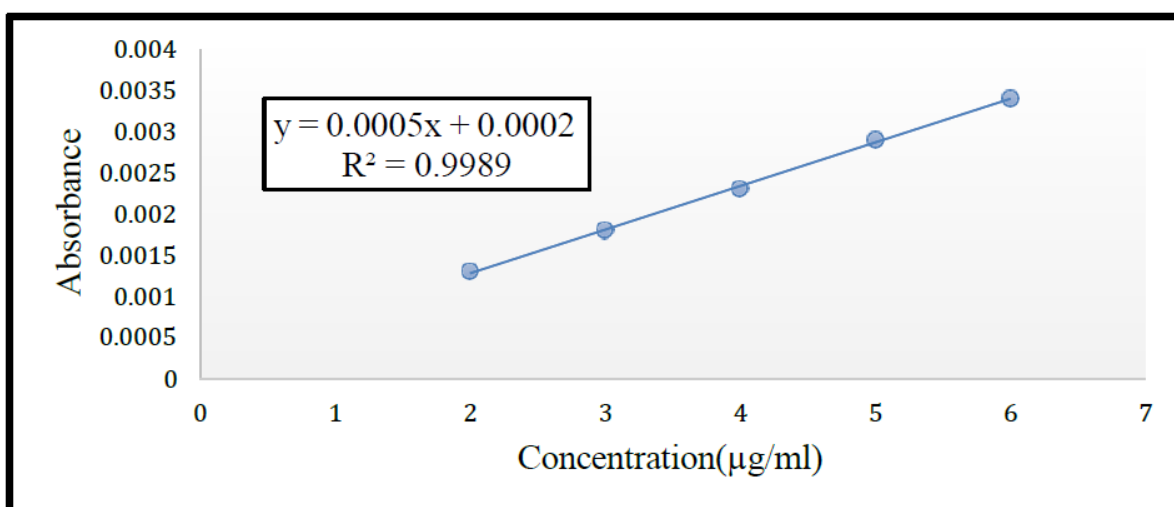


Fig.9. Calibration Curve of LOB at 282.6 nm (ZCP of GLM)

Table no.2 Linearity and Calibration Curve of GLM at 239.5 nm (ZCP of LOB)

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=5)	%R.S.D
1.	4	0.0127 ± 0.000092	0.7244
2.	6	0.0187 ± 0.000118	0.6310
3.	8	0.0239 ± 0.000129	0.5397
4.	10	0.0292 ± 0.000130	0.4452
5.	12	0.0347 ± 0.000124	0.3573

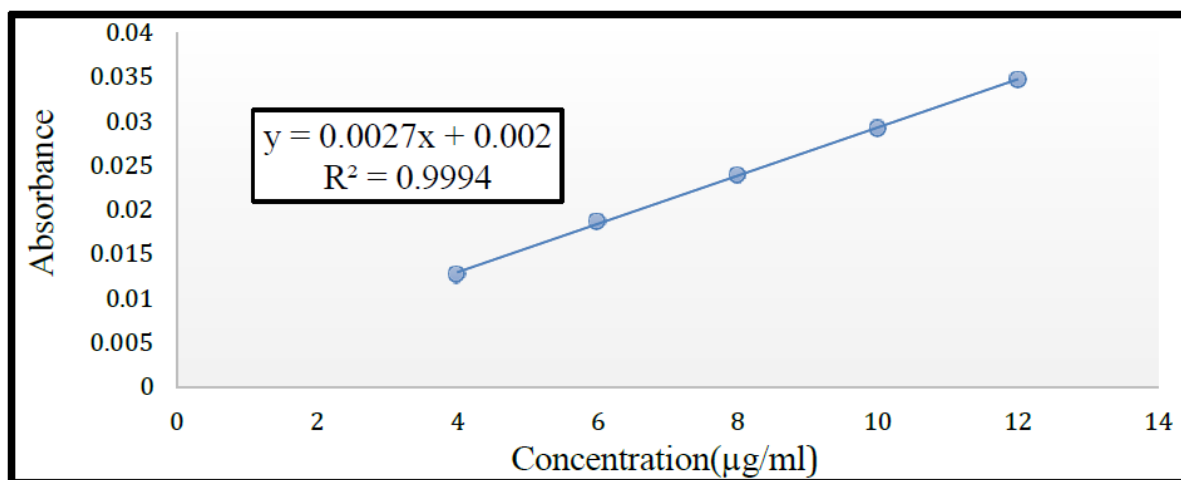


Fig. 10. Calibration Curve of GLM at 239.5 nm (ZCP of LOB)

Table no. 3. Regression line equation, Regression coefficient and correlation coefficient for LOB and GLM

Sr.No.	Drugs	Regression line Equation	Regression coefficient (R ²)	Correlation coefficient (r)
1.	LOB	y = 0.0005x + 0.0002	0.9989	0.9994
2.	GLM	y = 0.0027x + 0.002	0.9994	0.9996

2. PRECISION

A) Repeatability (n=6):

The data of Repeatability for LOB at 282.6 nm (ZCP of GLM) and GLM at 239.5 nm (ZCP of LOB) is shown in table.

Table no. 4. Repeatability data for LOB and GLM

Sr.No.	Drugs	Concentration (µg/ml)	Mean Abs. ± S.D. (n=6)	%R.S.D
1.	LOB	4	0.0024 ± 0.000016	0.6666
2.	GLM	8	0.0240 ± 0.000132	0.5500

B) Intraday Precision (n=3):

The data of Intraday Precision for LOB at 282.6 nm (ZCP of GLM) and GLM at 239.5nm (ZCP of LOB) is shown in table.

Table no. 5. Intraday Precision for LOB at 282.6 nm

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=3)	%R.S.D
1.	3	0.0021 ± 0.000016	0.7619
2.	4	0.0025 ± 0.000017	0.6800
3.	5	0.0032 ± 0.000018	0.5625

Table no. 6. Intraday Precision for GLM at 239.5 nm

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=3)	%R.S.D
1.	6	0.0189 ± 0.000125	0.6613
2.	8	0.0242 ± 0.000139	0.5743
3.	10	0.0295 ± 0.000141	0.4779

C) Interday Precision (n=3):

The data of Interday Precision for LOB at 282.6 nm (ZCP of GLM) and GLM at 239.5nm (ZCP of LOB) is shown in table.

Table no. 7. Interday Precision for LOB at 282.6 nm

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=3)	%R.S.D
1.	3	0.0031 ± 0.000026	0.8371
2.	4	0.0036 ± 0.000027	0.7500
3.	5	0.0043 ± 0.000027	0.6279

Table no. 8. Interday Precision for GLM at 239.5 nm

Sr.No.	Concentration (µg/ml)	Mean Abs. ± S.D. (n=3)	%R.S.D
1.	6	0.0198 ± 0.000143	0.7222
2.	8	0.0253 ± 0.000160	0.6324
3.	10	0.0304 ± 0.000167	0.5493

3. Accuracy:

Accuracy of the proposed method was assured by performing recovery study from marketed formulation at three levels by standard addition method. Percentage recovery for LOB and GLM at 282.6 nm and 239.5 nm were obtained respectively. The result is depicted in table. Recovery was found to be in the limit of 98-102%.

Table No.9. Determination of accuracy of LOB and GLM

Drugs	Level	Amount of sample (µg/ml)	Amount of std. spiked (µg/ml)	Total amount (µg/ml)	Amount of sample found (µg/ml)	% Recovery
LOB	0%	2	0	2	1.985	99.25
	80%	2	1.6	3.6	3.576	99.33
	100%	2	2	4	3.977	99.42
	120%	2	2.4	4.4	4.375	99.56
GLM	0%	4	0	4	3.969	99.22
	80%	4	3.2	7.2	7.165	99.51
	100%	4	4	8	7.965	99.56
	120%	4	4.8	8.8	8.812	100.13

4. Analysis of Marketed Formulation:

Analysing the marketed formulation was used in the method's suitability. The outcomes are shown in table.

Table no. 10. Determination of Assay of LOB and GLM

Tablet (LOBG-G1)	Actual conc. (mg/tablet)		Amt. obtained mean \pm S.D. (n=5) (mg/tablet)		LOB % Purity \pm S.D. (n=5)	GLM % Purity \pm S.D. (n=5)
	LOB	GLM	LOB	GLM		
	4	8	3.978 \pm 0.0266	7.966 \pm 0.0383	99.45 \pm 0.6671	99.57 \pm 0.4789

❖ AREA UNDER CURVE METHOD:

Selection of wavelength for estimation of Lobeglitazone Sulfate and Glimepiride:

The standard solution of Lobeglitazone Sulfate and Glimepiride were diluted with methanol individually to get the concentration of 2 $\mu\text{g/ml}$ and 4 $\mu\text{g/ml}$ of respectively of Lobeglitazone Sulfate and Glimepiride. The drugs were scanned in the UV range 200-400 nm. The areas were selected for Lobeglitazone Sulfate 245-255 nm and for Glimepiride 225-235 nm. As linearity of drugs are good in this selected wavelength range.

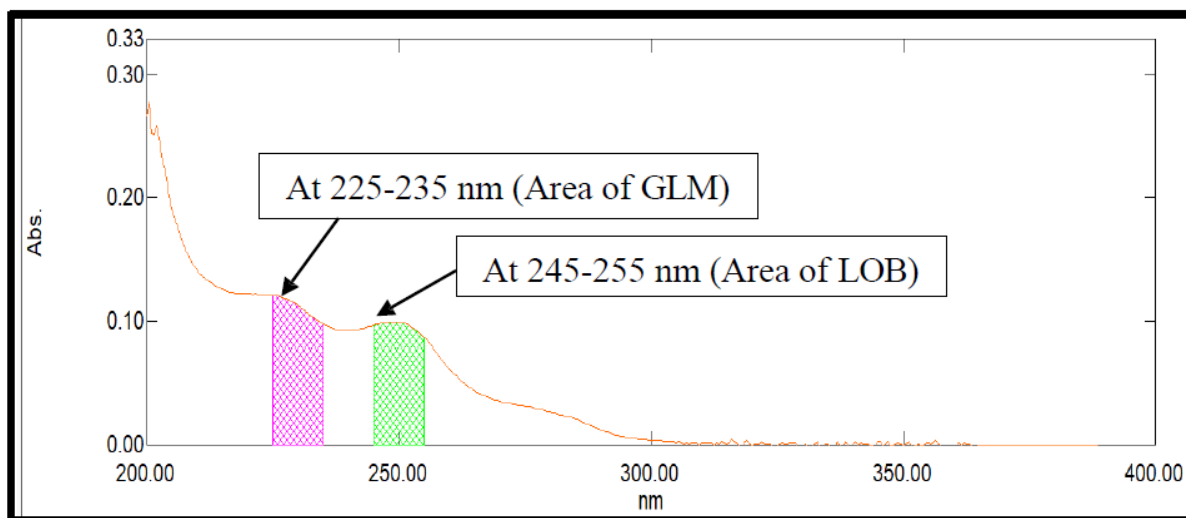


Fig. 11. AUC Spectra of Lobeglitazone Sulfate (LOB) 2 $\mu\text{g/ml}$ in wavelength range 225-235 nm and 245-255 nm

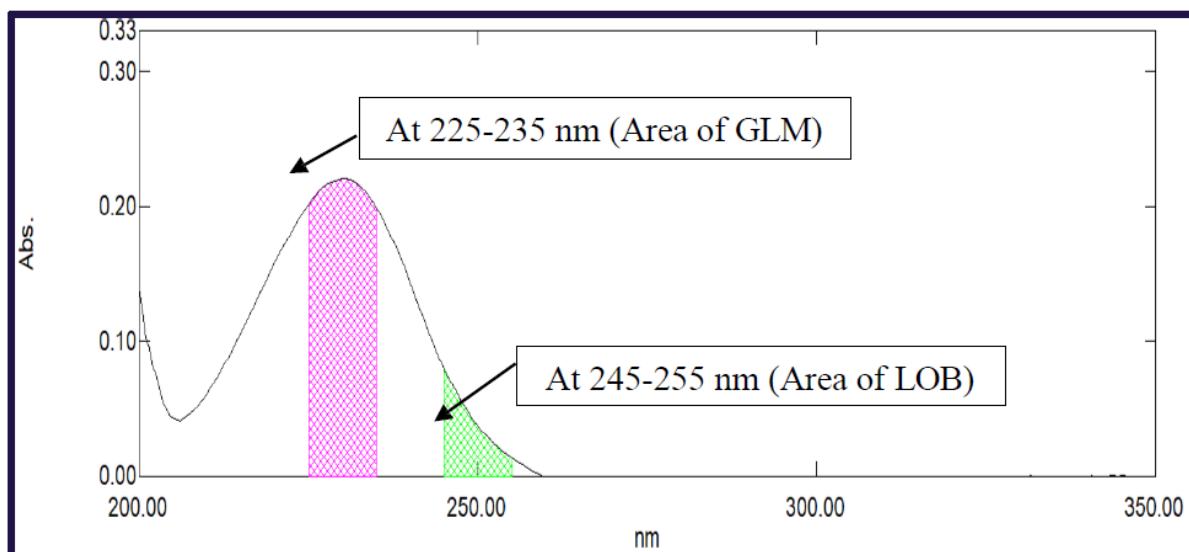


Fig. 12. AUC Spectra of Glimepiride (GLM) 4 µg/ml in wavelength range 225-235 nm and 245-255 nm

❖ Validation of proposed UV method

1. Linearity:

The linearity range for LOB and GLM was found to be in range of 2-6 µg/ml and 4- 12 µg/ml respectively. Linearity data for LOB at 245-255 nm and GLM at 225-235nm.

Table no. 11. Linearity of LOB at 245-255 nm (Area of GLM)

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=5)	%R.S.D
1.	2	1.1121 ± 0.00950	0.8545
2.	3	1.4731 ± 0.01121	0.7611
3.	4	1.8992 ± 0.01296	0.6825
4.	5	2.3523 ± 0.01222	0.5197
5.	6	2.8542 ± 0.01326	0.4647

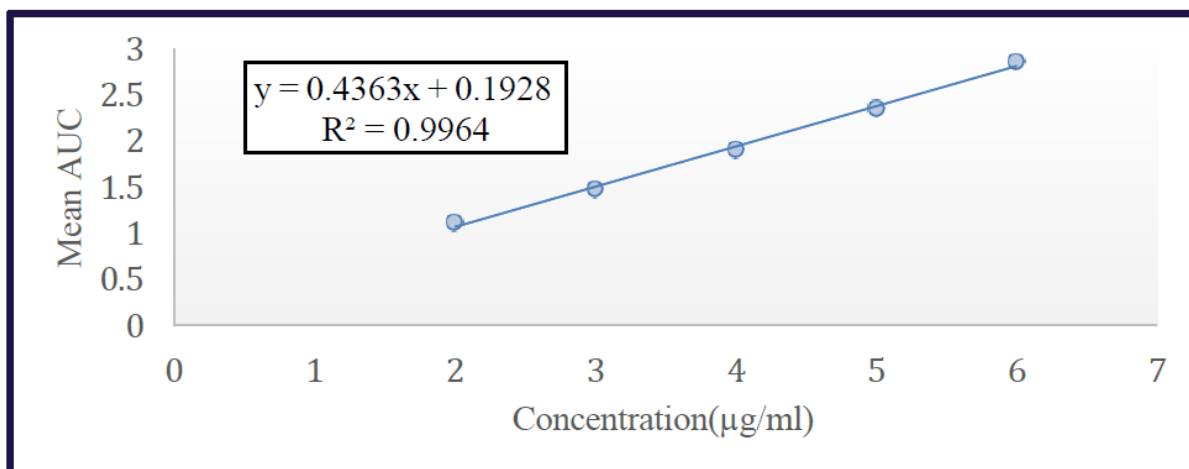


Fig. 13. Calibration Curve of LOB at 245-255 nm (Area of GLM)

Table no. 12. Linearity of LOB at 245-255 nm (Area of LOB)

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=5)	%R.S.D
1.	2	0.9623 ± 0.00811	0.8427
2.	3	1.3012 ± 0.01001	0.7692
3.	4	1.6681 ± 0.01153	0.6912
4.	5	2.0921 ± 0.01154	0.5515
5.	6	2.5482 ± 0.01110	0.4356

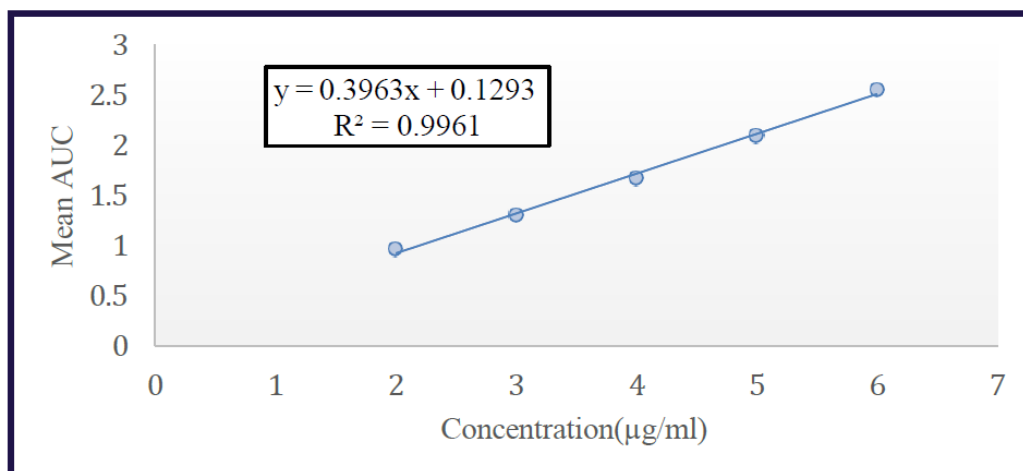


Fig. 14. Calibration Curve of LOB at 245-255 nm (Area of LOB)

Table no. 13. Linearity and Calibration Curve of GLM at 225-235 nm (Area of GLM)

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=5)	%R.S.D
1.	4	2.1362 ± 0.01549	0.7251
2.	6	3.2571 ± 0.02102	0.6453
3.	8	4.2041 ± 0.02316	0.5508
4.	10	5.1883 ± 0.02429	0.4681
5.	12	6.1642 ± 0.02162	0.3507

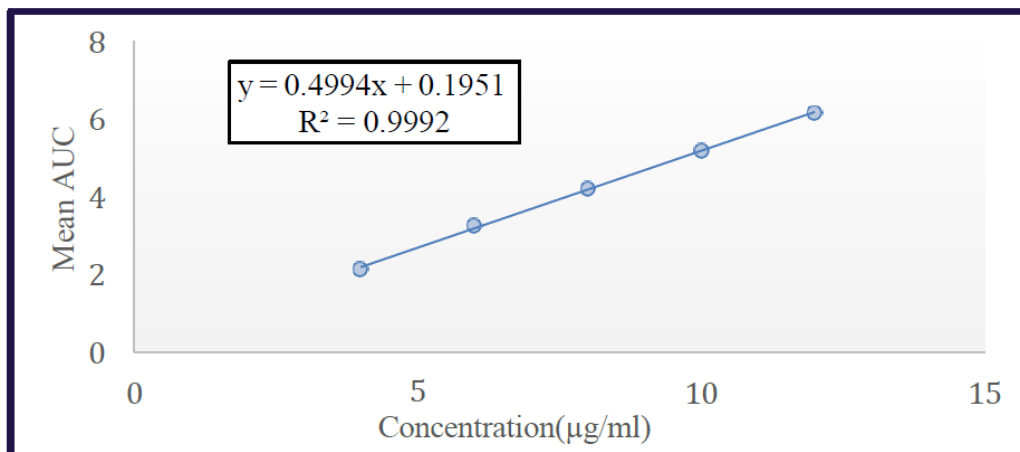


Fig. 15. Calibration Curve of GLM at 225-235 nm (Area of GLM)

Table no. 14. Linearity and Calibration Curve of GLM at 225-235 nm (Area of LOB)

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=5)	%R.S.D
1.	4	0.4042 ± 0.00284	0.7026
2.	6	0.6311 ± 0.00417	0.6607
3.	8	0.8323 ± 0.00487	0.5851
4.	10	1.0121 ± 0.00489	0.4831
5.	12	1.2042 ± 0.00459	0.3811

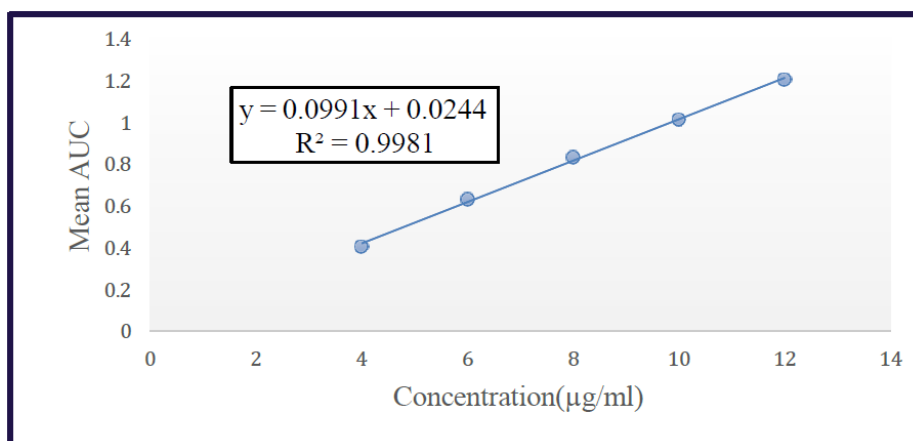


Fig. 16. Calibration Curve of GLM at 225-235 nm (Area of LOB)

Table no. 15. Regression line equation, Regression coefficient and correlation coefficient for LOB and GLM

Sr.No	Drugs	Regression line Equation	Regression coefficient (R ²)	Correlation coefficient (r)
1.	LOB	y = 0.4363x + 0.1928	0.9964	0.9981
		y = 0.3963x + 0.1293	0.9961	0.9980
2.	GLM	y = 0.4994x + 0.1951	0.9992	0.9995
		y = 0.0991x + 0.0244	0.9981	0.9990

2. PRECISION

A) Repeatability (n=6):

The data of Repeatability for LOB at 245-255 nm (Area of LOB) and GLM at 225-235nm (Area of LOB) is shown in table.

Table no. 16. Repeatability data for LOB and GLM

Sr.No.	Drugs	Concentration (µg/ml)	Mean AUC ± S.D. (n=6)	%R.S.D
1.	LOB	4	1.6684 ± 0.01188	0.7120
2.	GLM	8	4.2043 ± 0.02401	0.5710

B) Intraday Precision (n=3):

The data of Intraday Precision for LOB at 245-255 nm (Area of LOB) and GLM at 225-235 nm (Area of GLM) is shown in table.

Table no. 17. Intraday Precision for LOB at 245-255 nm

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=3)	%R.S.D
1.	3	1.3015 ± 0.01030	0.7913
2.	4	1.6685 ± 0.01233	0.7389
3.	5	2.0924 ± 0.01241	0.5930

Table no. 18. Intraday Precision for GLM at 225-235 nm

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=3)	%R.S.D
1.	6	3.2573 ± 0.02291	0.7033
2.	8	4.2044 ± 0.02773	0.6595
3.	10	5.1889 ± 0.02555	0.4923

C) Interday Precision (n=3):

The data of Interday Precision for LOB at 245-255 nm (Area of LOB) and GLM at 225-235 nm (Area of GLM) is shown in table.

Table no. 19. Interday Precision for LOB at 245-255 nm

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=3)	%R.S.D
1.	3	1.3024 ± 0.01161	0.8914
2.	4	1.6701 ± 0.01325	0.7933
3.	5	2.1935 ± 0.01478	0.6738

Table no. 20. Interday Precision for GLM at 225-235 nm

Sr.No.	Concentration (µg/ml)	Mean AUC ± S.D. (n=3)	%R.S.D
1.	6	3.2584 ± 0.02727	0.8369
2.	8	4.2053 ± 0.03226	0.7671
3.	10	5.1897 ± 0.03069	0.5913

3. Accuracy:

Accuracy of the proposed method was assured by performing recovery study from marketed formulation at three levels by standard addition method. Percentage recovery for LOB and GLM at 245-255 nm and 225-235 nm were obtained respectively. The result is depicted in table. Recovery was found to be in the limit of 98-102%.

Table no. 21. Determination of accuracy of LOB and GLM

Drugs	Level	Amount of sample (µg/ml)	Amount of std. spiked (µg/ml)	Total amount (µg/ml)	Amount of sample found (µg/ml)	% Recovery
LOB	0%	2	0	2	1.981	99.05
	80%	2	1.6	3.6	3.574	99.27
	100%	2	2	4	3.986	99.65
	120%	2	2.4	4.4	4.409	100.20
GLM	0%	4	0	4	3.972	99.30
	80%	4	3.2	7.2	7.162	99.47
	100%	4	4	8	7.966	99.57
	120%	4	4.8	8.8	8.778	99.75

4. Analysis of Marketed Formulation:

Suitability of the method was tested by analysing the marketed formulation. The data of assay for tablet using Cramer's Rule at wavelength 245-255 nm and 225-235 nm. The outcomes are show in table.

Table no. 22. Determination of Assay of LOB and GLM

Tablet (LOBG G1)	Actual conc. (mg/tablet)		Amt. obtained mean \pm S.D. (n=5) (mg/tablet)		LOB % Purity \pm S.D. (n=5)	GLM % Purity \pm S.D. (n=5)
	LOB	GLM	LOB	GLM		
	4	8	3.989 \pm 0.0208	7.968 \pm 0.0329	99.72 \pm 0.5216	99.60 \pm 0.4199

III. CONCLUSION:

The proposed UV Spectroscopic Methods were developed for simultaneous estimation of Lobeglitazone sulfate (LOB) and Glimepiride (GLM) and in pharmaceutical dosage form are simple, precise, accurate, and sensitive. They can be used for the routine analysis of both drugs in pharmaceutical formulations. The method was developed and validated as per ICH guidelines. The precision of the developed methods was confirmed by intra-day and inter-day analysis and the accuracy was confirmed by the recovery study. The %RSD was found to be <2.0%. It indicates that the method has good precision and accuracy.

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