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UV-Visible Spectrophotometric Method Development for p-Phenylenediamine Determination in Marketed Hair Dyes

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Abstract

Aim: To develop two simple, sensitive, rapid, robust and reproducible Spectrophotometric methods for determination of Para-phenylenediamine (PPD) in marketed Hair dyes. **Materials and methods:** Solvent used is 0.1N NaOH. Folin's reagent and ninhydrin reagent (were prepared in methanol) is used for analysis of P-phenylenediamine in marketed hair dyes by UV-Visible spectrophotometer at 453nm and 431nm respectively. **Result & Conclusion:** Regression coefficient values for both the methods were found to be within limits i.e., $r^2 \geq 2$. Paraphenylenediamine (PPD) concentration in all the tested brands of hair dyes were found to be within the limits.

Keywords
Folin's reagent, Ninhydrin reagent, p-Phenylenediamine, Spectrophotometric method.

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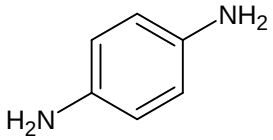
INTRODUCTION:

P-Phenylenediamine (PPD) (Figure 1) is a monocyclic aryl amine compound; the chemical system of PPD is C6H8N2 and 108.15 g is its molecular weight. It is powder shape that is white to light red which receives oxidized turning first purple, then brown then subsequently black when exposed to air. It is used as a hair colouring product at a maximum of 4%. Not only in hair dyes, is PPD also present in rubber compounds, textile dyes. People operating in organizations may be occupationally uncovered to PPD at some point of its manufacture or use, and the exposure may also occur through skin, eye contact, inhalation and ingestion. [1]Long-term exposure may lead to Eczematous touch dermatitis (chronic effect) in human beings. [2]



p-phenylenediamine

Currently, most of hair dye formulations advertised all around the globe will include PPD. [3, 4, 5] Analytical methods are evolved for the determination of PPD by HPLC GC/MS, volt metric technique, emission spectroscopy. These Spectrophotometric techniques have their advantages but the methods are time consuming. The present work was aimed to develop an easy, speedy, simple Spectrophotometric method for the estimation of PPD in different brands of hair dyes through using Folin's reagent and Ninhydrin reagent. These reagents are very financial while in comparison to other reagents and usage of those reagents is very common in laboratory. [6, 7, 8]

Folin's reagent which is Sodium 1, 2-naphthoquinone-4-sulfonate is used to estimate the amines and amino acids. [5]A bright red colour in alkaline solutions is produced by using this reagent and is also fluorescent [9, 10, 11]. The disadvantage of this procedure is it produces colour in slightly alkaline solution room temperature. Large number of substances of pharmaceutical interest and many amino compounds scan be determined by using Folin's reagent. [12, 13, 14, 15] The mechanism of reaction between PPD and folin's reagent is shown in Figure 2.

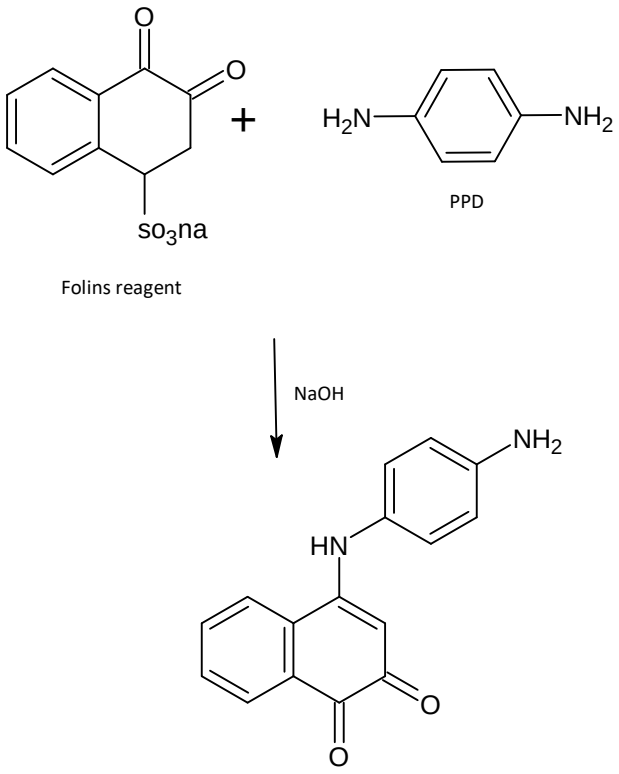


Figure 2. Mechanism of Folins reagent with PPD (Method A)

One of the important reagents for detecting amino acids, both technically and historically is ninhydrin reagent, which has been conventionally used to detect their microgram amounts [15, 16]. A purple-coloured product is produced when amino acids with a free alpha amino group are treated with an excess of ninhydrin. The colour formed is directly proportional to the amino acid present, under appropriate situations. After separation of amino acids through chromatography the remaining amino acids are quantified by colorimeter. [17] The mechanism of reaction between PPD and ninhydrin reagent is shown in Figure 3[18]

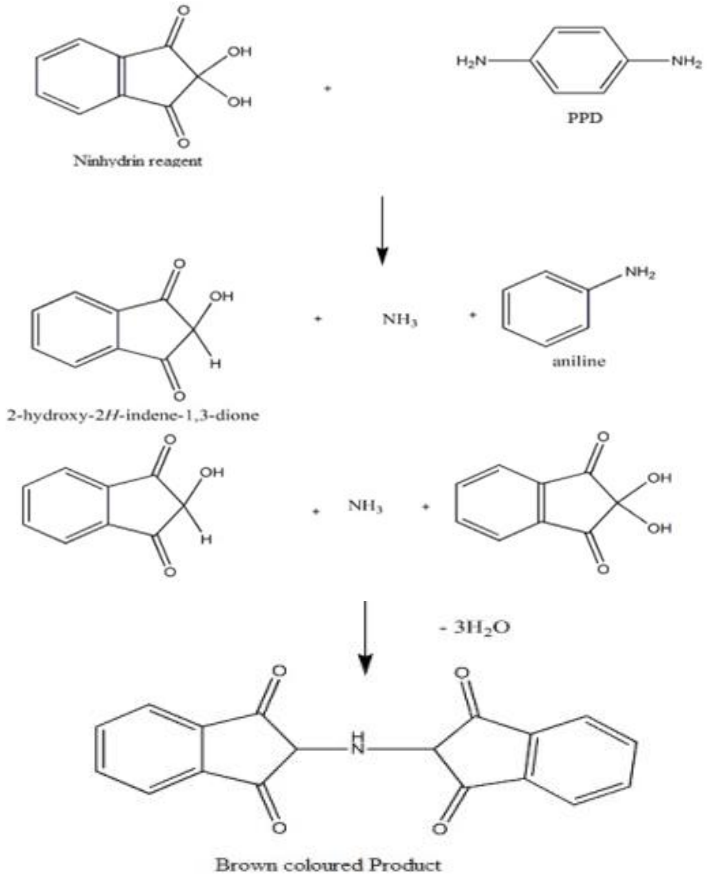


Figure 3. Mechanism of ninhydrin with PPD (Method B)

MATERIALS AND METHODS:

Instrument:

Double Beam UV-Visible Spectrophotometer "ElicoSl 210" were used for analysis.

Chemicals and Reagents

3 different hair dyes containing PPD were purchased from local market, standard PPD was obtained as a gift from Pharma company, Ninhyrin reagent, folin's reagent and acetone used were of analytical grade. All the other chemicals were of analytical grade.

METHOD:

Preparation of stock solutions

Weigh accurately about 100mg of standard PPD and then dissolve in 100ml of 0.1N NaOH solution to make the solution of 1mg/ml concentration. From this solution, serial dilutions were made to obtain 100 µg/mL and 10 µg/mL for method development.

Preparation of sample solution

1g of different hair dye products were separately weighed and dissolved in 25 mL of 0.1 N NaOH solutions. Whitman filter paper is used to filter the solution from these working samples concentration was prepared by using two different methods (method-A & method-B) using 0.1 N NaOH solution.

Method A

In this method 2ml of the sample solution were pipette and were taken in three different test tubes, add1.0 mL of Folin's reagent and 1mL sodium hydroxide in all three test tubes. The absorbance is checked at 453nm after making the volume up to 10ml with water and water is used as blank.

Method B

From above solution 2ml was taken in three different test tubes and then add 1.0 mL of ninhydrin reagent and 1 mL sodium hydroxide in all the three test tubes. The absorbance is checked at 431nm after making the volume up to 10ml with water and water is used as blank.

RESULTS& DISCUSSION:

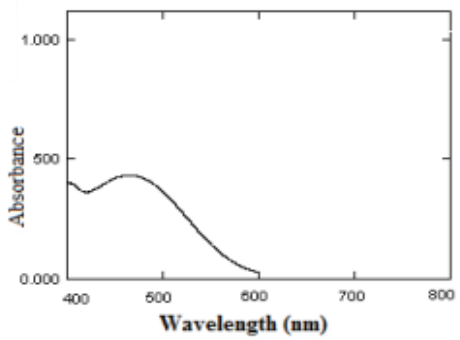


Figure 4. Absorption spectra of PPD with Folin's reagent

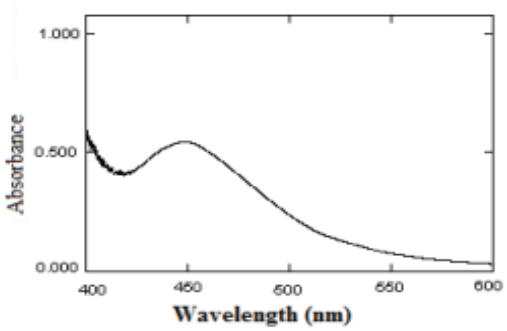


Figure 5. Absorption spectra of PPD with ninhydrin reagent

Table1: Optical parameters for Method A and Method B

Parameters obtained	Obtained values	
	Method A	**Method B**
λ_{max}	453 nm	431nm
Linearity (µg/mL)	2 - 12	0.1 - 0.6
Slope ± SEM	0.0721± 0.003	1.5227 ± 0.01
Intercept ± SEM	0.0487 ± 0.004	0.0276 ± 0.003
Regression coefficient	0.9919	0.9954
Wavelength	453 nm	431 nm
LOD (µg/mL)	0.0091	0.0019
LOQ (µg/mL)	0.0277	0.0059

Table 2. P-phenylenediamine (PPD) concentration present in different hair dyes by using method-A and method-B is shown below

Brands	Absorbance at 453nm (METHOD-A)	Concentrations(nm) (METHOD-A)	Absorbance at 453nm (METHOD-A)	Concentrations(nm) (METHOD-B)
Brands-A	0.143	1.319	0.1109	0.055
Brands-B	0.4376	5.411	0.1199	0.0612
Brands-C	0.3827	4.648	0.848	0.038

CONCLUSION:

Estimation of PPD in three different hair dyes by Spectrophotometric method was performed successfully. The method was found to be simple and easy for estimation of PPD in hair dyes. All the brands which are tested contain p-phenylenediamine (PPD) and their concentrations were found to be within limits.

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REFERENCES:

[1] https://en.wikipedia.org/wiki/P-Phenylenediamine
[2] https://dermnetnz.org/topics/allergy-to-paraphenylenediamine#:~:text=Paraphenylenediamine%20(PPD)%20is%20a%20chemical,permed%20without%20losing%20its%20colour.
[3] Ayesha Al-Suwaidi 1 and Hafiz Ahmed 2, Determination of para-Phenylenediamine (PPD) in Henna in the United Arab Emirates, Int. J. Environ. Res. Public Health 2010, 7, 1681-1693; doi:10.3390/ijerph7041681

[4] https://onlinelibrary.wiley.com/doi/10.1002/14356007.a12_571.pub2
[5] https://www.cdc.gov/niosh/idlh/106503.html
[6] Department of Health and Human Services Hazardous Substances Data Bank (HSDB, online database). National Toxicology Information Program, National Library of Medicine. Bethesda, MD, USA, 1993.
[7] Stanley LA, Skare JA, Doyle E, Powrie R, D'Angelo D, Elcombe CR, Lack of evidences for metabolism of Phenylenediamine by human hepatic cytochrome P450 enzymes, J Toxicol 210(2- 3), 147-157, 2005.
[8] Kind F, Scherer K, Bircher AJ, Contact dermatitis to para-phenylenediamine in hair dye following sensitization to black henna tattoos-an ongoing problem, J Dtsch Dermatol Ges 10(8), 572-578, 2012.
[9] https://www.cdc.gov/niosh/idlh/106503.html
[10] Nafisur Rahman, Mohammad Kashif, Application of ninhydrin to Spectrophotometric determination of famotidine in drug formulations, Il Farmaco, 58(10), 1045–1050, 2003.
[11] Stambouli A, Bellimam MA, El Karni N, Bouayoun T, El Bouri A, Optimization of an analytical method for detecting paraphenylenediamine (PPD) by GC/ MSiontrap in biological liquids, Forensic Sci Int 146, S87-S92, 2004.
[12] Inzhang Gao; Xiaodong Chen Jie Ren; Wu Yang, Determination of p-phenylenediamine by perturbation of a non-equilibrium stationary state in the B-Z reaction, Journal of the Brazilian Chemical Society, J Braz Chem Soc 22 (4), 648-651, 2011
[13] KessarinNgamdeeSurangkhanaMartwisetThawatchai TuntulaniWittayaNgeontae, Selective fluorescence sensors for Phenylenediamine using formyl boronate ester with an assistance of micelles, Sensors & Actuators: B. Chemical, 173, 682-691, 2012.
[14] Nitin S Jadhav, Muthu Lakshmi M, Meena S, Duraivel S, validated visible Spectrophotometric estimation of paraphenylenediamine, a carcinogenic ingredient in henna hair dyes, Int J Pharm Technol 2(4), 900-906, 2010.
[15] Hilton CL, Spectrophotometric determination of flexzone 3C and other p-phenylenediamine derivatives, Anal Chem 32 (12), 1554–1557, 1960.
[16] Yoshiaki Ikarashi, Masa-akiKaniwa, Determination of p-phenylenediamine and related antioxidants in rubber boots by high performance liquid chromatography. Development of an analytical method for N- (1-Methylheptyl)-N'-phenyl-pphenyenediamline. J Health Science 46(6), 467 - 473, 2000.
[17] Ursula Vincent, Guy Bordin, and Adela R. Rodrigue, Validation of an analytical procedure for the determination of oxidative hair dyes in cosmetic formulations, J Cosmet Sci 53, 43-58, 2002.
[18] Maria Luisa Di Gioiaa, Antonella Leggioa, Adolfo Le Peraa, Angelo Liguoria, Determination by gas chromatography/mass spectrometry of p-phenylenediamine in hair dyes after conversion to an imine derivative, J Chromatogr A 1066 (1–2), 143–148, 2005.