*EJPMR, 2025, 12(12), 338-347* **Research Article** **Impact Factor 7.065**

**EUROPEAN JOURNAL OF PHARMACEUTICAL AND MEDICAL RESEARCH**

www.ejpmr.com

**ISSN (O): 2394-3211**
**ISSN (P): 3051-2573**
**Coden USA: EJPMAG**

# ESTIMATION AND EVALUATION OF LAWSONE IN DIFFERENT HENNA SAMPLES BY USING VARIOUS ANALYTICAL TECHNIQUES

**Laya M., Sridevi P.***

[1]Student, [2]Professor
Department of Pharmaceutical Analysis, Sri Venkateshwara College of Pharmacy, Madhapur, Hitech City Road, Hyderabad, 500081.

***Corresponding Author: Sridevi P.**

Professor & Head, Department of Pharmaceutical Analysis, Sri Venkateshwara College of Pharmacy, Madhapur, Hitech City Road, Hyderabad, 500081.

**DOI:** https://doi.org/10.5281/zenodo.17735755

**How to cite this Article:** Laya M., Sridevi P.* (2025). ESTIMATION AND EVALUATION OF LAWSONE IN DIFFERENT HENNA SAMPLES BY USING VARIOUS ANALYTICAL TECHNIQUES. European Journal of Biomedical and Pharmaceutical Sciences, 12(12), 338–347.


**Article Received on 03/11/2025** **Article Revised on 24/11/2025** **Article Published on 01/12/2025**

## ABSTRACT

Lawsonia inermis, often known as henna, is a natural dye plant that is widely used for cosmetic and medicinal purposes. Its main active component, lawsone (2-hydroxy-1,4-naphoquinone), is what gives henna its characteristic reddish-brown colour and therapeutic properties. In order to determine and assess the amount of Lawsone in both natural and commercial henna samples, the current study used UV-visible spectroscopy, HPLC, atomic absorption spectroscopy, and infrared spectroscopy. Cold maceration and sonication were used to create methanolic extracts for efficient Lawsone recovery. UV-visible spectroscopy was utilized to get λ max and initial quantification. Additionally, an HPLC method was created in compliance with ICH guidelines. The use of Atomic Absorption Spectroscopy was used to assess sample safety and heavy metal detection. The developed analytical techniques were shown to be suitable for henna and were straightforward, accurate, precise, and reliable.

**KEYWORDS:** Lawsone, Henna, UV-Visible Spectroscopy, IR, HPLC, Atomic Absorption Spectroscopy.

## INTRODUCTION

Determining the quality, purity, and authenticity of various henna samples requires the quantification and assessment of Lawsone. The primary bioactive pigment in henna, lawsone, may be precisely identified and measured using a variety of analytical methods. Reliable data on the chemical makeup, functional groups, and concentration of Lawsone can be obtained using techniques like UV–visible spectroscopy, HPLC, and infrared (IR) spectroscopy. These analytical techniques enhance quality control in both commercial and research applications and aid in the standardization of henna products.

### Analytical methods of Modern spectroscopic and chromatographic methods

**UV-VISIBLE SPECTROSCOPY:** UV-visible spectroscopy is based on the idea that molecules absorb ultraviolet or visible light, which causes electrons to shift from lower to higher energy levels. According to Beer-Lambert's law, the amount of light absorbed is directly proportional to the substance's concentration**.**

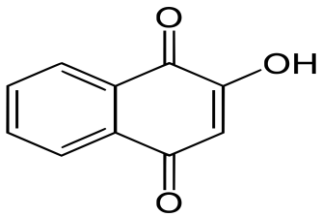
**HPLC:** HPLC is that various components in a mixture separate because they interact differently with the stationary phase and the mobile phase. As a result, each compound moves through the column at a different speed and elutes at a different time

**INFRARED SPECTROSCOPY:** Infrared spectroscopy is that molecules absorb infrared light, which causes vibrational changes like stretching and bending. Only vibrations involving a change in dipole moment result in an IR absorption signal.

**ATOMIC ABSORPTION SPECTROSCOPY:** AAS operates on the basis that light of a certain wavelength is absorbed by free atoms in their ground state. When a sample is atomized in a flame or furnace, the amount of light absorbed by these atoms is exactly proportional to the concentration of the element present.

**DRUG PROFILE**

- **Scientific name:** Lawsonia inermis
- **Family:** Lythraceae
- **Genus:** Lawsonia
- **Kingdom:** Plantae
- **Order:** Myrtales




$C_{10}H_6O_3$

**MATERIALS AND METHODS**

**Study Area**

The entire study was carried out in Sri Venkateshwara College of Pharmacy, Hyderabad, Telangana in 2025.

**CHEMICALS USED**

The chemicals used in this study include standard Lawsone from Sigma Aldrich with 96.5% purity, methanol for HPLC from Merck Life Science and ethanol also supplied by Merck Life Science. Hydrochloric acid was obtained from SD Fine Chem Limited, while HPLC-grade water was sourced from Avantor. Additionally, acetonitrile for HPLC was procured from SD Fine Chem Limited.

**INSTRUMENTS USED:** The laboratory is equipped with a UV/VIS Spectrophotometer from Lab India, an HPLC system from Shimadzu and Analytical balance manufactured by Sartorius. Additionally, Borosil beakers and conical flasks are used for sample handling, along with pipettes and test tubes for precise measurement and transfer of liquids.

**METHODS**

**MORPHOLOGICAL ANALYSIS**

Different types of Henna samples were collected and morphological characteristics were examined such as plant type, leaves, flowers, leaf type, shape, leaf surface, leaf texture, colour, taste and odour.

**PHYTOCHEMICAL ANALYSIS**

1. **ALKALOIDS:** The Wagner's test involved dissolving a plant extract in Wagner's reagent, which is iodine in potassium iodide. Additionally, the colour look suggests that the Henna extract contains alkaloids.
2. **AMINO ACIDS:** NINHYDRIN TEST: After dissolving plant extract in 0.25% Ninhydrin reagent and boiling it for a short while, the colour that forms shows that amino acids are present.
3. **PHENOLIC COMPOUNDS:** FERRIC CHLORIDE TEST: 3-4 drops of ferric chloric solution were added to plant extract and the colour that formed show the presence of phenolic chemicals.
4. **REDUCING SUGARS:** BENEDICTS TEST: 3-4 drops of Benedict's reagent were applied to a plant extract, which was then gently heated. The emergence of colour indicated the presence of reducing sugars.
5. **FLAVONOIDS:** LEAD ACETATE TEST: After adding a few drops of lead acetate to plant extract, the colour changes to show the presence of flavonoids.

**QUALITATIVE ANALYSIS OF LAWSONE**

**THIN LAYER CHROMATOGRAPHY**

**Preparation of Standard & Sample solution**

Standard solution preparation involved accurately weighing one gram of standard Lawsone in a 10-millilitre volumetric flask and then adding 10 millilitres of ethanol to the flask.

Sample solution preparation involved transferring 10 mL of the extracted sample solution into a 10 mL volumetric flask.

**Procedure:** The TLC analysis was carried out by spotting the standard (plot A) and three samples solutions (plot B, C and D) on to the prepared TLC plate using capillary tubes. The plate was then placed in a beaker containing the mobile phase, prepared with ethanol and acetonitrile and sealed with a Petric plate. Once the solvent front reached about 70% of the plate's height, the TLC plate was removed and allowed to air dry. After drying, the plate was observed under a UV chamber at 283 nm to compare the spots with the standard and confirm the presence of Lawsone.

**METHOD DEVELOPMENT OF LAWSONE BY UV SPECTROSCOPY**

**Preparation of Standard solution**

In a 10 mL volumetric flask, one gram of standard Lawsone was dissolved in methanol. After 20 minutes of sonication, the solution was let to cool ambient temperature. Methanol was used to dilute one millilitre of this solution to 10 mL.

**Preparation of Sample solution**

A 10 mL volumetric flask was filled with one gram of each henna sample, which was then filtered through Whatman paper after being extracted with methanol using cold maceration, occasional shaking, and 15 minutes of sonication. For additional examination, 1 mL of the extract was diluted to 10 mL with methanol after the filtrate had cooled to room temperature.

**MULTIVARITE CALIBRATION OF LAWSONE BY UV SPECTROSCOPY**

**Preparation of sample solution**

In a 10 mL volumetric flask, one gram of each sample was dissolved in methanol. For twenty minutes, the mixtures were sonicated. They were filtered through

Whatman filter paper once they had cooled. Methanol was used to dilute one milliliter of each filtrate to ten millilitres. Methanol was used to dilute one milliliter of each of the test and reference solutions. Next, each solution's absorbance was measured.

## VALIDATION PARAMETERS

### LINEARITY

Consider the standard to be the blank. Now, make 10 µg/mL of mixture from the sample solution. A number of wavelengths around the drugs ƛ max (450 nm) are then used to measure absorbance. Accordingly, 444, 446, 448, 450, 452, 454 and 456 nm. The absorbance values of each solution were noted and shown. At various wavelengths, the residual plots and calibration graphs were displayed.

### ACCURACY

The test solution receives known additions of Standard Lawsone at 75%, 100% and 125% respectively. 10 mL of the standard solution was pipetted into three separate volumetric flasks from the produced stock solutions of the standard and sample. Then, 0.1, 0.6 and 1.1 mL of the sample solution were added to the volumetric flasks and the volume was increased to 10 mL with methanol. The recovery percentages were computed. Recovery study findings are noted.

### PRECISION

Precision is a repeatability process. It is done by taking 30 µg/mL concentration. Precision is performed as intraday and inter-day precision, where one concentration is analysed for 5 times and mean is calculated to find standard deviation and %RSD at different selected wavelengths.

### LOD AND LOQ DETECTION

The lowest level of analyte found in the sample is known as the LOD. The lowest quantity of analyte in the sample, as established by accuracy and precision, is known as the limit of quantification (LOQ).

Where,

LOD = 3.3* σ/m

LOQ = 10* σ/m

### ASSAY

The amount of Lawsone in the sample was assessed by measuring the extracted samples absorbance at 450 nm.

### Residual plots

Analyte (X) absorbance is measured at seven different wavelengths (λ= 444, 446, 448,450, 452, 454 and 456 nm). The following equation can be expressed for each of the chosen wavelengths.

$$C_x = \frac{A_T - K_T}{a=b=c=d=e=f=g}$$

## METHOD DEVELOPMENT OF LAWSONE BY RP-HPLC

### Procedure

Accurately weight and transfer 1 gram of standard and sample Lawsone into a 10 mL clean dry volumetric flask separately. To this add methanol as diluent and sonicate to dissolve it completely and made-up volume to the mark with the same solvent. This makes the concentration of 0.1mg/ml solution. Measure the regions to determine the amount of lawsone present after injecting 10 µL of the reference sample into the chromatographic apparatus.

## DETERMINATION OF LAWSONE BY INFRARED SPECTROSCOPY

### Procedure

One millilitre of the extract was diluted with methanol, and then a tiny drop was put on a NaCl plate. To spread the sample into a thin, even layer, the second plate was carefully placed on top. For additional examination, the produced plates were then stored in the sample holder.

## DETERMINATION OF LAWSONE BY ATOMIC ABSORPTION SPECTROSCOPY

### Preparation of standard & Sample solution

- Lawsone stock standard and sample solutions were prepared.
- Diluted in increments of 0.5, 1, 2, 5 and 10 parts per million.
- Distilled water to dilute the solution to volume after filtering it.

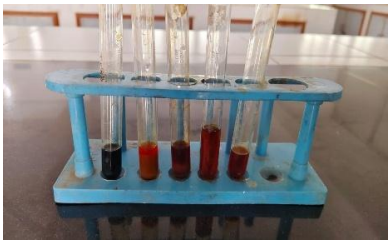
## RESULTS AND DISCUSSIONS

### Morphological Evaluation

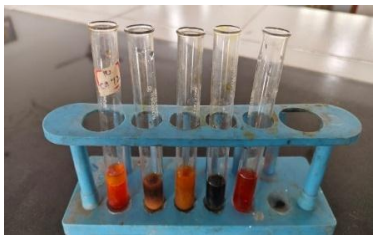
| Characteristics | Observations |
|---|---|
| Leaf | Simple leaf |
| Shape of the leaf | Ovate to elliptic |
| Leaf surface | Smooth and glabrous |
| Leaf texture | Thin, papery |
| Colour | Fresh leaves- Bright green |
| | Dried leaves- Brownish |
| Taste | Slightly bitter |
| Odour | Mildly aromatic when crushed |

### PHYTO-CHEMICAL ANALYSIS

| S.NO | Test parameters | Observations |
|---|---|---|
| 1 | Alkaloids (Wagners Test) | Brown Precipitate Was Observed |
| 2 | Amino acids (Ninhydrin Test) | Blue Colour Was Observed |
| 3 | Phenolic compounds (Ferric Chloride Test) | Bluish Black Colour Was Observed |
| 4 | Reducing sugars (Benedicts Test) | Orange Red Precipitate Was Observed |
| 5 | Flavonoids (Lead Acetate Test) | Yellow Colour Was Observed |



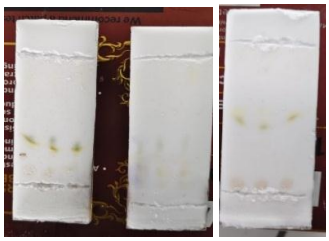
**Phyto-chemical analysis of standard**



**Phyto-chemical analysis of sample**

**THIN LAYER CHROMATOGRAPHY OF LAWSONE**

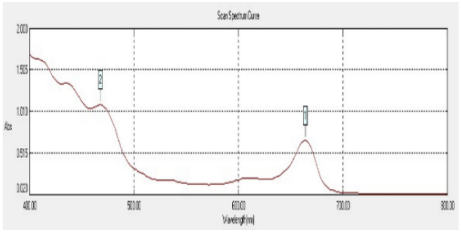
**TLC Profiling of Standard**



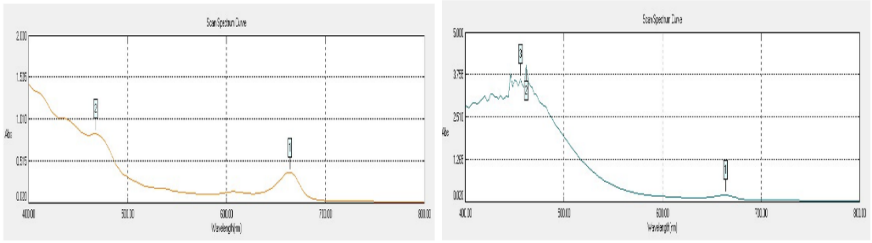
**TLC Profiling of different samples**

**METHOD DEVELOPMENT OF LAWSONE BY UV-VISIBLE SPECTROSCOPY**

The wavelenghts of Lawsone was found at 450 nm by comparing it with standard Lawsone and the samples.



**UV – Spectrum of standard Lawsone**

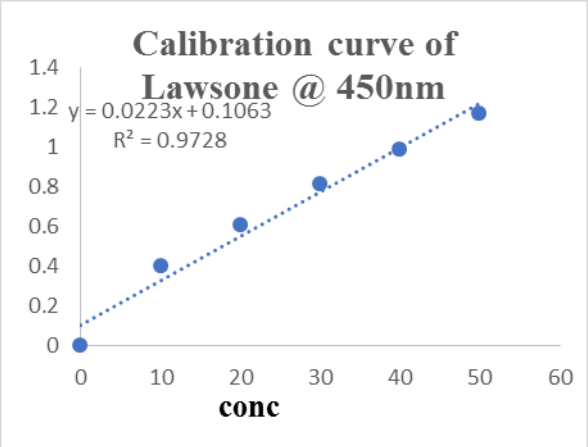


**UV – Spectra of Sample Lawsone**

**Linearity Data**

**1. Linearity data of Lawsone observed at 450 nm.**

| Conc (µg/mL) | Absorbance (nm) |
|---|---|
| | 450 |
| 10 | 0.402 |
| 20 | 0.611 |
| 30 | 0.816 |
| 40 | 0.986 |
| 50 | 1.171 |




**ACCURACY DATA**

| CONCENTRATION LEVELS | AMOUNT RECOVERED | STD DEVIATION | % RSD |
|---|---|---|---|
| 75% | 280.7 | 0.40 | 0.5 |
| 100% | 247.6 | 0.4 | 0.5 |
| 125% | 219.91 | 0.31 | 0.4 |

**PRECISION DATA**

**2. Precision data of Lawsone observed at 450 nm.**

| S.NO | INTRADAY | INTERDAY |
|---|---|---|
| 1 | 0.816 | 0.816 |
| 2 | 0.816 | 0.816 |
| 3 | 0.815 | 0.816 |
| 4 | 0.816 | 0.815 |
| 5 | 0.816 | 0.816 |
| Average | 0.8158 | 0.8158 |
| STD deviation | 0.0004472 | 0.000447214 |
| % RSD | 0.05 | 0.05 |

**LOD AND LOQ DETECTION**

**Limit od detection (LOD) µg/mL**

| Y=3.3*0.0223/0.1063 | Y VALUE (Abs) | σ (slope) | m (Intercept) | LOD |
|---|---|---|---|---|
| LOD= 3.3*σ/m | 0.816 | 0.0223 | 0.1063 | 0.692 |

**Limit of Quantification (LOQ) µg/mL**

| Y=10*0.0223/0.1063 | Y VALUE (Abs) | σ (slope) | m (Intercept) | LOQ |
|---|---|---|---|---|
| LOQ=10*σ/m | 0.816 | 0.0223 | 0.1063 | 2.098 |

**ASSAY**

| | ASSAY PARAMETERS (n=3) | | AMOUNT OF LAWSONE FOUND |
|---|---|---|---|
| **Observation- 1** | **Standard Abs** | 2.231 | |
| | **Sample Abs** | 1.823 | 81.757 |
| **Observation-2** | **Standard Abs** | 2.231 | |
| | **Sample Abs** | 1.823 | 81.712 |
| **Observation- 3** | **Standard Abs** | 2.231 | |
| | **Sample Abs** | 1.831 | 81.757 |
| | | Mean | 245.23 |
| | | Average | 81.74 |
| | | STD deviation | 0.03 |
| | | % RSD | 0.1 |

**VALIDATION PARAMETERS**

**ACCURACY**

| WAVELENGTH | CONCENTRATION LEVELS | AMOUNT RECOVERED | STD DEVIATION | % RSD |
|---|---|---|---|---|
| **444** | 75 | 252.3 | 0.40 | 0.2 |
| | 100 | 222.6 | 0.4 | 0.2 |
| | 125 | 198.19 | 0.16 | 0.1 |
| **446** | 75 | 259.2 | 0.40 | 0.2 |
| | 100 | 228.7 | 0.4 | 0.2 |
| | 125 | 203.04 | 0.31 | 0.2 |
| **448** | 75 | 253.1 | 0.20 | 0.2 |
| | 100 | 222.9 | 0.2 | 0.2 |
| | 125 | 198.10 | 0.16 | 0.1 |
| **450** | 75 | 280.7 | 0.40 | 0.5 |
| | 100 | 247.6 | 0.4 | 0.5 |
| | 125 | 219.91 | 0.31 | 0.4 |
| **452** | 75 | 265.9 | 0.20 | 0.1 |
| | 100 | 234.1 | 0.2 | 0.1 |
| | 125 | 207.61 | 0.16 | 0.1 |
| **454** | 75 | 275.5 | 0.20 | 0.1 |
| | 100 | 242.9 | 0.2 | 0.1 |
| | 125 | 215.9 | 0.16 | 0.1 |
| **456** | 75 | 275.6 | 0.20 | 0.1 |
| | 100 | 243.2 | 0.2 | 0.1 |
| | 125 | 216.14 | 0.16 | 0.1 |

**PRECISION**

**ABSORBANCE VALUES FOR INTRADAY PRECISION**

| Conc (µg/mL) | ABSORBANCE (nm) | | | | | | |
|---|---|---|---|---|---|---|---|
| | **444** | **446** | **448** | **450** | **452** | **454** | **456** |
| 10 | 0.736 | 0.754 | 0.735 | 0.816 | 0.772 | 0.801 | 0.802 |
| 20 | 0.736 | 0.754 | 0.735 | 0.816 | 0.772 | 0.801 | 0.801 |
| 30 | 0.734 | 0.756 | 0.735 | 0.815 | 0.771 | 0.802 | 0.802 |
| 40 | 0.736 | 0.754 | 0.736 | 0.816 | 0.772 | 0.801 | 0.802 |
| 50 | 0.736 | 0.754 | 0.735 | 0.816 | 0.772 | 0.801 | 0.802 |
| Average | 0.7356 | 0.7544 | 0.7352 | 0.8158 | 0.7718 | 0.8012 | 0.8018 |
| STD | 0.0008944 | 0.0008944 | 0.0004472 | 0.0004472 | 0.0004472 | 0.0004472 | 0.0004472 |
| % RSD | 0.12 | 0.12 | 0.06 | 0.05 | 0.06 | 0.06 | 0.06 |

**ABSORBANCE VALUES FOR INTERDAY PRECISION**

| Conc (µg/mL) | ABSORBANCE (nm) | | | | | | |
|---|---|---|---|---|---|---|---|
| | **444** | **446** | **448** | **450** | **452** | **454** | **456** |
| 10 | 0.736 | 0.754 | 0.735 | 0.816 | 0.771 | 0.801 | 0.802 |
| 20 | 0.734 | 0.754 | 0.736 | 0.816 | 0.772 | 0.802 | 0.802 |
| 30 | 0.736 | 0.754 | 0.735 | 0.816 | 0.772 | 0.801 | 0.802 |
| 40 | 0.736 | 0.756 | 0.735 | 0.815 | 0.772 | 0.801 | 0.801 |
| 50 | 0.736 | 0.754 | 0.735 | 0.816 | 0.772 | 0.801 | 0.802 |
| Average | 0.7356 | 0.7544 | 0.7352 | 0.8158 | 0.7718 | 0.8012 | 0.8018 |
| STD | 0.000894427 | 0.000894427 | 0.000447214 | 0.000447214 | 0.000447214 | 0.000447214 | 0.000447214 |
| % RSD | 0.12 | 0.12 | 0.06 | 0.05 | 0.06 | 0.06 | 0.06 |

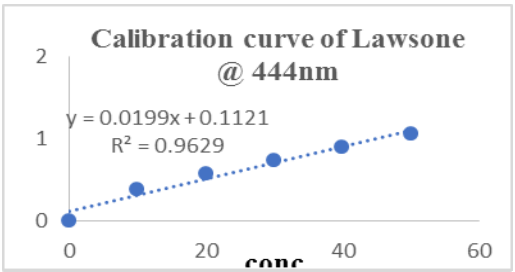
**LOD AND LOQ DETECTION**

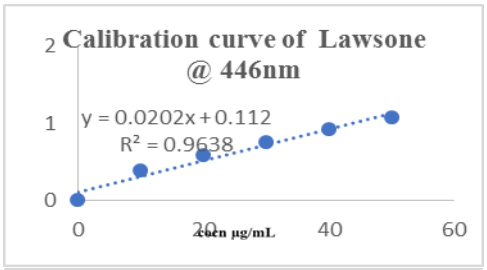
$$LOD = 3.3 * \frac{\sigma}{m} \quad LOQ = 10 * \frac{\sigma}{m}$$

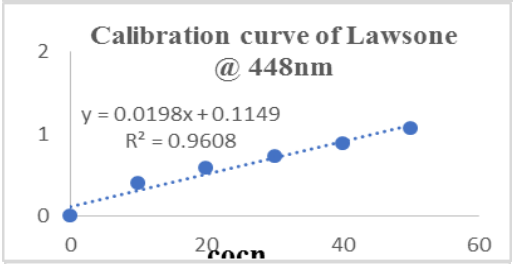
**LOD AND LOQ DATA**

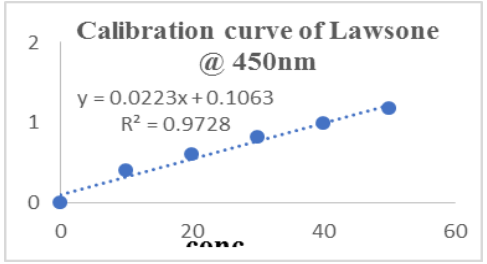
| WAVEWLENGTH (nm) | σ (Slope) | m (Intercept) | LOD Values | LOQ Values |
|---|---|---|---|---|
| **444** | 0.0199 | 0.1121 | 0.350 | 0.022 |
| **446** | 0.0202 | 0.1120 | 0.595 | 1.803 |
| **448** | 0.0164 | 0.2412 | 0.224 | 0.679 |
| **450** | 0.0223 | 0.1063 | 0.692 | 2.098 |
| **452** | 0.0212 | 0.1138 | 0.615 | 1.863 |
| **454** | 0.0209 | 0.1267 | 0.544 | 1.650 |
| **456** | 0.0217 | 0.1167 | 0.061 | 0.186 |

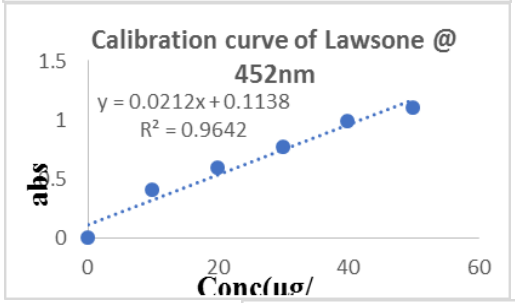
**LINEARITY**

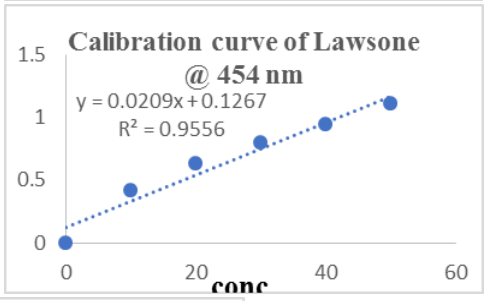


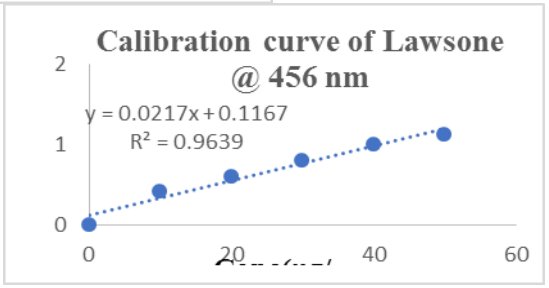



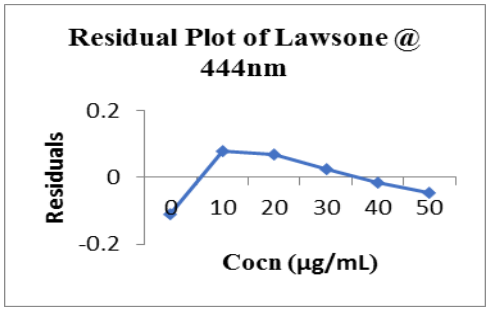



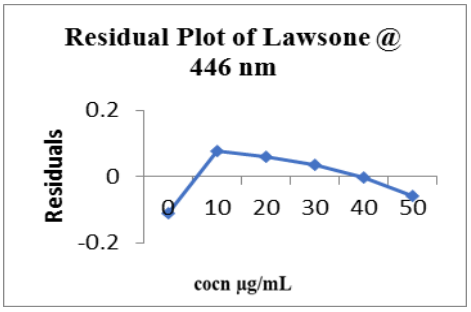



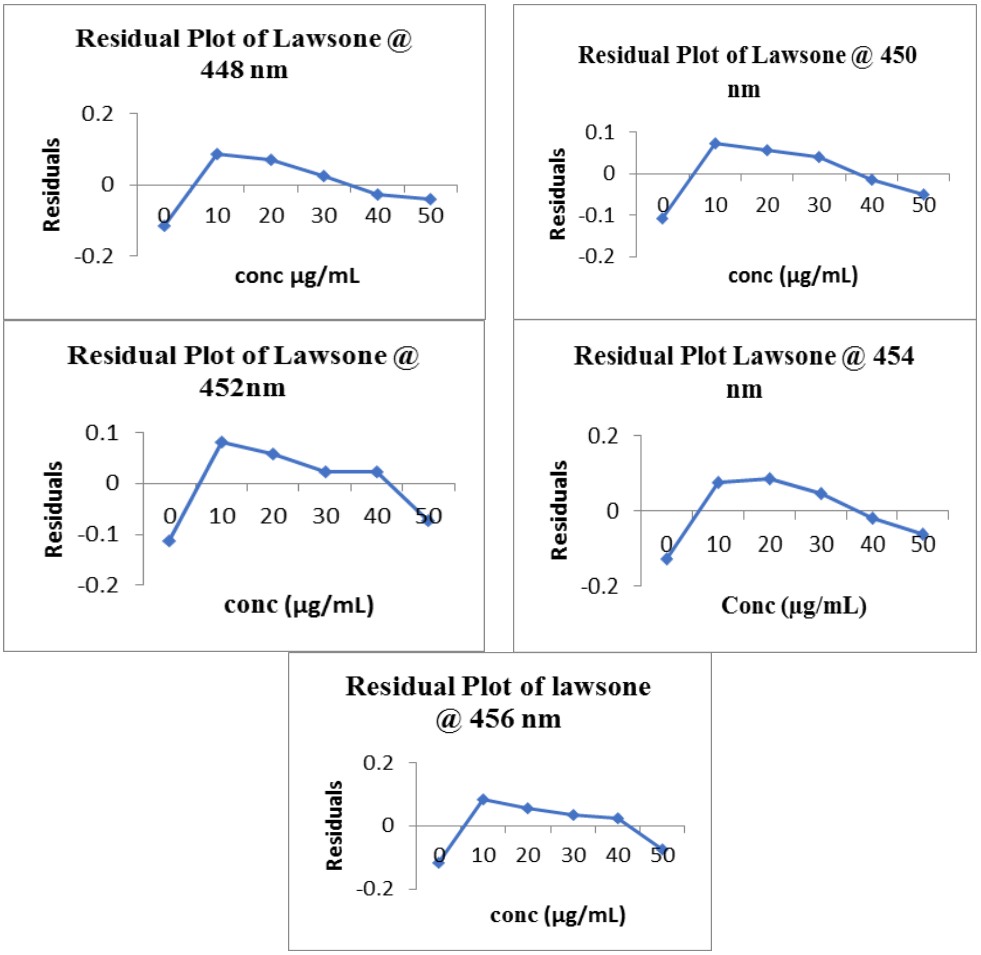




**Residual Plots**






**METHOD DEVELOPMENT OF LAWSONE BY RP-HPLC**

**Optimized method**

Stationary phase- Inertsil RP- C18

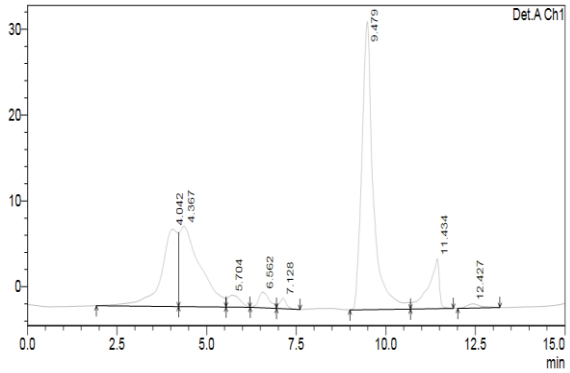
Eluent Composition – MP A -Methanol: MP B- HPLC water (60:40)

Spruge rate- 1.0 mL/min

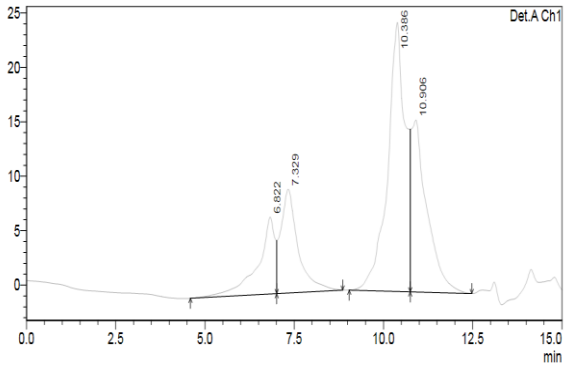
Quantification – 282nm

Runtime – 15 min

Inject volume- 20 µL



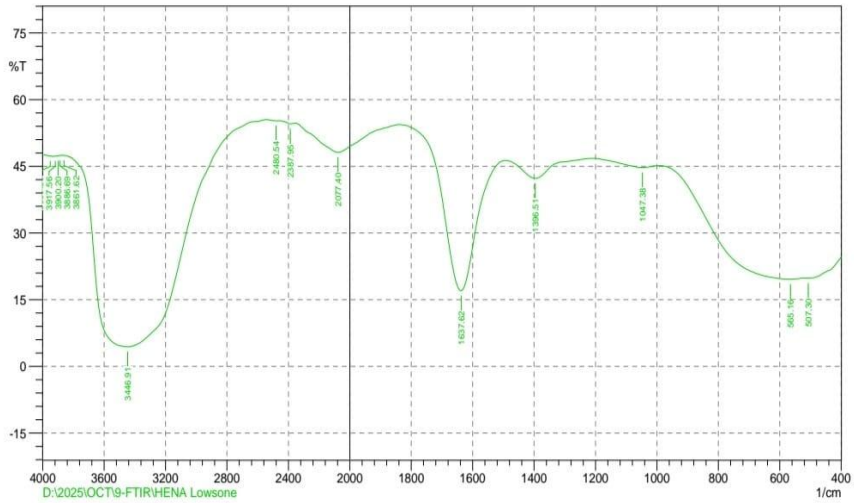

**Standard chormatogram of Lawsone**




**Sample chromatogram of Lawsone**

**DETERMINATION OF LAWSONE BY INFRARED SPECTROSCOPY**

| Functional group | Absorption Range ($cm^{-1}$) | Values Observed ($cm^{-1}$) |
|---|---|---|
| O-H | 3000 – 3700 | 3446 |
| C=O | 1650 – 1750 | 1637 |
| C-O | 1000 – 1350 | 1047 |
| C=C | 1600 - 1700 | 1396 |




**DETERMINATION OF LAWSONE BY ATOMIC ABSORPTION SPECTROSCOPY (AAS)**

| Elements | Limits (ppm) | Values Observed (ppm) |
|---|---|---|
| Nickel (Ni) | ± 1 | 0.0244 |
| Copper (Cu) | ± 20 | 0.0964 |
| Lead (Pb) | ± 10 | 0.2113 |
| Cadmium (Cd) | ± 0.3 | 0.0139 |

## CONCLUSION

Using a variety of analytical methods, including UV–visible spectroscopy, IR spectroscopy, HPLC, and AAS, the quantification and evaluation of Lawsone in diverse henna samples revealed significant variations in Lawsone content between natural and commercial samples. The identity and purity of Lawsone were supported by the IR spectrum analysis, which verified the existence of distinctive functional groups. While AAS guaranteed the identification of any heavy metal impurities, UV-visible and HPLC techniques offered precise quantification. All things considered, the combined analytical technique worked well for evaluating the safety, authenticity, and quality of henna products.

## REFERENCES

1. Muhammad Aslam, Sana Siddique, Muhammud Zia-Ur-Rehman, Zahra Noreen, Yousuf Abbas, Aamir Sohail, Muhammad Aneeq Javed, Madiha Rashid, Aamir Ali. Isolation and Identification of Lawsonia content from Leaves of Henna (Lawsonia inermis).Scientific Inquiry and Review, 2023.
2. Vaihnavi Dinesh Sarmalkar, Palvi Satish Kamut, Dr. Naomita Dhume, Maria Seelan Nadar, Sakshi Suryant Shinde, Asfina Shaikh. Lawsonia Inermis (Henna): A Comprehensive Review of its therapeutic potential, Phytochemistry and Risks of chemical Adulteration. World Journal of Pharmaceutical Research, Febraury 25, 2025.
3. Ivana Petzel, Sylvia I. Meier, stefan W. Toennes, Manfred Schubert- Zsilavecz. Detection of Lawsone (2-hydroxy-1,4-napthoquinone) in Henna treated hair. Forensic Science International, April 2019.
4. Nagwa S. El-Shaer, Jihar M. Badr,Maha A.Aboul-Ela, Yousry M. Gohar. Determination of Lawsone in henna powders by High Performance liquid chromatography. Journal of Separation Science, December 11, 2007.
5. Sahar Younes Ibrahim, Marwa M Fawzi, Mohammed Gader Saad M Abdel Rahman.Determination of Heavy Metals and Other Toxic Ingredients in Henna (Lawsonia inermis). Journal of Environmental and Analytical Toxicology, January 2016.
6. Ismail A. Ajaj, Wafa K. Amhimmid, Hala Ismail, Tahani Al-Arabi, Khawla Al-Oraibi. Determination of Heavy Metals in Henna Leaves and Cosmetic Henna Products Available in Zliten, Libiya.Al-Mukhkar Journal of Science, May 18, 2022.
7. Jayaganesh Sankar, mridula Kini, Sudhakar Mhaskar, Puja sangle. Estimation of Lawsone. Release from Lawsonia inermis Leaf Powder. Trends in Agricultural Sciences 2022.
8. Tsanuo Kassim Mohammed, Jaleny paul Ochieng. Levels of some Heavy Metals in the leaves of Henna Plant (Lawsonia inermis) with in Lamu county, Kenya. International Journal of Science and Healthcare Research, December 2020.
9. Shaziz Sulthana, muhammad Zafar.Authentication of Herbal Medicine Henna (Lawsonia inermis) by using Taxonomic and pharmacognostic Techniques. January 2013.
10. Muhammad Aslam, Sana Siddique, Muhammud Zaheer, Muhammud Zia-Ur-Rehman, Zahra Noreen, Yousuf Abbas, Aamir Sohail, Muhammad Aneeq Javed, Madiha Rashid, Aamir Ali. Isolation and Identification of Lawsonia Content from the Leaves of Henna (Lawsonia Inermis). Feburaury 7, 2025.

11. Jayaganesh Sankar, Mridula Kini, Puja Sangle. Extraction of Lawsone Active from the Henna Leaves (Lawsonia inermis) by solvent Extraction Method. Medicinal and Analytical Chemisrty International Journal. December 29, 2023.
12. Snehal S.Phrike, Moitreyee Saha. Determination of Lawsone by HPTLC in Lawsonia inermis L.Callus and Plantlets Regenerated invitro. Asian Journal of Chemistry, January 30, 2014.
13. Mauanang M. Patel, Bhavna R. Solanki, Nilesh C. Gurav, Prateek H.Patel,Shweta S. Verma. Method development for Lawsone estimation in Trihup herbal Hair powder by High performance thin layer chromatography. Journal of Advanced Pharmaceutical Technology and Research, September, 2013.