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UV METHOD DEVELOPMENT TO FOR QUALITATIVE AND QUANTITATIVE ESTIMATION OF LAWSONE IN ITS GEL FORMULATION

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ABSTRACT

Lawsone is the main active ingredient in the henna plant that is lawsonia inermis. Based on the staining properties of lawsone, it is mainly used for morethan 5000years as hair dye, but also applied to the body. Lawsone corresponds to 2-hydroxy-1,4-naphthoquinone and is present at 1-2% in dried leaves of the plant. Whereas, water hyacinth flowers also used as orange red dye. Michael addition is the process where protein keratin reacts with lawsone ingredient to obtain permanent stain on the skin and hair until it sheds. Lawsone highly interacts with protein keratin to produce dark colored ink. In order to determine the drug in biological fluid or in pharmaceutical preparations, there are no. of methods available, that is HPTLC, HPLC, and spectrophotometery. The new, simple, reliable, rapid, precise ultraviolet spectrophotometric method has to validate and been developed to analyses lawsone in bulk & poly-herbal formulation. It can be concluded that the proposed method is simple, rapid, accurate, precise, economic and reproducible for UV spectro-photometric estimation of Lawsone from pharmaceutical formulation. This method for routine estimation of Lawsone in bulk and pharmaceutical dosage form was successfully applied.

Keywords: Lawsone, UV, Development, Estimate.

INTRODUCTION

Lawsone is the main active ingredient in the henna plant that is lawsonia inermis. Based on the staining properties of lawsone, it is mainly used for morethan 5000years as hair dye, but also applied to the body. Lawsone corresponds to 2-hydroxy-1,4-naphthoquinone and is present at 1-2% in dried leaves of the plant. Whereas, water hyacinth flowers also used as orange red dye. Michael addition is the process where protein keratin reacts with lawsone ingredient to obtain permanent stain on the skin and hair until it sheds. Lawsone highly interacts with protein keratin to produce dark colored ink. When this lawsone concentration decreases, it undergoes breakdown and shows fading of tattoo ink. UV light was strongly absorbed by lawsone and aqueous extracts are effective to use sunless tanning lotion & sunscreen. Lawsone extract from henna found in walnuts which is similar to juglone. Both are naphtholenes. The study of lawsone provided both are effective in blocking UV light through sunscreen products [1-4].

In order to determine the drug in biological fluid or in pharmaceutical preparations, there are no. of methods available, that is HPTLC, HPLC, and spectrophotometery. The new, simple, reliable, rapid, precise ultraviolet spectrophotometric method has to validate and been developed to analyses lawsone in bulk & poly-herbal formulation. Statistical tests are conducted on validation data [5-7].

MATERIALS AND METHODS

Instrument Used:

UV-Vis spectrophotometer 1700, Make: Shimadzu, Kyoto, Japan, Scan speed: 40nm/min, Bath Sonicator

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Reagents and Solutions

All the reagents used in this assay were of analytical grade. Poly herbal gels of Lawsone (Henna) were purchased.

EXPERIMENTAL

Determination of λ_{max}

Weighed amount of Lawsone was dissolved in 0.1N NaOH to obtain a 100µg/ml solution. Scan this solution between 200-400nm to determine maximum absorption. Therefore, they studied the dilution effect on maximum absorption by diluting the stock solution 20ug/ml and has been scanned in the range of 200-400nm.

Preparation of Standard Stock Solution

Prepare the standard drug solution of Lawsone by dissolving 10 mg Lawsone in 100 ml 0.1N NaOH to obtain 100 µg/ml Concentration of stock solution.

Preparation of Calibration Curve

Prepare the calibration curve in 0.1N NaOH at λ_{max} 276nm by using UV-Visible spectrophotometer Model 1700. Prepare 100ug/ml for this stock solution. Serial dilution of 10, 15, 20, 25, 30µg/ml were prepared and absorbance was taken at λ_{max} 276nm. 6sets of average values have been taken for calibration curve and solution is scanned between 200-400 nm against blank.

Assay

500mg of gel containing of 5 mg of Lawsone was weighed. Gel equivalent to 100 mg of Lawsone was transferred into 100 ml volumetric flask dissolved in 0.1N NaOH. The solution was then filtered through Whatmann filter paper No 40 (0.45 micron).Aliquots of the sample were removed and diluted to 10 ml of 0.1N NaOH to obtain strengths of 20µg/ml determined at the respective absorbance of 276nm against 0.1N NaOH as a blank [8-11].

Limit of Detection (LOD) and Limit of Quantification (LOQ)

Determine the LOD and LOQ of GLP by using standard deviation of the response and slope approach as defined in ICH guidelines. The LOD and LOQ are seen in table 1. Claculate the LOD & LOQ by the equation (1) LOD= 3.3δ/*s* and (2) LOQ = 10 δ/*s* respectively, where δ is the standard deviation of blank and *s* is slope.

Recovery studies

Judgement of the accuracy method was performed by recovery studies. Recovery studies are performed by the addition of known quantity of pure drug to pre-analyzed formulation and follow the proposed method. Recovery % is calculated from the amount of drug present. At 3 different concentration levels, recovery study were conducted that standard drug is added to the sample.

RESULTS AND DISCUSSION

The UV scan of standard solution between 200 – 400 nm showed the absorption maxima at 271 nm. The overlay spectra of different concentration range of standard Lawsone was recorded. The Beer's law was verified from the calibration curve by plotting a graph of concentration vs. absorbance. The linearity range was observed between 11-33µg/ml. The plot clearly showed a straight line passing through origin with equation Y= 0.0315X - 0.0358 with correlation coefficient of 0.998. The coefficient of correlation was highly significant. The optical characteristics and other validation parabymeters are thus summarized in table1. The assay method was validated by low values of standard deviation and standard error, indicating accuracy and precision in table2 of the methods. Excellent recovery studies further prove the accuracy of the method table3. The assay result was repeated for three times which was found to be 100.23-101.78% of labelled claim in table 4 [12,13].

Table 1: Optical Parameters for Lawsone

S. No.	Parameters	values
1	max(nm)	271
2	linearity range	11-33µg/ml
3	regression equation	Y=0.0315X-0.0358
4	correlation coefficient	0.998
5	slope	0.0347
6	intercept	0.0296
7	Limit of detection(µg/ml)	0.6564
8	Limit of quantification(µg/ml)	2.7829

Table 2: Precision Data for Lawsone

S. No.	Conc. ug/ml	intraday	cv	Interday	cv
1	15	0.4792±0.00587	0.8912	0.4197±0.0053	2.353
2	20	0.6450±0.0192	2.698	0.6196±0.0089	3.401
3	25	0.7924±0.0046	0.4821	0.795±0.0042	0.694

Table 3: Recovery Study Data for Lawsone

S. No.	Amount of sample (ug/ml)	Added drug (ug/ml)	Drug recovered (ug/ml) ±sd	%recovery
1	20	0	20.7581±0.36322	99.5986
2	20	10	30.8149±0.2790	99.1472
3	20	20	40.3268±0.4934	99.3109
4	20	30	50.2923±0.5907	99.4143

Table 4: Assay Results for Lawsone

S. No.	Actual conc. (µg/ml)	Amount obtained (µg/ml)	%drug
1	20	19.95	100.23
2	20	20.12	101.78
3	20	20.49	100.54

CONCLUSION

It can be concluded that the proposed method is simple, rapid, accurate, precise, economic and reproducible for UV spectro-photometric estimation of Lawsone from pharmaceutical formulation. This method for routine estimation of Lawsone in bulk and pharmaceutical dosage form was successfully applied.

REFERENCES

1. Goodman and Gilman's, The pharmacological basis of therapeutics,11th edition, edited by Laurence L. Brunton, John S. Lazo, Keith L. Parkar, McGraw-Hill, Medical publishing division,2006:1635-1638.
2. Dhabale, Seervi C.R: Simple, accurate, precise, reproducible and economical procedures for simultaneous estimation of Glipizide and Metformin hydrochloride in tablet dosage form. International Journal of ChemTech Research, 2009; Vol.2: 813-817.
3. Darshana K. Modi and Bhavesh H. Patel, Simultaneous determination of metformin hydrochloride and glipizide in tablet formulation by HPTLC, J. of Liquid Chromatography and Related Technologies, 2012; 35(1):28-39.
4. P. Venkatesh, T. Harisudhan, Hira Choudhury, Ramesh Mullangi, Nuggehally R. Srinivas: Simultaneous estimation of six anti-diabetic drugs-glibenclamide, gliclazide, glipizide, pioglitazone, repaglinide and rosiglitazone: Development of a novel HPLC method for use in the analysis of pharmaceutical formulations and its application to human plasma assay. Biomedical Chromatography, 20: 1043–1048.
5. Yu H Nola, Ho NM Emmie, Tang P. W Francis, Wan S.M. Terence:To develop the simple, stability-indicating reversed-phase high-performance liquid chromatographic (RP-HPLC) method for determination of Glipizide in guinea pig plasma, Journal of Chromatography A,2008; Vol.1, Issues 1-2: 426-434.
6. S Dhawan, A K Singla, High Performance liquid chromatographic analysis of glipizide: application to *in vitro* and *in vivo* studies. J Chromatogr Sci. 2003; 41 (6):295-300.
7. Swaroop R. Lahoti, Prashant K. Puranik, Ashish A. Heda, Rajesh B. Navale: Development and Validation of RP-HPLC Method for Analysis of Glipizide in Guinea Pig Plasma and its Application to Pharmacokinetic Study, International Journal of Pharm Tech Research, 2010 ;2(3) : 1649-1654.
8. Shaikh Rahila, Karigar Asif: Reverse phase high performance liquid chromatographic method for the analysis of glipizide in pharmaceutical dosage forms, International Journal of Research in Ayurveda & Pharmacy, 2010; 1(2): 455-458.
9. S. AbuRuza, b, J. Millershipb and J. McElnayb: The development and validation of liquid chromatography method for the simultaneous determination of metformin and glipizide, gliclazide, glibenclamide or glimepiride in plasma, J of Chromatography B, 2005; 817 (2): 277-286.
10. B. Udaykumar Rao and Anna Pratima Nikalje: Determination of Glipizide, Glibenclamide and Glimeperide in a Tablet Dosage Form in the Presence of Metformin Hydrochloride by Ion Pair –Reversed Phase Liquid Chromatographic Technique, J Anal Bioanal Techniques, 2010; 1(2):105.

11. Anna Gumieniczek and Anna Berecka, Quantitative analysis of gliclazide and glipizide in Tablets by a new validated and stability-indicating RPTLC method, J of Planar Chromatography, 2010; 23(2): 129-133.
12. International conference on Harmonization, Guidance for Industry In; Q2A Text on Validation of Analytical Methods, Switzerland; IFPMA 1994;1-4.
13. International conference on Harmonization, Guidance for Industry In; Q2B Validation of Analytical Procedures, Methodology, Switzerland; IFPMA 1996; 1-8.