



iJRASET

International Journal For Research in
Applied Science and Engineering Technology



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Volume: 14 **Issue:** VIII **Month of publication:** August 2026

DOI: <https://doi.org/10.22214/ijraset.2026.84633>

www.ijraset.com

Call: ☎ 08813907089

E-mail ID: ijraset@gmail.com

Study of Harmful Chemical (para-phenylenediamine) Effects in Commercial Hair Dyes using TLC (Thin Layer Chromatography) and UV Visible Spectrophotometer: Current Issues and Future Directions (Hair Dyes)

Sadhna Gour¹, Dr. Mohammad Athar²

Institute for Excellence in Higher Education Bhopal, MP

Abstract: Hair dyes are really common, but they can have some not so great stuff in them. Things like phenylenediamine, Resorcinol, ammonia, and heavy metals can be bad for our health. So, we decided to take a closer look at what's actually in these hair dyes. We used a different methods to test them, like preliminary examination, chemical tests, and TLC. We also used UV visible spectrophotometry to get a better result. Our goal was to figure out if these hair dyes are really safe for us to use.

Commercial hair dyes sample 1a black hair dye, 1b brown hair dye, sample 2 Black, sample 3 brown, sample 4a brown, 4b black and sample 5a brown, 5b black were studied. Preliminary examination included Visible inspection, label verification, colour and consistency evaluation, odour assessment. Chemical test were conducted to detect ammonia, hydrogen peroxide, and PPD. Chromatography experiments allowed the separation of individual components, and RF values were determined and compared with scientific literature.

UV spectrophotometry further confirmed the presence of multiple chemical components. It turns out that hair dyes made from synthetic materials had Higher RF values, and that's because they contain less polar compound like PPD. On the other hand, natural henna based dyes had lower RF values due to polar components such as lawsone. When it comes to mixed formulations, they fell somewhere in between. What's really important to take away from this study is that even hair dyes that claim to be herbal can still contain harmful chemicals. So, it's crucial for consumers to be aware of what they are using and to carefully analyze the ingredients. This is not just about being mindful of what we put in our hair, but also about being informed about the potential risks associated with certain hair dyes. By being more aware and taking the time to read labels, we can make better choices for our health and well-being.

Keywords: Para-phenylenediamine, hair dyes, Thin layer chromatography, UV spectrophotometry.

I. INTRODUCTION

Hair dyes are cosmetic products widely used to change or enhance hair color. They work by depositing natural or synthetic color molecules on the hair surface or penetrating the hair shaft. Hair dyes may be natural, containing plant derived pigments like lawsone from henna, or synthetic, containing chemicals such as para phenylenediamine (PPD), resorcinol and hydrogen peroxide, ammonia and heavy metals.

Natural henna, derived from the leaves of the lawsonia inermis family Lythraceae plant, has been used for centuries for hair coloring. In addition to its coloring properties, henna is known for its conditioning effects, helping to strengthen hair and promote scalp health that is highly regarded for making it a popular choice for adornment women's bodies hair during celebration of the wedding and social events particularly in Indian subcontinent and in the Arab world.

The composition the active dye molecule the active in leaves of henna which give red color lawsone (2-hydroxy-2,4-naphthoquinone) Fig. 1a. para-phenylenediamine; its chemical formula ($C_6H_8N_2$) (fig. 1b.) This aromatic amine produced by many manufacturing companies, due to it is not found in nature. It's a derivative of aniline a white and water soluble solid, then due to air oxidation change darkens color.

A. Types of Hair dyes

- 1) Temporary Haicoat the hair surface and wash out quick.
- 2) Semi-permanent hair dyes - penetrate slightly into hair shaft and last for a few washes.
- 3) Permamnent hair dyes -Contain oxidizing agaents and ammonia , cause chemical changes in hair structure.
- 4) Natural hair dyes- Plant based , such as henna,considerd safer but sometimes mixed with synthetic additives.

Overall ,the sample include three major categories : Permanent synthetic dyes (Sample 1a,1b and 2) ,Semi-permanent dyes (Sample 3),mixed herbal formulations (Sample 4),and natural henna -based dyes (Sample 5). This classification helps in comparing their chmical behaviour using TLC,UV spectrophotometry and chemical tests.

B. History of Hair dyes

- 1) Ancient Egyptians used plant – based dyes especially henna, to color hair.
- 2) Romans experimented with metallic salts for hair Coloring.
- 3) Synthetic dyes, including aromatic amines were developed in the 19 century.th Today , commercial hair dyes are chemical advanced but may pose health risks if they contain harmful chemicals.

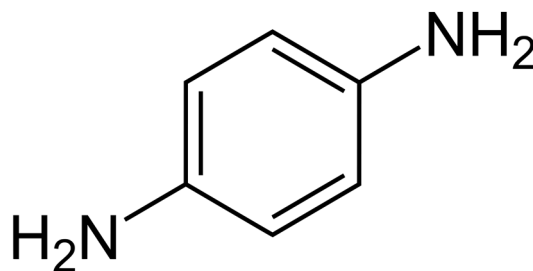


fig.1A structure of para- phenylenediamine

II. PROBLEM IDENTIFICATION AND PROPOSED WORK

A. Problem Identification

Commercially available hair dyes often contain a variety of synthetic chemicals that may not always be safe for long.fTerm used. In many cases, the information provided on product labels can be in complete, making it difficult for consumers to fully understand what they are applying to their skin and hair. The use of such products may pose several potential health risks, including skin irritation, allergic response, breast cancer and possible long term toxic effects due to repeated exposure to certain chemical compounds .

B. Proposed Work

To better understand the composition and safety of commercial hair dyes, this study propose a systematic chemical analysis of selected samples. The work will begin with preliminary and confirmatory chemical tests aimed at identifying potentially harmful substances present in the dyes. Further, chromatographic

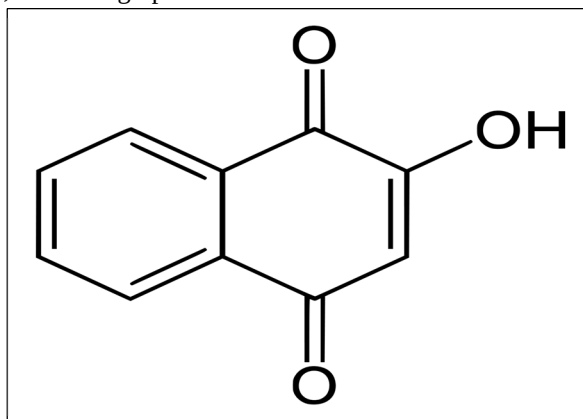


fig.1B Structure of lawsone

techniques such as thin layer chromatography will be employed to separate the components of the dyes. The experiment ally with standard literature values to help in identifying the compounds. In addition, UV spectrometry will be used to analyze the absorption behavior of the sample , providing further insight into their chemical composition and concentration of active constituents.

III. METHODOLOGY

A. Visual inspection: Each hair dye sample

was checked for packaging condition, label details (batch number, expiry date, safety warnings) and overall appearance including color and consistency everything is good .

B. Color and Consistency

Samples were examined for uniformity and checked for lumps , sediment, or any visible irregularities, we don't find anything suspicious .

C. Odor Evaluation

The samples were gently smelled to assess odor, with special attention to the presence of ammonia.

D. Solubility Test

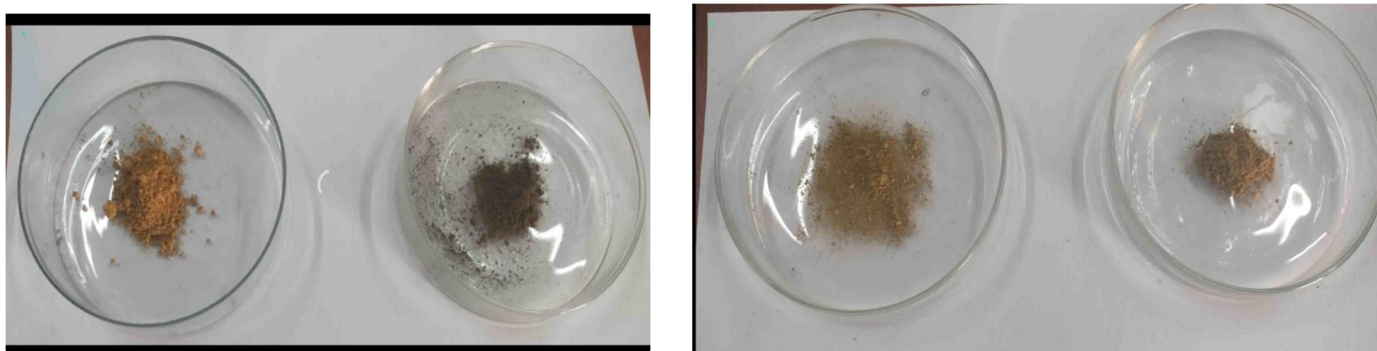
1g of hair dye was first mixed with 10ml of ethanol for the solubility test .At room temperature most sample didn't dissolve except the Sample 4A brown color dye .Which showed slightly solubility. The samples were then gently heated, and the temperature at which better solubility was observed was noted different temperature for different brand of dyes. For further analysis, each sample (hair dyes) was treated with water or dilute HCL, followed by filtration. The filtrate was used for preliminary testing. Ab oxidation test was performed by adding hydrogen peroxide (H_2O_2) , and a ferric chloride ($FeCl_3$) test was also carried out. The oxidation test gave a brown to black color, while the ferric chloride test showed a violet color . These results suggested the possible presence of PPD.

E. Stability Test

The stability of the sample was observed over 24-48 hours at room temperature under light and dark condition. Sample 1A(black), 2(black dye) ,3(brown dye) ,4B (black) ,5B(black) showed slightly color changes, becoming darker . In contrast, sample 1B (brown) , 4A(brown dye) and 5A(brown) remained stable with no change.

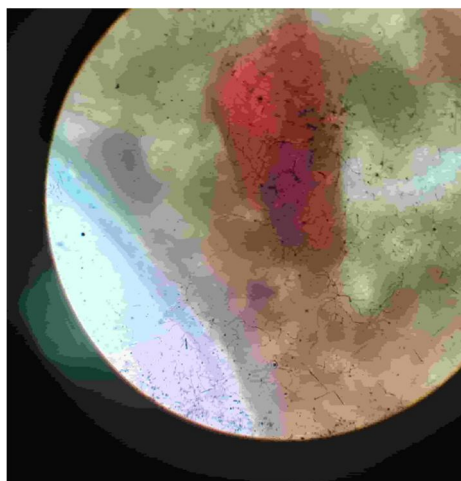
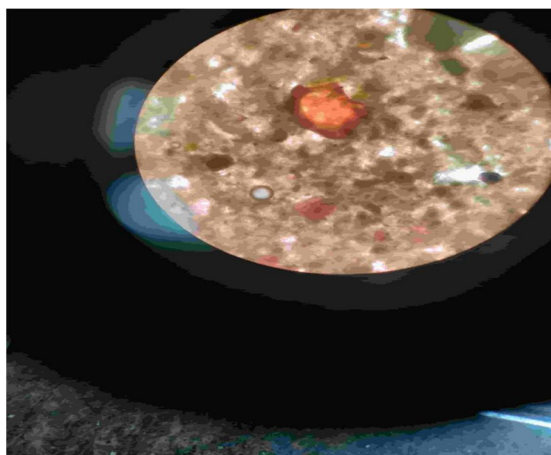
F. Microscopic Examination

A small amount of each sample was placed on glass slide pour few drops of glycerin on it then cover it with the help of coverslip and observed under a microscope. No foreign particles or impurities were founding any of the samples



Sample 4a, 4b

Fig A) visual examination of hair dye



Sample 5a, 5b

Fig B) Microscopic examination of hair dyes

Table1 Physical characteristics of hair dyes.

Sample No.	Colour	Consistency	odour
Sample 1a	Black	Smooth creamy	Chemical smell
Sample 1b	Brown	Smooth creamy	Slightly chemical smell
Sample 2	Black	Smooth	Cosmetic fragrance
Sample 3	Brown	Smooth	Mild cosmetic fragrance
Sample 4a	Brown	Smooth fine powder	Soft herbal fragrance or mild chemical
Sample 4b	Black	Slightly gritty powder	Soft herbal fragrance or mild chemical
Sample 5a	Brown	Rough fine powder	Herbal fragrance
Sample 5b	Black	Slightly gritty powder	Herbal fragrance

Table 1.1 physical characteristics (solubility) of hair dyes

Sample No.	Solubility	Temperature
Sample 1a	Insoluble in water Soluble on heating	48-51 °C
Sample 1b	Insoluble in water Soluble on heating	50-60 °C
Sample 2	Insoluble in water Soluble on heating	40-42 °C
Sample 3	Insoluble in water Soluble on heating	30-33 °C
Sample 4a	Partially soluble in water Completely soluble on heating	25 °C
Sample 4b	Insoluble in water Soluble on heating	32 °C
Sample 5a	Soluble in water	-
Sample 5b	Soluble in water	-

IV. THIN LAYER CHROMATOGRAPHY

Although the thin layers manufacturer was artificial silica gel G, the first step into thin plate was a clean glass plate. Then the plate was dried and activated by heating for a few minutes. After that, a baseline about 2cm from the bottom of the plate was marked by pencil. Beneath it, different positions to indicate where different samples would be loaded on subsequent operation were designated “ **Sample 1a, 1b, Sample 2, Sample 3, Sample 4a,4b, and Sample 5a, 5b**”. Next , use a capillary to drop a small amount of each hair dye concentrate at each marked position of the chromatogram. To make the solvent

Table 2. Detection of PPD by Thin layer chromatography

system consisting of” **Benzene: Acetone (8:2)**, ready and to pour this mixture into a Saturation chamber. This chamber was closed and left for 15-20 minutes until both sides accumulated enough solvent vapor inside. After saturation, the TLC plate was carefully placed inside th chamber, ensuring that the solvent level was below the baseline. The chamber was closed, and the solvent was allowed to rise up the plate. When the solvent front reached near the top of the plate, the plate was removed and the solvent front was immediately marked with a pencil. The plate was allowed to dry. After drying, the separated spots where observed under ultraviolet light. The RF values obtained were compared with standard PPD values to confirm its presence in the samples. The estimated PPD content varied across the different samples. Sample 1a showed $\pm 11.4\%$ PPD, while Sample 1b contained $\pm 11\%$, Sample 2 showed a higher concentration $\pm 20.4\%$,sample 3 contained $\pm 8.82\%$ PPD, whereas sample 4a and 4b showed relatively low values of $\pm 3.2\%$ and $\pm 2.9\%$, respectively .In contrast, PPD was not detect in both sample 5a and 5b . Using technical like TLC ,you can compare dyes from different brands. Each dye produces a unique pattern of spots called an RF value pattern. This makes it easy to tell whether 2 dyes are similar or different. Standard RF value PPD is 0.2 0.7 and it’s depends on solvent system.

Rf = Distance travelled by solute/ distance travelled by solvent front

Sample No.	Rf	PPD Content %	Observation
Sample 1a	0.52	114%	Present
Sample 1b	0.50	11%	Present
Sample 2	0.51	20.4%	Present
Sample 3	0.49	8.82%	Present
Sample 4a	0.32	3.2%	Present
Sample 4b	0.29	2.9%	Present
Sample 5a	Not detected	Not detected	Absent

Table 2. Detection of PPD by Thin layer chromatography

Sample No.	PPD Content %	Observation
Sample 1a	114%	Present
Sample 1b	11%	Present
Sample 2	20.4%	Present
Sample 3	8.82%	Present
Sample 4a	3.2%	Present
Sample 4b	2.9%	Present
Sample 5a	Not detected	Absent

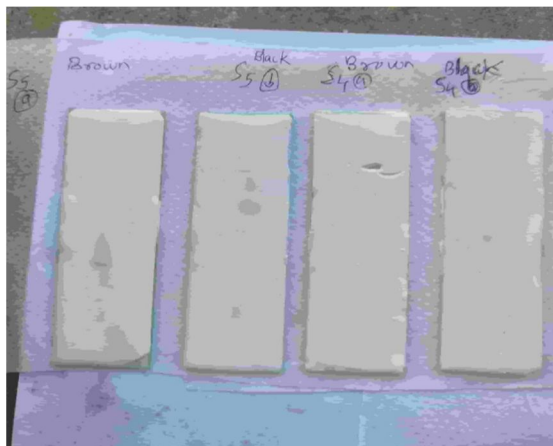


Fig 2.1 sample 4a, 4b and 5a, 5b

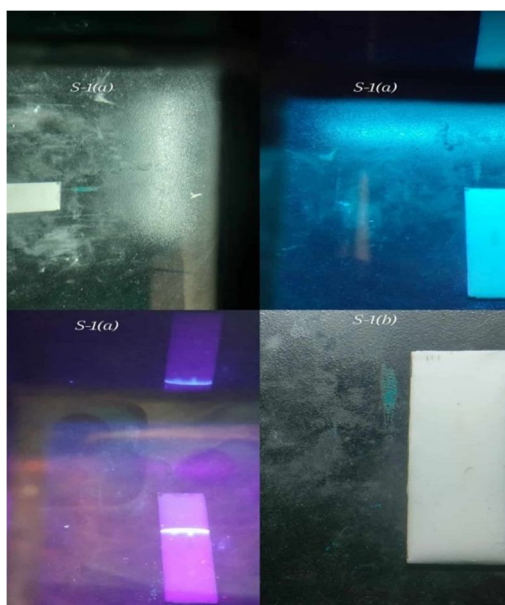


Fig 2.4 Sample 2,3,4a,4b and 5a,5b



Fig 2.1 Sample 1a,1b,2 and 3

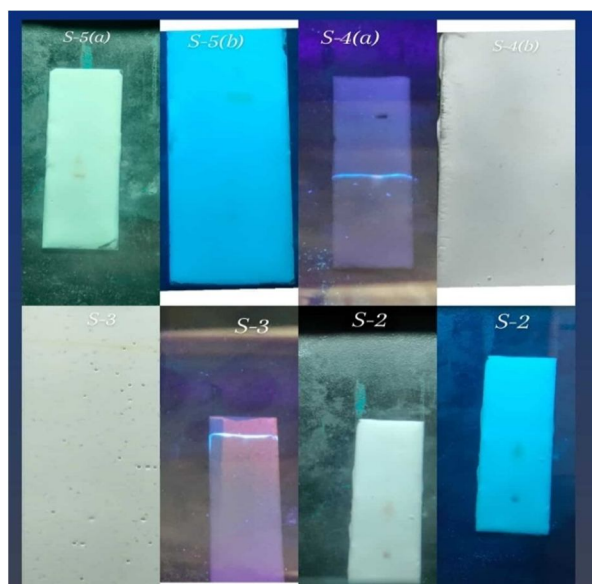


Fig 2.3 Sample 1a,1b under UV light fig

V. CHEMICAL DETECTION TEST

A. Detection of Ammonia

- 1) A small amount of the hair dye sample was taken in test tube, and its odour was carefully checked for the characteristics pungent smell of ammonia. Then, moist red litmus paper was held near the mouth of the test tube. Red litmus paper turned blue, it confirmed the presence of ammonia.



Fig i) litmus paper test for ammonia

- 2) Detection of p-phenylenediamine (PPD) A small amount of the hair dye extract was placed in a clean test tube. A few drops of hydrogen peroxide solution were then added. The mixture was gently shaken and observed carefully. Solution quickly turned dark black color, it indicated the presence of PPD in the sample.

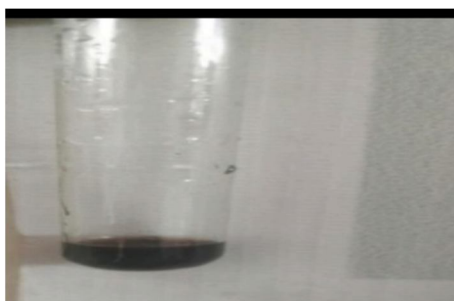


Fig ii) PPD test

B. Detection of hydrogen peroxide

Sample was taken in a test tube. A few drops of potassium iodide (KI) solution were added, followed by starch solution. Appearance of a blue colour confirmed the presence of hydrogen peroxide. Observations: Sample 1a,1b,2,3,4a,and 4b tested positive for the presence of ammonia ,PPD and hydrogen peroxide. However, samples 5a and 5b didn't show the presence of these chemicals and were considered free from them.

VI. CONFORMATORY TEST AND DATA ANALYSIS

A. UV spectrophotometric Analysis

UV spectrophotometry analytical determine technique the is used absorption ultraviolet light by an to of chemical substances present in a sample. Compounds such as p phenylenediamine and other dye intermediates absorb UV radiation in the range of 200-300 nm due to electronic transition within their molecular structure. The wavelength at which maximum absorption occurs is characteristic of specific compounds and can be used for their identification. UV spectrophotometry is used to study the absorption of ultraviolet light by different chemical compounds. Compounds such as para-phenylenediamine (PPD) and related aromatic dye intermediates show characteristics absorption in the range of 240-300 nm. In this study, ethanol was used as the solvent because all hair dye samples were extracted in Ethanol (C₂H₅OH), ensuring complete solubility and consistent spectral behavior. Standard PPD solutions (2-10µg/mL) were prepared in ethanol to construct a calibration curve, and ethanol was used the blank for instrument calibration. The coded samples were dissolved, filtered, and analyzed using a UV spectrophotometer. The results showed that sample 1 and sample 2(permanent dyes) exhibited strong absorbance peaks in the 240- 260 nm region, including a higher concentration of PPD or similar aromatic amine. Sample 3(brown shampoo based dye) showed moderate absorbance, suggesting the presence of lower concentration of synthetic colorants typical of Semi-permanent formulations. Sample 4(herbal labelled product) showed weak absorbance, indicating possible trace synthetic additives. Sample 5(pure henna based product) showed negligible absorbance in this region.

Table 3. Detection of PPD by UV spectrophotometry

Sample No.	UV absorption range(nm)	Observation
Sample 1a	280-285 nm	Trace PPD
Sample 1b	282-290 nm	Moderate
Sample 2	285-295 nm	High
Sample 3	282-288 nm	Moderate
Sample 4a	280-286 nm	Low presence
Sample 4b	280-285 nm	Low presence
Sample 5a	No peak	PPD Absent
Sample 5b	No peak	PPD Absent

VII. RESULT AND DISCUSSION

In this study, different analytical techniques like chemical test ,TLC , UV spectrophotometry were performed to detect thepresence of harmful chemicals in dyes sample Out of 8 samples analyzed, 6 samples were found to contain chemicals components such as ammonia, PPD, and hydrogen peroxide. Only 2 samples (5a, 5b) were free from these chemicals and appeared to formulations. The TLC be and natural UV spectrophotometric results further confirmed the variation in PPD concentration among brands. different

VIII. CONCLUSION AND FUTURE WORK

A. Future Work

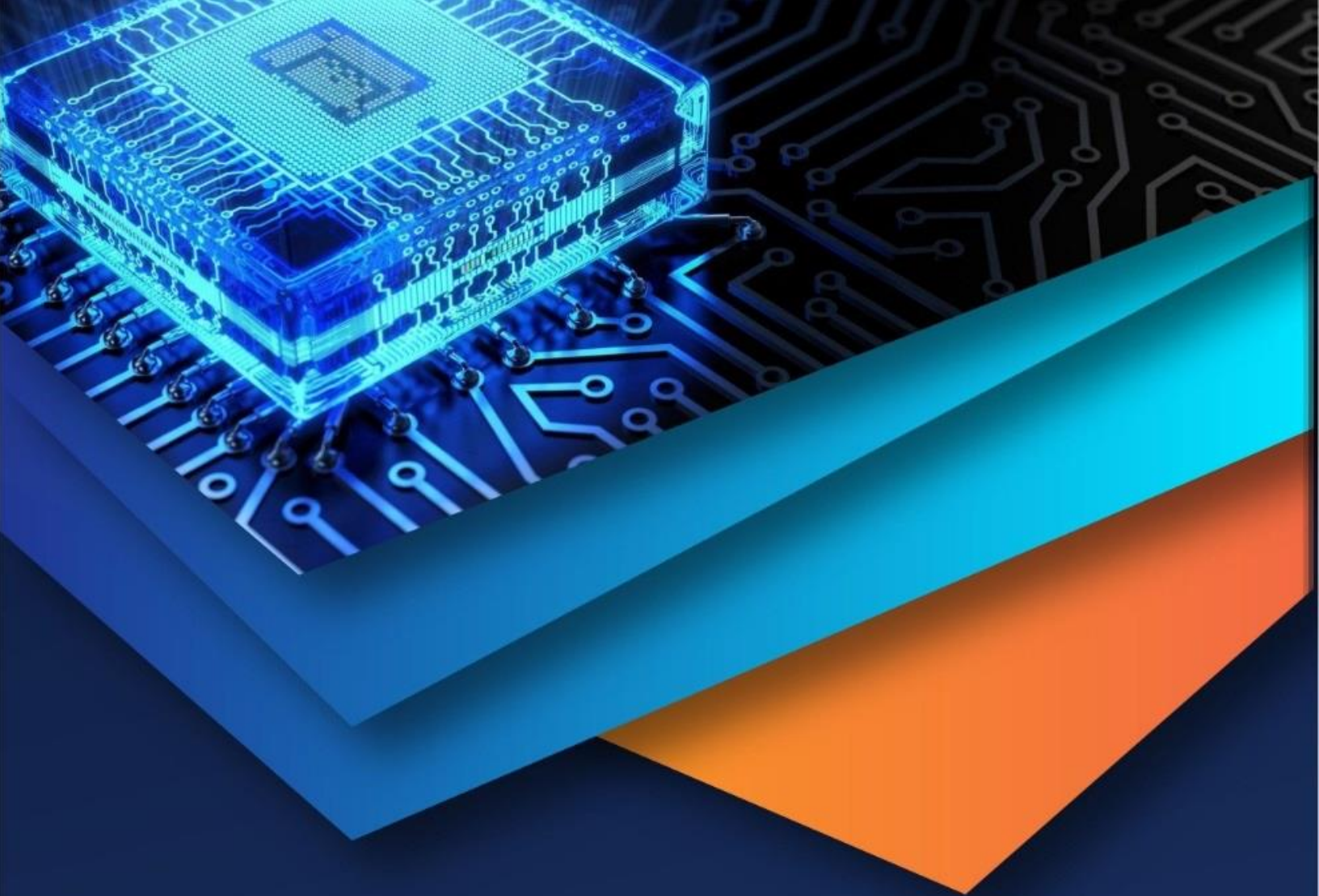
- 1) Development of Simple and Rapid Detection Methods In the future, this study can be extended to develop simple, quick, and user-friendly methods for detecting harmful chemicals such as Para phenylenediamine in commercial hair dyes. These methods should aim to provide instant results without requiring complex procedures.
- 2) Design of Paper-Based Detection Systems One possible advancement is the development of paper strip tests, similar to litmus paper, that can instantly indicate the presence of Para-phenylenediamine. Such low cost and easy-to-use tools would be highly beneficial for quick screening and everyday use.

- 3) Development Detection Kits of Portable Future research can focus on creating portable detection kits that are compact, easy to carry, and simple to operate. These kits could allow consumers to test hair dye products at home without needing specialized knowledge or equipment
- 4) Advancement in Electronic Detection Devices Advanced studies may lead to the development of small electronic devices, similar to alcohol detectors, that can quickly identify harmful chemicals like Para- phenylenediamine in cosmetic products. Such devices would provide fast and reliable results with improved accuracy.
- 5) Improvement in Consumer Awareness and Safety These innovations would significantly enhance consumer awareness by enabling individuals to test products before use. This would help reduce health risks associated with exposure to harmful chemicals and promote safer use of cosmetic products.
- 6) Expansion for Broader Cosmetic Analysis In the future, these detection methods can also be extended to identify other harmful substances present in cosmetics, making the approach more comprehensive and useful for overall product safety evaluation This study evaluates the presence of harmful chemicals in commercial hair dyes, with special focus on Para phenylenediamine using analytical techniques such as Thin Layer Chromatography and UV-Visible Spectrophotometry. The results show that these methods are effective for identifying and estimating the presence of PPD in various hair dye samples. They are also cost effective and easily accessible, making them suitable for routine laboratory analysis. contain The study highlights that most commercially available hair dyes harmful chemicals such as Para phenylenediamine, ammonia, and hydrogen peroxide, which may pose health risks to users. Only a few products, such as pure henna-based dyes, are free from these toxic substances and are comparatively safer for use. However, certain limitations were observed, including interference from other ingredients present in hair dye formulations, limited sensitivity of conventional techniques, and challenges in achieving high specificity. These issues indicate the need for improved analytical methods and better detection approaches.

Overall, this study provides a strong analytical foundation for detecting harmful chemicals in hair dyes and emphasizes the importance of consumer awareness and product safety. Future advancements in analytical techniques and portable detection methods can further improve accuracy, sensitivity, and real-time analysis, ensuring safer use of cosmetic products

REFERENCES

- (11) Jacob, S. E., Zapolanski, T., (1) Horev, L. Exogenous Factors in Hair Disorders. *Exogenous Dermatology* 2006, 3 (5), 237–245. (2) Guerra-Tapia, A.; Gonzalez-Guerra, E. Hair Cosmetics: Dyes. *Actas Dermosifiliogr* 2014, 105 (9), 833–839. (3) Shiel, S. Hair Health and Management of Common Hair Disorders. *J. Cosmet Dermatol* 2007, 6 (SUPPL. 1), 12–17. (4) Trüeb, R.; Hoffmann, R. Aging of Hair. *J. Cosmet Dermatol* 2005, 4 (2), 60–72. (5) Bolduc, C.; Shapiro, J. Hair Care Products: Waving, Straightening, Conditioning, and Coloring. *Clin Dermatol* 2001, 19 (4), 431–436. (6) Sinclair, R. D. Healthy Hair: What Is It? *Journal of Investigative Dermatology Symposium Proceedings* 2007, 12 (2), 2–5. (7) Oliveira, R. A. G. d.; Zanon, T. B.; Bessegato, G. G.; Oliveira, D. P.; Umbuzeiro, G. A.; Zanon, M. V. B. The Chemistry and Toxicity of Hair Dyes. *Quim. Nova* 2014, 37 (6), 1037–1046. (8) Nohynek, G. J.; Skare, J. A.; Meuling, W. J. A.; Wehmeyer, K. R.; de Bie, A. T. H. J.; Vaes, W. H. J.; Dufour, E. K.; Fautz, R.; Steiling, W.; Bramante, M.; Toutain, H. Human Systemic Exposure to [14C] Paraphenylenediamine-Containing Oxidative Hair Dyes: Absorption, Kinetics, Metabolism, Excretion and Safety Assessment. *Food Chem. Toxicol.* 2015, 81, 71–80. (9) Almeida, P. J., Borrego, L., Pulido-Melián, E., & González-Díaz, O. (2012). Quantification of p-phenylenediamine and 2-hydroxy-1,4 naphthoquinone in henna tattoos. *Contact Dermatitis*, 66(1), 33–37.
- (10) Brancaccio, R. R., Brown, L. H., Chang, Y. T., Fogelman, J. P., Mafong, E. A., & Cohen, D. E. (2002). Identification and quantification of para-phenylenediamine in a temporary black henna tattoo. *American Journal of Contact Dermatology*, 13(1), 15–18. Chayavichitsilp, P., Connelly, E. A., & Eichenfield, L. F. (2008). P Phenylenediamine in black henna tattoos: A practice in need of policy in children. *Archives of Pediatrics & Adolescent Medicine*, 162(8), 790–792.
- (12) Kang, I.-J., & Lee, M.-H. (2006). Quantification of para-phenylenediamine and heavy metals in henna dye. *Contact Dermatitis*, 55(1), 26–29. (13) Pretzel-Witt, S., Meier, S. I., Schubert-Zsilavecz, M., & Toennes, S. W. (2019). Detection of lawsone (2-hydroxy 1,4-naphthoquinone) in henna-treated hair. *Forensic Science International*, 297, 184–188. (14) Sienkiewicz-Szlapka, E., Janysz, A., & Wróbel, M. H. (2025). Quantification of lawsone and p-phenylenediamine in natural henna products for hair colouring available in Poland. *Contact Dermatitis*, 92(4), 261–272. V. (15) Singh, M., Bhardiya, S. R., Rai, A., & Rai, K. approach (2022). Electrochemical for recognition and quantification of p-phenylenediamine: A review. *Sensors & Diagnostics*, 1, 376–386 Such innovations would help increase consumer awareness and safety by allowing people to test products before use.
10. Bibliography • Almeida, P. J., Borrego, L., Pulido-Melián, E., & González-Díaz, O. (2012). Quantification of p phenylenediamine and 2-hydroxy-1,4 naphthoquinone in henna tattoos. *Contact Dermatitis*, 66(1), 33–37. <https://pmc.ncbi.nlm.nih.gov/articles/PMC1226873/2/pdf/ao5c01576> • Kang, I.-J., & Lee, M.-H. (2006). Quantification of para-phenylenediamine and heavy metals in henna dye. *Contact Dermatitis*, 55(1), 26–29. <https://doi.org/10.1111/j.0105-1873.2006.00845> • Brancaccio, R. R., Brown, L. H., Chang, Y. T., Fogelman, J. P., Mafong, E. A., & Cohen, D. E. (2002). Identification and quantification of para phenylenediamine in a temporary black henna tattoo. *American Journal of Contact Dermatology*, 13(1), 15–18. <https://doi.org/10.1053/ajcd.2002.30466> • Pretzel-Witt, S., Meier, S. I., Schubert-Zsilavecz, M., & Toennes, S. W. (2019). Detection of lawsone (2-hydroxy-1,4-naphthoquinone) in henna-treated hair. *Forensic Science International*, 297, 184–188. <https://doi.org/10.1016/j.forsciint.2019.01.0377> • Singh, M., Bhardiya, S. R., Rai, A., & Rai, V. K. (2022). Electrochemical approach for recognition and quantification of p-phenylenediamine: A review. *Sensors & Diagnostics*, 1, 376–386. <https://doi.org/10.1039/D1SD00070E> • Determination of p-phenylenediamine and its metabolites MAPPD and DAPPD in biological samples using HPLC-DAD and amperometric detection – ScienceDirect <https://share.google/pCf3ymEBsIusIn9Sn> • Jacob, S. E., Zapolanski, T., Chayavichitsilp, P., Connelly, E. A., & Eichenfield, L. F. (2008). P Phenylenediamine in black henna tattoos: A practice in need of policy in children. *Archives of Pediatrics & Adolescent Medicine*, 162(8), 790–792. <https://doi.org/10.1001/archpedi.162.8.790> • Sienkiewicz-Szlapka, E., Janysz, A., & Wróbel, M. H. (2025). Quantification of lawsone and p phenylenediamine in natural henna products for hair colouring available in Poland. *Contact Dermatitis*, 92(4), 261–272. <https://doi.org/10.1111/cod.14739>



10.22214/IJRASET



45.98



IMPACT FACTOR:
7.129



IMPACT FACTOR:
7.429



INTERNATIONAL JOURNAL FOR RESEARCH

IN APPLIED SCIENCE & ENGINEERING TECHNOLOGY

Call : 08813907089  (24*7 Support on Whatsapp)