WORLD JOURNAL OF PHARMACEUTICAL RESEARCH
SJIF Impact Factor 8.453
Volume 14, Issue 2, 332-344. Review Article ISSN 2277–7105

ANALYTICAL METHOD DEVELOPEMENT AND VALIDATION FOR ESTIMATION OF LOBEGLITAZONE SULFATE IN PHARMACEUTICAL DOSAGE FORMS BY UV SPECTROPHOTOMETRIC METHOD

C. A. Shaik Fayaaz Ahamed[1]*, M. Sathish[2], B. Sarumathy[3], M. Seenuvasan[4], E. Shuruthiga[5] and E. S. Sharma[6]

[1]Associate Professor Department of Pharmaceutical Analysis,

[2,3,4,5,6]B. Pharm Student,

Aadhibhagawan College of Pharmacy, Rantham, Thiruvannamalai, Tamil Nadu.

Article Received on
25 November 2024,

Revised on 15 Dec. 2024,
Published on 15 Jan. 2025

DOI: 10.20959/wjpr20252-35233

***Corresponding Author**
C. A. Shaik Fayaaz Ahamed
Associate Professor
Department of Pharmaceutical Analysis, Aadhibhagawan College of Pharmacy, Rantham, Thiruvannamalai, Tamil Nadu.

ABSTRACT

The developed UV-Visible spectroscopy method for analyzing lobeglitazone sulfate tablet formulation is simple, rapid, accurate, precise, and sensitive. The method has been validated, with n-butanol used as the solvent. All validation parameters, including linearity, precision, accuracy, and robustness, showed less than 2% RSD, in accordance with ICH guidelines, indicating the method's sensitivity. The correlation coefficient was 0.998. Both intra-day and inter-day %RSD values were within the acceptable range, and accuracy remained within normal limits. The method's limit of detection (LOD) and limit of quantification (LOQ) were determined to be 0.231 µg/mL and 1.015 µg/mL, respectively, demonstrating its suitability for the rapid quantification of lobeglitazone sulfate. In conclusion, the proposed method is accurate, precise, reliable, and compliant with ICH guidelines, making it ideal for routine analysis of lobeglitazone sulfate in bulk formulations.

KEYWORDS: Lobeglitazone Sulfate, UV-Visible Spectroscopy, Validation.

1. INTRODUCTION

Ultraviolet (UV) visible spectroscopy is a widely used analytical technique for the qualitative and quantitative analysis of various compounds, particularly in the pharmaceutical industry. The method is based on the absorption of UV or visible light by a sample, which leads to electronic transitions in the molecules. These transitions provide valuable information about the chemical structure and concentration of the analyte. UV-visible spectroscopy is favored for its simplicity, rapidity, and non-destructive nature, making it an essential tool for routine analysis in quality control and formulation development.

The development of robust analytical methods using UV spectroscopy is critical to ensure the accuracy, precision, and sensitivity required for pharmaceutical testing. For the effective application of UV spectroscopy in drug analysis, the method must be validated according to guidelines such as those provided by the International Council for Harmonisation (ICH). This includes evaluating parameters such as linearity, precision, accuracy, robustness, and the limits of detection (LOD) and quantification (LOQ).

In the case of lobeglitazone sulfate, a drug used in the management of metabolic disorders, the development of a reliable and efficient UV spectroscopy method is essential for its routine analysis in tablet formulations. This method must meet the required standards for quality assurance, ensuring that the drug formulation is both consistent and effective for patient use. The aim of this study is to develop and validate a UV-visible spectroscopy method for the quantification of lobeglitazone sulfate, focusing on its precision, sensitivity, and compliance with regulatory guidelines.

Validation is a critical process in the development of analytical methods, ensuring that the method is suitable for its intended purpose and consistently produces reliable, accurate, and precise results. In the pharmaceutical and chemical industries, method validation is essential to confirm that an analytical technique performs as expected under the specified conditions. This process is particularly important for regulatory compliance, ensuring that all products meet safety, efficacy, and quality standards.

The validation of an analytical method involves a comprehensive evaluation of key performance characteristics, including specificity, linearity, accuracy, precision, robustness, limit of detection (LOD), limit of quantification (LOQ), and system suitability. Each of these parameters provides essential information about the reliability and effectiveness of the

method in various conditions. For instance, precision evaluates the consistency of results over time, while accuracy assesses the closeness of measured values to the true value. Robustness ensures the method's stability under slight variations in experimental conditions, and LOD/LOQ help determine the method's sensitivity.

In compliance with regulatory guidelines such as those outlined by the International Council for Harmonisation (ICH), method validation is crucial for ensuring that analytical techniques can be confidently applied in routine testing and quality control. For pharmaceutical formulations, this means that validated methods can be reliably used to assess the identity, purity, and concentration of active pharmaceutical ingredients (APIs), ensuring that each product batch is safe and effective for patient use.

This study aims to validate a UV-visible spectroscopic method for the analysis of lobeglitazone sulfate tablet formulations. By evaluating the method's performance across various validation parameters, we ensure that it meets the rigorous standards required for accurate and reproducible quantification of the drug in bulk and tablet forms.

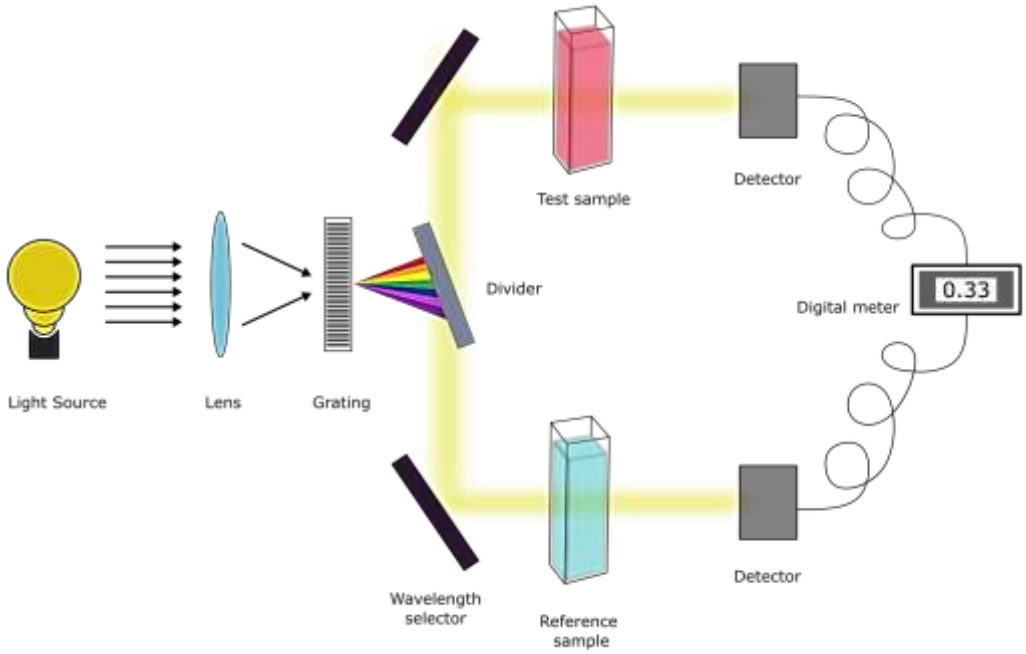

Fig. 1: Double beam spectrophotometer.

2. Drug profile

- **Drug name:** Lobeglitazone Sulfate (TZDs)
- **Chemical name:** 5-(4-(2-((6- (4-methoxy phenoxy) pyrimidin- 4-yl) (methyl) amino) ethoxy) benzyl) thiazolidine -2,4-dione sulfate.

- **Molecular formula:** $C_{24}H_{26}N_4O_9S_2$
- **Molecular weight:** 578.6g
- **Strength:** 0.5 mg
- **Description:** Lobeglitazone sulfate is an anti diabetic medication from the thiazolinedione class of drug. It functions as an insulin sensitizer by binding and activating and peroxisome proliferator –activated receptors (PPAR) in fat cells. Lobeglitazone sulfate is a oral administration, agonist for both PPAR α and PPAR γ receptors in fat cells and making the cell more responsive to insulin.
- **Appearance:** White to-off white solid powder.
- **Solubility:** Completely soluble in dimethyl sulfoxide (DMSO) and other organic solvents such as ethanol and n-butanol, decreased soluble in water due to sulfate form is present.
- **Selection of solvent:** n-butanol is used as a solvent
- **Melting point:** 165-167^0 c
- **Boiling point:** 711.0±60.0◦c (Predicted)
- **Bioavailability:** Oral- absolute bioavailability ~ 95%, IV - absolute bioavailability 100%
- **Elimination Half-Life:** 7.8 – 9.8 hrs
- **Brand name:** LOBG (Glenmark pharmaceuticals)
- **Mechanism of action:** Lobeglitazone act as an insulin sensitizer by binding and activating peroxisome proliferator- activated receptor (PPAR) gamma within fat cells. By promoting the binding of insulin at fat cells. Lobeglitazone has been shown to reduce blood sugar level, lower haemoglobin A1C (HbA1C) levels and improve lipid and liver profiles. Unlike Pioglitazone, which is a dual agonist at PPAR-alpha and PPAR-gamma, Lobeglitazone is a pure PPAR- alpha agonist.

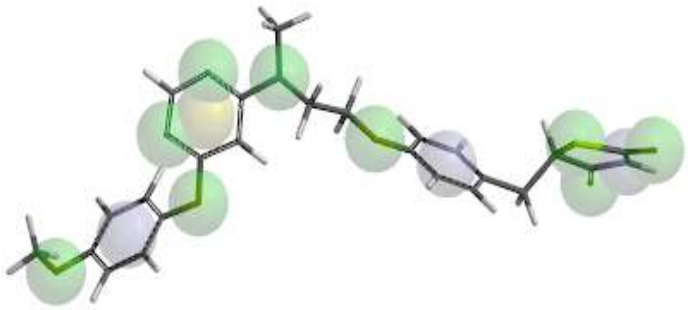

Fig. 2: Structure of lobeglitazone sulfate.

3. MATERIALS AND METHODS

3.1 Instrumentation

For the selection of analytical wavelength PC based Double Beam Labindia UV- visible spectrophotometer with 1.0cm matching quartz cell were used for measurement of absorbance. The UV spectra were recorded over the wavelength **200-400nm**.

All the drug and chemical were weighed on digital laboratory electronic balanced.

3.2 Procedure

3.2.1 Preparation of standard stock solution

A precisely measured amount of 50 mg of LBG was separately transferred into 50 ml volumetric flasks. 10 ml n-butanol was added to the flask, shake it to dissolve the drugs, and then diluted with n-butanol to the mark, resulting in a stock solution with a concentration of 1000 µg./ml.

3.2.2 Preparation of working standard stock solution

A 10ml of standard stock solution was withdrawn and transferred to 100ml volumetric flask. Volume is made up to diluents to get the working standard solution 100 g/ml of lobeglitazone sulfate.

3.2.3 Procedure for determination of wavelength for measurement (λ max)

For the selection of analytical wavelength range for method 100µg/ml lobeglitazone sulfate was scanned in the spectrum mode from 200-400nm. n-butanol was used as blank. It was seen at the **252nm** maximum absorbance.

3.2.4 Preparation of calibration curve of lobeglitazone sulfate

From the standard stock solution, the various dilutions with 2µg/ml, 3µg/ml, 4µg/ml, 5µg/ml, 6µg/ml were prepared. The solutions were scanned at **252nm**, and absorbance was recorded. Lobeglitazone sulfate was plotted the graph and linearity represented the correlation coefficient was found to be 0.998.

3.2.5 Parameters of analytical method validation

Analytical method validation: World journal of pharmaceutical research to ICH guidelines, the developed method was validate to assure the reliability of results of the analysis for different parameters like linearity, precision, accuracy, robustness, ruggedness, limit of detection (LOD), limit of quantification (LOQ), specificity.

3.3 Linearity

The linearity was determined by analysing absorbance of the lobeglitazone sulfate standard concentration (2-6µg/ml) **252nm.** The absorbance are measured. A regression equation and correlation coefficient were determined for lobeglitazone sulfate standard concentration. The results of linearity is shown in table 1. The correlation coefficient was found to be 0.998.

3.4 Accuracy

Accuracy was preparing by 3 sample of the solution 80,100 and 120% of working standard and added concentration of lobeglitazone sulfate in each sample solution and dissolved in 10ml of volumetric flask. Accuracy was assessed using a minimum of 9 determinations over a minimum 3 concentrations levels for each sample. The results of accuracy are shown in the table.2

3.5 Repeatability

Repeatability was expresses the closeness of the results obtained with the same sample (or) subsample of the same sample, same location over a short period of time. Repeatability was expected to give the smallest possible variation in result. select the middle concentration i.e. 10µg/ml and carry out the repeatability by taking the absorbance of the solution six times and the %RSD was found to be 1.05% with RSD≤2. The results of repeatability are as shown in table 3.

3.6 Precision

The precision of this method was estimated by two method variation. These two method variation are Interday and Intraday. Thus the above method at different time interval (morning, afternoon, evening) on the same day (Intraday precision) and on three consecutive day (Interday precision). Interday precision was determined by checking absorbance is 10$\mu g/ml$. on three different days. Calculate the mean of absorbance and %RSD. The results of precision are shown in the table.4&5

3.7 Robustness

Robustness was carryout by doing variation in method of parameter was done (i.e change in wavelength). Robustness of lobeglitazone sulfate was determined by variance if wavelength analysis by person to person. No significant difference was found in the absorbance and hence the proposed method was considered as robust which is shown in table.6

3.8 Limit Of Detection (LOD) And Limit Of Quantification (LOQ)

Limit of detection is taken in the lowest amount of sample can be detected. The limit of quantification is taken in the lowest amount of analyte in the sample under the experimental conditions. The LOD and LOQ for lobeglitazone sulfate were found to be **0.231µg/ml** and **1.015µg/ml.**

3.9 Specificity

A solution containing mixture of tablet excipients were using the sample preparation procedure to evaluate the possible interference of the excipients. From the absorbance result no interference was observed from the excipients present in the formulation indicated that the method is specific.

3.10 Assay of lobeglitazone tablet for formulation

For the analysis 10 tablet for lobeglitazone sulfate were weighed and finely powdered an accurately weighed quantity of powder is equivalent to 112.2mg of lobeglitzone sulfate was taken in 100ml volumetric flask. n-butanol is used as a solvent. The solution was sonicated for 15mins and then filtered through by whatmann filter paper (No:41) and volume adjusted the solvent. Pipette out 10ml of the solution and makeup 100ml of solvent. From this further dilution was made to get the final concentration of 100$\mu g/ml$. The results of assay was shown in table 7.

4. RESULTS AND DISCUSSION

4.1 Solubility

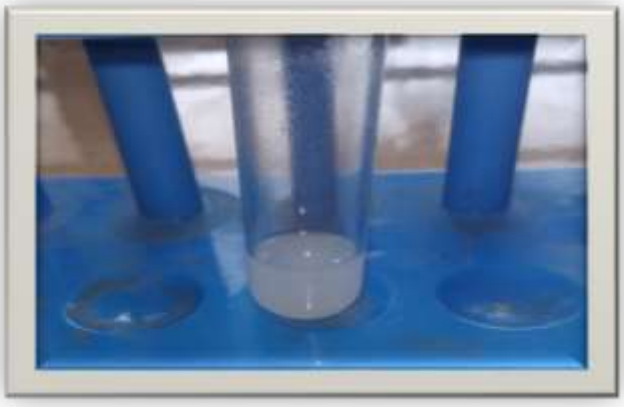

Fig. 3: Solubility profile.

Result: Soluble In n-Butanol

4.2 Determination of wavelength for measurement (λ MAX)

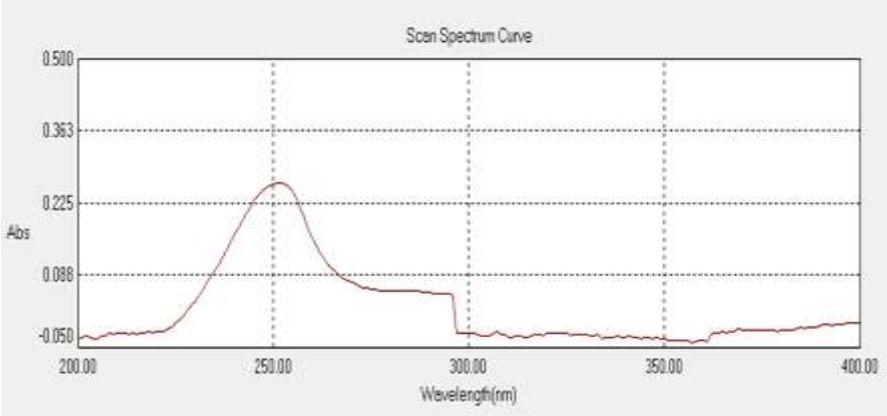

Fig. 4: Determination of wavelength for measurement (λ Max).

4.3 Linearity

Table 1: Linearity.

S. No.	Concentration ($\mu g/ml$)	Absorbance
1	2	0.234
2	3	0.332
3	4	0.445
4	5	0.552
5	6	0.661

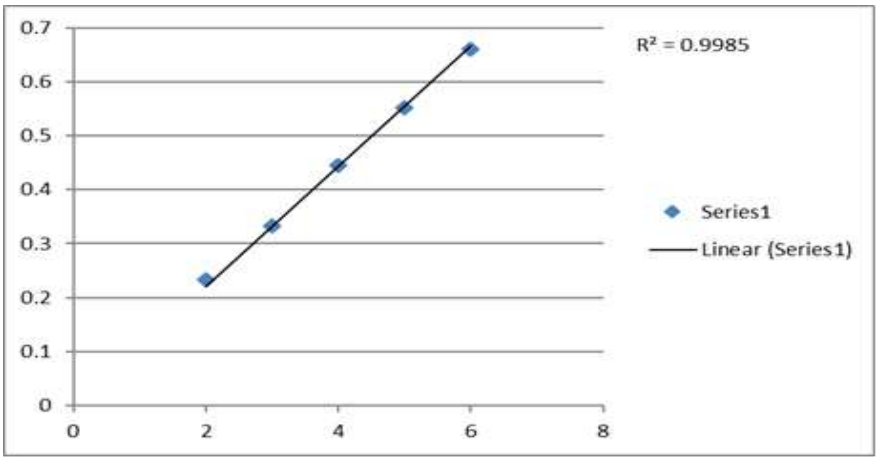

Fig. 5: Linearity.

4.4 Accuracy

Table 2: Accuracy.

S. No	Concentration	% Recovery	%RSD
1.	80%	98.45	1.10
2.	100%	98.71	1.05
3.	120%	99.72	0.90

4.5 Repeatability

Table 3: Repeatability.

S. No	Concentration (µg/ml)	Absorbance	Mean	Std. Deviation	%RSD
1.	10µg/ml	1.012	1.054	0.01054	1.05%
2.	10µg/ml	1.124			
3.	10µg/ml	1.051			
4.	10µg/ml	1.022			
5.	10µg/ml	1.011			
6.	10µg/ml	1.104			

4.6 Precision

Table 4: Interday precision data.

S. No	Concentration (µg/ml)	Absorbance			Mean	Std. Deviation	%RSD
		1	2	3			
1.	2 µg/ml	0.234	0.242	0.239	0.238	0.00238	0.23%
2.	3µg/ml	0.332	0.329	0.340	0.333	0.00333	0.33%
3.	4µg/ml	0.445	0.459	0.446	0.45	0.0045	0.4%

Table 5: Intraday precision data.

S. No	Concentration (µg/ml)	Absorbance			Mean	Std. Deviation	%RSD
		1	2	3			
1.	2 µg/ml	0.231	0.222	0.233	0.228	0.00228	0.22%
2.	3 µg/ml	0.335	0.342	0.339	0.339	0.00338	0.33%
3.	4 µg/ml	0.434	0.445	0.438	0.438	0.00439	0.43%

4.7 Robustness

Table 6: Robustness.

Parameter	Parameter sequence	Absorbance	Mean	Std. deviation	%RSD
Wavelength	252nm 250nm 248nm	0.264 0.254 0.250	0.256	0.00256	0.25%

4.8 Assay of lobeglitazone tablet for formulation

Table 7: Assay results for tablets using proposed methods.

Drug	Labelclaim (mg)	Amount found (mg)	% Label Claim Assay (n=3) ± SD
Lobeglitazone sulfate	0.5MG	0.503	100.13±0.95

4.9 Summary

Table 8: Summary of validation parameter.

S. No	Parameter		Normal range	Result
1.	Linearity		0.999	0.998
2.	Precision	Intraday	NMT2%	0.23-0.4%
		Interday	NMT2%	0.22-0.43%
3.	Accuracy	80%	98.45	1.10
		100%	98.71	1.05
		120%	99.72	0.90
4.	Robustness		%RSD≤2	0.25%
5.	Limit of detection		-	0.231
6.	Limit of quantification		-	1.015
7.	Assay		-	100.13±0.95

5. DISCUSSION

The method discussed in the present work provided a convenient and accurate way for analysis of lobeglitazone sulfate in its pharmaceutical dosage form. n- butanol is used as a solvent. Absorbance maxima for lobeglitazone sulfate at 252nm were selected for the analysis. Linearity for detector response was observed in the concentration of 2-6µg/ml. percent label claim for lobeglitazone sulfate in tablet analysis, was found to be 100%. Accuracy of proposed methods was studied and the results expressed as %recovery. Percent recovery for lobeglitazone sulfate was found in the range is 100% indicating the accuracy of all methods.

6. CONCLUSION

The proposed method development of UV – visible spectroscopy isquite, simple, rapid, accurate, precise and sensitive for lobeglitazone sulfate tablet formulation. In validation of UV-visible spectroscopy, it can be concluded that spectroscopic method has been validated. n-butanol is used as a solvent. All the validation parameter like linearity, precision, accuracy, robustness was found to be less than 2%RSD according to ICH guidelines that indicate proposed method is sensitive. Correlation coefficient is 0.998. The value of %RSD for intra-day and inter-day were within normal range and the accuracy was found to be normal range. The limit of detection and limit of quantification of the projected methodology was found to be **0.231µg/ml** and **1.015µg/ml** that is simple and rapid for quantification of lobeglitazone sulfate tablet formulation. It was concluded that developed method is simple, rapid, almost accurate, precise and reliable. In compliance with ICH guideline the method is valid and appropriate for estimation of lobeglitazone sulfate with excellent linearity, precision,

accuracy, robustness and ruggedness. This method can ne used for routine analysis of lobeglitazone sulfate in bulk formulation.

7. REFERENCES

1. Gustav Kirchhoff (left) and Robert Bunsen (right). Introduction to analytical chemistry, copyright © by U. A. Deokate, all rights reserved, 2018.
2. Dr. Abdul Rehman, Introduction to spectroscopy.
3. Harpreet Kaur, Introduction to UV Spectroscopy.
4. Narasimha Kumar. Yadati, Analytical method development.
5. P. P. Sharma, "Validation in pharmaceutical Industry- concepts, approaches & guidelines", vandana publication House, 2007; 1.
6. B. T. Loftus & R.A Nash, "Pharmaceuticals Process Validation" Drugs and pharm Sci. series, Dekkar Inc N. Y, 129: 3.
7. ICH harmonized tripartite guidelines – Validation of analytical procedures: Text and methodology- ICH Q2 (R1).
8. Vikas Vasant Patil, Amol Pawar. Validation of lifecycle, Asian Journal of pharmaceutical analysis, 2022; 215.
9. Sushila Dagadu Chavan, Deepa Mahendra desai, key parameters and statistics of analytical method validation. World journal of advanced research and reviews, 2022; 389-402.
10. Drug profile Lobeglitazone Sulfate **C24H26N4O9S2** https://pubchem.ncbi.nlm.gov.
11. Jeel Dobariya, Dhara Patel, Prasanna Pradhan, Grishma Patel, Dhananjay Meshrama Development and Validation of UV Spectrophotometric methods for simultaneous estimation of Lobeglitazone Sulfate and Glimepiride in combined dosage form.
12. Kalepu Eswar Krishna Sai, Medidi Srinivas, Bula Udaya Kumari, Chepyala Sumalatha Arram Madhavi. Development and validation of an HPLC Method for the Determination of Lobeglitazone in Bulk and in Tablet Formulation. Int. J. Pharm. Investigation, 2024; 14(1): 204-11.
13. Priyanka V and Murugan S. Stability indicating method development and validation of lobeglitazone sulfate by RP-HPLC Method, 2024; 853-861.
14. Sarumathy S, Balamurugan M, Robinson R. Lobeglitazone and Its Therapeutics Benefits: A Review, 2023. Doi 10.7759/cureus. 50085.

15. Chetan Sharma, Dinesh K. Badyal.A comprehensive review on combination of lobeglitazone and glimepiride in management of type 2 (DM) ©2024 National journal of pharmacology and therapeutics.
16. Jung Won Kim, BS; Jung-Ryul Kim, MD, PhD; SoJeong Yi, MS; Kwang-Hee Shin, MS; Hyun-Suk Shin, MS; Seo Hyun Yoon, PhD; Joo-Youn Cho, PhD; Dal-Hyun Kim, PhD; Sang-Goo Shin, MD, PhD; In-Jin Jang, MD, PhD; and Kyung-Sang Yu, MD, PhD. Tolerability and Pharmacokinetics of Lobeglitazone (CKD-501), a Peroxisome Proliferator-Activated Receptor-γ Agonist: A Single- and Multiple-Dose, Double-Blind, Randomized Control Study in Healthy Male Korean Subject, 2011; 33: 1819-1830. Doi: 10.1016/j.clinthera.
17. Kambiz Hassanzadeh, Arman Rahimmi, Mohammad Raman Moloudi, Rita Maccarone, Massino Corbo, Esmeal Izadpanah, Marco Feligioni. Effect of lobeglitazone on motor function in rat model of parkinson's disease with diabetes with diabetes co-morbidity, 2021; 184-192. https://doi.org/10.1016/j.brainressbull.
18. Joonsang Yoo, Jimin Jeon, Minyoul Baik and Jinkwon Kim. Lobeglitazone: A novel thiazolidinedione, for secondary prevention in patients with ischemic stroke: a nationwide nested case-control study. Cardiovasc Diabetol, 2023; 22(1): 106. doi:10.1186/s12933-023-01841-4, PMID 37147722, PMCID PMC10163714.
19. Vibhuti Jadhao, Shashank Joshi, Monika Tandon, Rahul Kodgule, Wen Wu, Sachin Suryawanshi, Hanmant Barkate. Randomized, Double-blind, Phase III Trial of Lobeglitazone Add-on to Metformin in Type 2 Diabetes (SENSITIZE INDIA). Journal of the Association of Physicians India, 2024; 72(1): 32–42.
20. Kwi-Hyun Bae, Jung Beom Seo, Yun-A Jung, Hye-Young Seo, Sun Hee Kang, Hui-Jeon Jeon, Jae Man Lee, Sungwoo Lee, Jung-Guk Kim, In-Kyu Lee, Gwon-Soo Jung, Keun-Gyu Park. Lobeglitazone, a Novel Peroxisome Proliferator-Activated Receptor γ Agonist, Attenuates Renal Fibrosis Caused by Unilateral Ureteral Obstruction (UUO) in Mice. Endocrinol Metab, 2017; 32: 115-123. https://doi.org/10.3803/EnM.2017.32.1.115
21. Piyusha D Gulhane, Shailesh G. Jawarkar. Bioanalytical method development and validation for determination of lobeglitazone in human plasma. Int Res J Modernization Eng Technol Sci, 2023; 5(05): 8103-16.
22. Bong-Hoi Choi, Zhen Jin, Juhong oh, Eun Ae Jeong, Jong Youl Lee, Kyung-ah Park, Kyung Eun Kim, Jung Eun Lee, Hyun-Jin Kim, Jong Ryeal Hahm, Gu Seob Roh. Effects of lobeglitazone on insulin resistant and hepatic seatosis in high-fat die –fed mice, 2018.

23. Kyoung Min Kim, Hyun-Jin Jin, Seo Yeon Lee, Hyo Jin Maeng, Gha Young Lee, Tae Jung Oh, Sung Hee Choi, Hak Chul Jang, Soo Lim, Effects of lobeglitazone, a new thiazolidinedione, on oesteoblastogenesis and bone mineral density in mice. Endocrinol. Metab, 2017; 32(3): 389-395. https;//doi.org/10.3803/EnM.2017.
24. Sin Gon Kim, Doo Man Kim, Jeong-Taek Woo, Hak Chul Jang, Choon Hee Chung, Kyung Soo Ko, Jeong Hyun Park, Yong Soo Park, Sang Jin Kim, Dong Seop Choi. Efficacy and safety of lobeglitazone monotherapy in patients with type 2 diabetes mellitus over 24- weeks: a multi center, randomized, double-blind, parallel-group, placebo-controlled trial. PLOS ONE, 2014; 9(4): e92843. Doi: 10.1371/journal.pone.0092843, PMID 24736628.
25. Jin Ah Jung, Soo-Yun Lee, Tae-Eun Kim, Jung-Ryul Kim, Chin Kim, Wooseong Huh, Jae-Wook Ko: Lack of the effect of lobeglitazone, a peroxisome proliferator-activated receptor-γ agonist, on the pharmacokinetics and pharmacodynamics of warfarin, 2015; 737-743. Doi: 10.2147DDDT.S76591.
26. Kyung-Soo Kim, Sangmo Hong, Hong-Yup Ahn, Cheol-Young Park. Comparative Efficacy of Lobeglitazone Versus Pioglitazone on Albuminuria in Patients with Type 2 Diabetes Mellitus. Diabetes Ther, 2021; 12: 171–181. https://doi.org/10.1007/s13300-020-00948-1.
27. Soo Lim, Eu Jeong Ku, Seo Young Lee, Ji Hyun Lee, Jie- Eun Lee, Kyoung Min Kim, Melanie J Davies. Therapeutic efficacy and safety of initial triple combination of metformin, sitagliptin, and lobeglitazone in drug-naïve patients with type 2 diabetes: initial triple study, 2020; 8: e000807. doi:10.1136/bmjdrc-2019-00080.