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A selective quantitative assay of p-phenylenediamine in henna and hair dyes

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ABSTRACT

P-phenylenediamine (PPD) is an aniline derivative with crystalline appearance easily oxidized to provide a darkness color, therefore used as a color developer or as permanent color in hair dye. PPD has been determined in henna and hair dyes of black, brown, grey, and blonde color of different origins. The aim of this study is to know the concentrations of this substance in hair dyes and compare them with each other. the principle of oxidative-coupling criteria has been followed by the oxidation a PPD in aqueous medium then coupling with 2,7-DHN in basic medium to form a stable complex measured at 582nm. Beer's law was obeyed from 1.02 to38 µg/mL, The method has high sensitivity and low limits of detection and quantitation (0.36771x104 L.mol-1.cm-1, 0.052 µg/mL and 0.176 µg/mL respectively). Henna contains 0.625 mg PPD /0.2 g which consider the highest value, while hair dyes contain from 0.0525 to 0.3075 mg PPD /0.2 g with an average recovery 99% and average standard deviations of 1.198%.

التقدير الطيفي للبارافينيلين ثنائي الامين في الحنة وصبغات الشعر

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الخلاصة:

بارا فينيلين ثنائي الامين من مشتقات الانيلين يظهر بشكل بلورات سهلة التأكسد الى لون غامق بتأثير الهواء لذلك تم استخدامه بكثرة في منتجات صبغات الشعر. تم تقدير بارا فينيلين ثنائي الامين في الحنة وصبغات الشعر السوداء، البنية والرصاصية، والشقراء في انواع مختلفة من الصبغات المتداولة في الاسواق. والهدف من هذه الدراسة هي معرفة تركيز هذه المادة في صبغات الشعر المتوفرة في الاسواق المحلية ومقارنتها مع بعضها. وهذه الطريقة اعتمدت على اكسدة بارافينيلين ثنائي الامين في الوسط المائي ثم اقترانه مع الكاشف 2,7−DHN في الوسط القاعدي وتكونت صبغة مستقرة ذات لون ازرق ذائبة في الماء تعطي اعلى امتصاص عند 582 نانومتر. واعطت الطريقة خطية من 1.02الى 38 ميكروغرام لكل

مللتر ومعامل امتصاص مولاري 0.36771×10^4لتر. مول$^{-1}$. سم$^{-1}$ وLOD وLOQ 0.025 و0.167 ميكروغرام لكل مللتر على التوالي. واظهرت الطريقة ان الحنة تحوي على اعلى تركيز من بين المنتجات التي تم تطبيق الطريقة عليها حيث احتوت على 0.625 ملغم من PPD لكل 0.2 غرام بينما احتوت صبغات الشعر على تركيز 0.0525 الى 0.3075 ملغم من PPD لكل 0.2 غرام وكانت نسبة الاسترجاع 99% وبمعدل انحراف معياري1.198%.

الكلمات المفتاحية: بارافينيلين ثنائي الامين، حنة، صبغات الشعر، تقدير طيف

1- NTRODUCTION

Paraphenylenediamine (PPD) is an organic compound with a chemical formula of $C_6H_4(NH_2)_2$, and molecular weight 108.15g/mole It's openly used in hair dye. (PPD) is an aniline derivative with crystalline appearance easily oxidized to provide a darkness color by the effect of air, therefore used as a color developer or as permanent color in hair dye. It is used as a component of oxidative hair coloring products at a maximum concentration of 4.0%. PPD also can be used in wool or knitting dyes, photo graphic developing agent and as an antioxidant in rubber compounds. People may be position to PPD during its industrialization or use, and the position of PPD may occur through inhalation, skin and eye contact, and ingestion [1-2]. Temporary position to high levels of PPD (severe effects) may cause strong dermatitis, eye irritation and tearing, asthma, gastritis, renal failure, vertigo, tremors, convulsions and coma in humans. Eczematous contact dermatitis may result from long-term exposure in humans [3-4]. Currently, PPD is existing in more than 1000 hair dye pointing marketed all over the world [5].

Some analytical methods are developed for the determination of PPD; these in include: HPLC [10-12], GC/MS [6,7], voltametric method [8], emission spectroscopy in addition to spectrophotometric method [9], involving the time-consuming method diazotization and coupling with N-(1-naphthyl)ethylenediamine [10], oxidation of the compound to convert in to salt measured calorimetrically [10], coupling with reagent 2-aminonaphthalene-4,8-disulfonic acid with low level detection [11]. Another method was based on the use of PPD in the reaction of sodium nitrite with p-sulfanilic acid in an acidic medium to form the azo compound in an alkaline medium [12], also a method of first-order derivative spectrophotometry was developed [13].

In this study, PPD was determined by a proposed simple, sensitive, rapid, reproducible, precise and accurate spectrophotometric analysis depending on the oxidation a PPD in aqueous medium then coupling with 2,7-DHN in basic medium to form blue product solution measured in 582nm. followed by determination of PPD in henna and hair-dyes.The aim of this study is to know the concentrations of this substance in hair dyes and compare them with each other

2-Material and methods

2.1. Instrument:

-Shimadzu UV-VIS 1900i spectrophotometer - double beam ,1cm matched glass cells, Sartorius BL 20 S, Germany Balance.

2-2 Materials:

All chemical ‘materials used were of analytical grade.

p-phenylene diamine (PPD) (BDH), 2,7dihydroxynaphthalene (2,7DHN) (BDH), Ethanol (ABS).

2-3 Prepared solutions:

2.3.1. PPD solution (1×10^{-3}) This solution was prepared by dissolving 0.0108 g of pure reagent PPD in 6 ml of ethanol and dilute with distilled water in a 100 mL calibrated flask.

2.3.2. 2,7DHN solution (1×10^{-3}) was prepared by dissolving 0.0161 g of reagent in 3ml ethanol and then diluting with distilled water in a 100 mL calibrated flask.

2.3.3. $NaIO_4$ (0.2٪) solution was prepared by dissolving 0.2g of pure solid in 100 ml of distilled water.

2.3.4. Preparation of hair dye and henna solution

2.3.4.1. 2 g of hair dye was weighed and dissolved in 25 ml of 0.1 M NaOH solution, then the solution was filtered using filter paper. filtered solution diluted to 50 ml with distilled water in a volumetric flask [1].

2.3.4.2. 2 g of henna powder was weighed and dissolved in 25 ml of distilled water, then solution was filtered using filter paper, and the filtered solution was diluted to 50 ml with distilled water in a volumetric flask.

3. Results and Discussion

When 2ml of ($NaIO_4$ 0.2%) was added to 1ml of (PPD) and waited for 3 min, followed by 2ml of (2,7-DHN), 0.5 of (NaOH 1M) and distilled water to make exactly 10 ml, a blue color is appeared against colorless blank solution prepared by the same way. the absorption spectrum shows a maximum response at 582 nm.

3.1. Select the conditions of the reaction:

The type and the amount of four bases on the sensitivity of the reaction have been studied,1 mL of 1M NaOH is the best medium for the oxidative coupling step. Figer 1 shows the effect of bases, and Figer 2 shows the effect of amount of the NaOH. The effect of 1 to 3 mL of the oxidant $NaIO_4$ (0.2%) has been checked, Table 1 shows that 1 mL is the best when making the measurements. The effect of 0.5 to 2.5 mL of 2,7DHN (1×10^{-3}) has been checked too, Table 2 shows that 2

mL is the best when making the measurements. Figure 3 shows that 10 minutes is the best time for the oxidation. Stability study in Table 3 shows that the reaction is stable for an hour. Table 4 show no effect of the surfactants on the absorbance of the reaction mixture.

3.2. Absorption spectrum and calibration curve

The absorption spectrum shows a maximum response at 582 nm. under the selected criteria conditions: (1.4-48 ppm) of PPD, 1 ml of PPD (1×10^{-3}M), 1 mL of $NaIO_4$ (0.2%),2ml of 2,7-DHN (1×10^{-3}M), 1 ml of NaOH (1 M) and distilled water to make exactly 10 ml, a blue color is appeared against colorless blank solution prepared by the same way. The method is repeated in the same way and in the same sequence for increasing series of PPD ($1x10^{-3}$M) ranging from 0.1 to 3.5 ml to evaluate the calibration curve. Figure 5 shows the spectrum of the formed dye and Figure 4 shows the calibration curve.

Figure 4 exhibits a linear relation between the absorbances with the increasing amount of PPD within the range of (1.36-48 µg/mL) ($0.1\ x10^{-4}$- $3.5\ x10^{-4}$ M) with a high sensitivity of $3.690x10^{3}$ $L.mol^{-1}\cdot cm^{-1}$. The calculated LOD and LOQ are 0.22 and 0.06 µg/mL respectively. The calculated accuracy and precision of three replications of selected concentrations within the calibration curve are listed in Table 5. And Table 6 shows that water as a diluent is the best, in spit that other solvents cause bathochromic shift as Figure 6 shows.

3.3. The influence of interfering compounds

Increasing volumes (1-3 mL) of $1x\ 10^{-3}$ M of three expected organic compounds as interferences on the determination of fixed amount of PPD (I mL of $1x\ 10^{-3}$ M) has been studied, the results in Table 6 show good selectivity of the method.

4. Application and validation of the method

4.1. Determination of PPD in hair dye products and henna

In a10 ml volumetric flask, 1ml of 1ml of the prepared hair dye solutions or henna solution,1ml $NaIO_4$, 2 ml of 2,7-DHN and 1 ml NaOH were added, and the solutions diluted to be 10 ml exactly, the absorbances were measured at 586 nm after 10 minutes. Three replications of the measurements were taken for each type of hair product. The results are listed in Table 7.

4.2. Standard addition method

The effect of the interfering substances in the hair dye products was monitored by adding 1mL of these products of hair dye (each one alone) to a series of standard solutions so that the total concentrations were within the standard curve

of PPD. Figure 7 shows no interferences caused by other additives present in the hair dye products, Table 8 summarizes the results.

5.Conclusions

Some people may think that henna is better than hair dyes, but what we have shown in this research is that the selected type of henna contains a higher concentration of the chemical compound P-phenylenediamine (PPD), which indicates that not all henna is safe or made of natural ingredients., as was done in this research. In this research, PPD has been determined sensitively in henna and hair dyes of different origins, the method is based on the oxidation of PPD by NaIO4 then coupled with the 2,7dihydroxynaphthalene. Henna contains 0.625 mg PPD /0.2 g which is considered the highest value, while hair dyes contain from 0.0525 to 0.3075 mg PPD /0.2 g with an average recovery of 99% and average standard deviations of 1.198%.

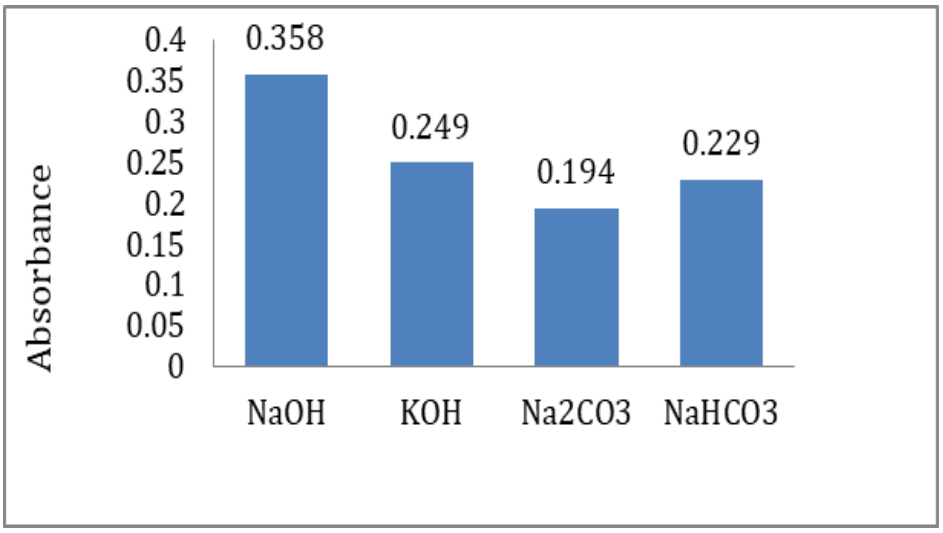


Fig. 1. The influence of different bases on the reaction

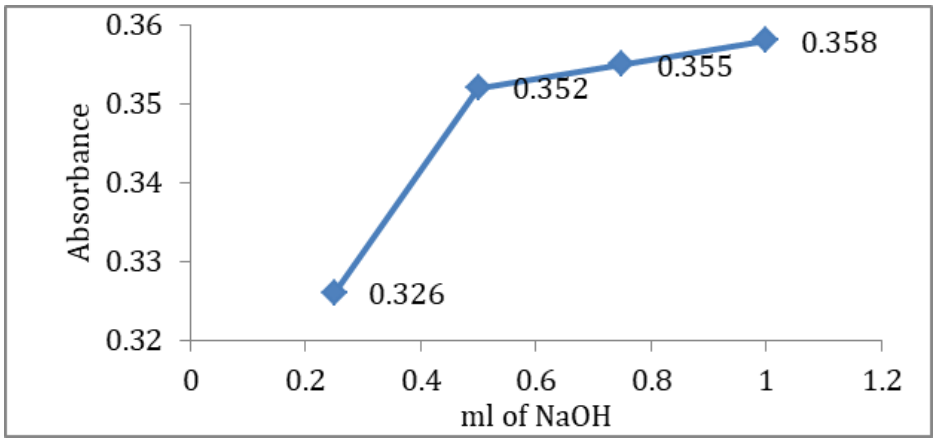


Fig. 2. The influence of the amount of

Table 1: The influence of the oxidant $NaIO_4$

mL of $NaIO_4$ (0.2%)	Absorbance /µg of PPD in 10ml				
	7	**14**	**20**	**27**	**R^2**
1.0	0.192	0.344	0.453	0.559	0.9917
1.5	0.193	0.351	0.482	0.567	0.9831
2.0	0.195	0.358	0.487	0.487	0.9885
2.5	0.209	0.365	0.491	0.589	0.9896
3.0	0.215	0.367	0.497	0.592	0.9898

Table 2: The influence of 0.5 to 2.5ml of 2,7DHN

mL of 2,7DHN (1×10^{-3}%)	Absorbance /µg of PPD in 10ml				
	7	**14**	**20**	**27**	**R^2**
0.5	0.079	0.097	0.111	0.141	0.9728
1.0	0.086	0.108	0.209	0.267	0.9510
1.5	0.103	0.226	0.290	0.353	0.9688
2.0	0.291	0.343	0.401	0.451	0.9993
2.5	0.301	0.354	0.413	0.476	0.9985

Table 3: Study of the stability

	Absorbance at min.									
immediately	**1**	**3**	**5**	**10**	**15**	**20**	**30**	**40**	**50**	**60**
0.441	0.441	0.441	0.443	0.446	0.446	0.446	0.446	0.446	0.443	0.443

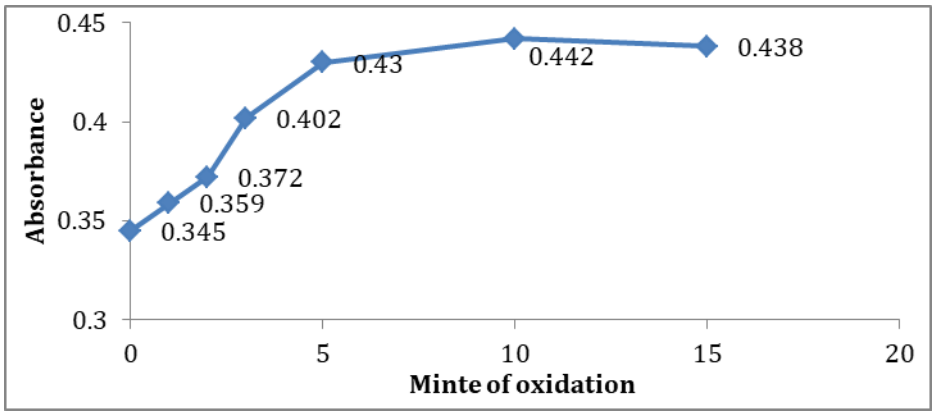


Fig. 3. The influence of oxidation time

Table 4: The influence of surfactants

Surfactant solution (SUR) (1×10^{-3} M)	Absorbance/Order* addition I	II	III	ƛ max
SDS	0.281	0.355	0.278	582
CTAB	0.333	0.425	0.276	580
CPC	0.425	0.336	0.341	581

Order*: I. 2,7DHN+$NaIO_4$+SUR.+PPD+NaOH, II. ,7DHN+$NaIO_4$+PPD+SUR.+NaOH, III. 2,7DHN+$NaIO_4$+PPD+ NaOH+ SUR.

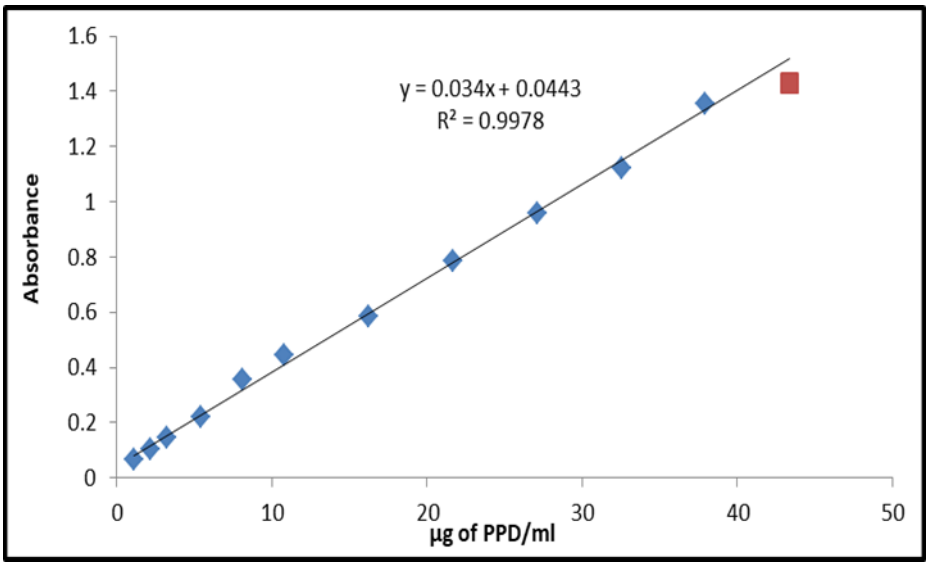


Fig. 4. The calibration curve for the determination of PPD

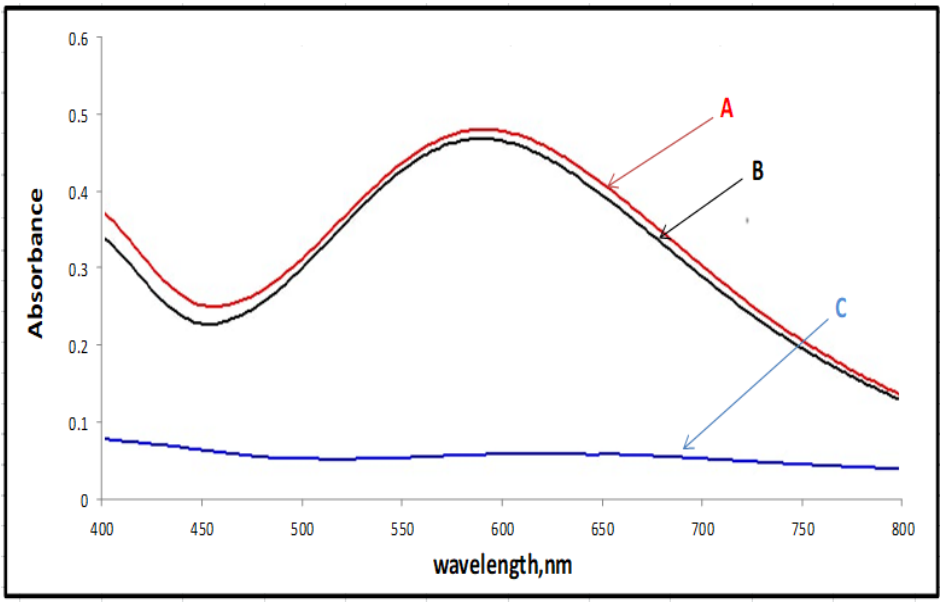


Fig. 5. The spectrum of 108 ppm of PPD-2,7DHN complex, A: against blank, B: against distilled water and C: Blank against distilled water.

Table 5: Accuracy and precision of the calibration curve

Ml of PADPA	Found	True	Recovery%	RSD%
0.5	0.214	0.214	100%	1.2
1.0	0.445	0.446	99%	0.38
2	0.783	0.785	90%	0.26

Table 6: The influence of solvents

Solvent	Absorbance	ƛ max
Ethanol	0.293	614
Methanol	0.215	612
DMSO	0.319	648
Acetone	turbid	turbid
Water	0.442	582

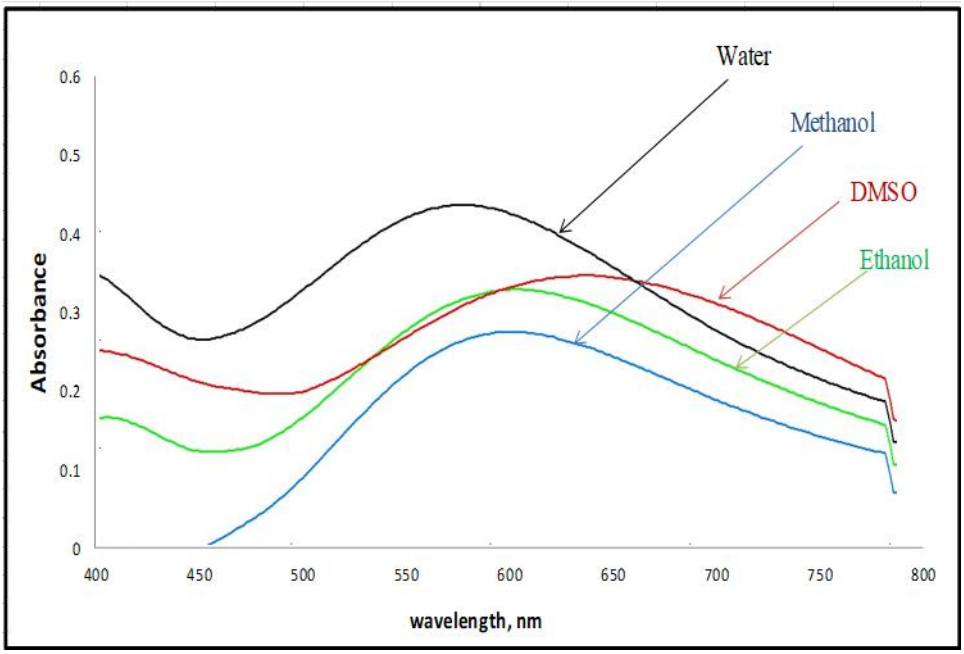


Fig. 6. The influence of solvents

Table 6: The influence of some organic compounds

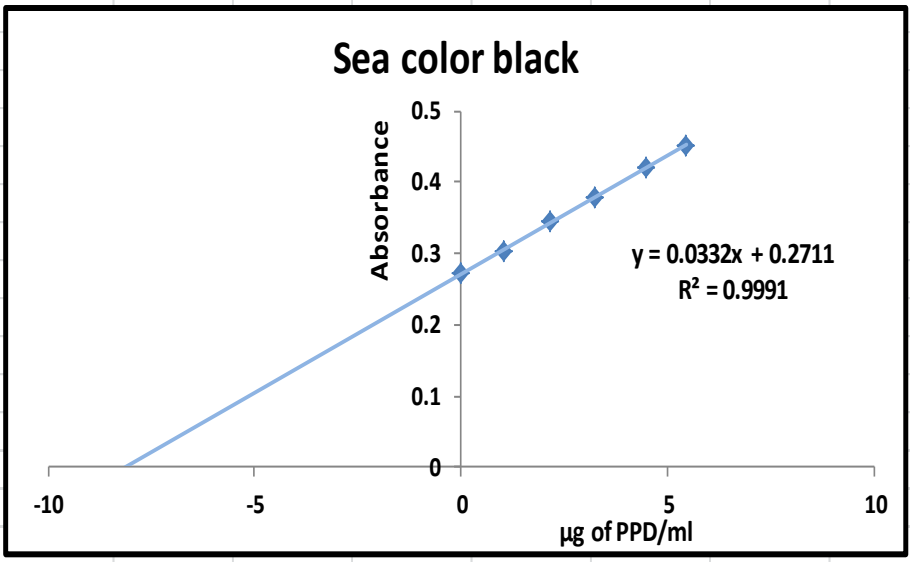
Compounds* ($1x\ 10^{-3}$ M)	Absorbance of PPD (I mL of $1x\ 10^{-3}$ M)/ mL of the compound		
	1	2	3
PADPA	0.445	0.443	0.447
PAP	0.442	0.445	0.439
RES	0.448	0.447	0.451

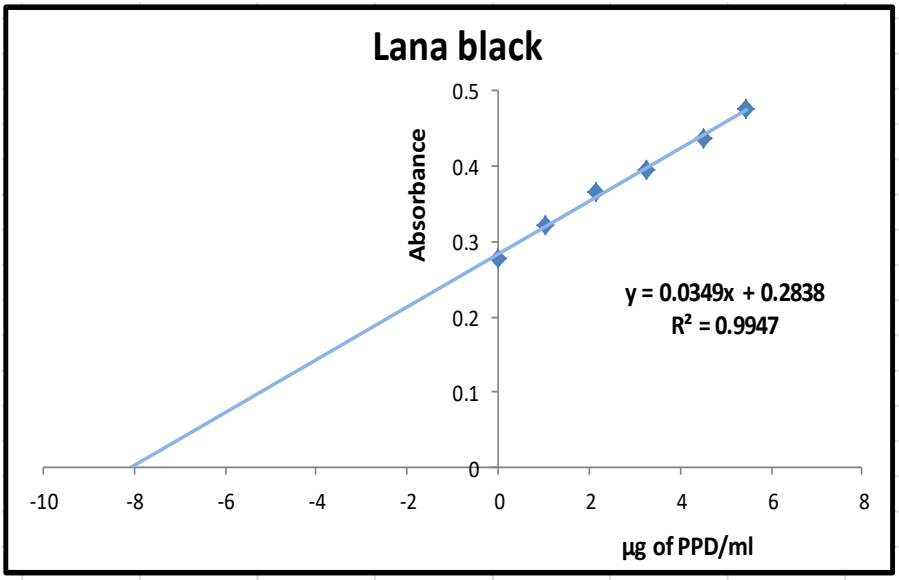
*PADPA :P-aminobiphenyl amine, PAP: p-aminophenol, RES: Resorcinol.

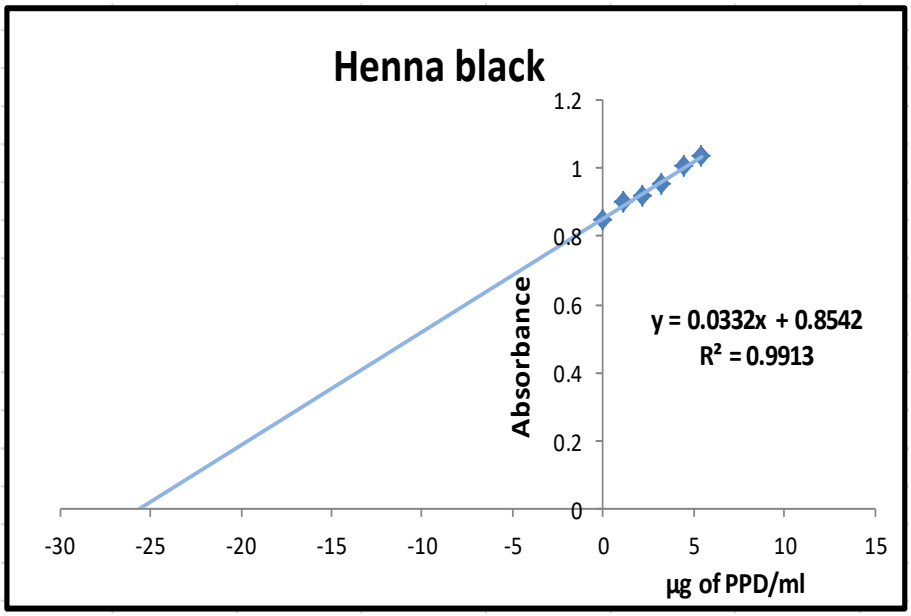
Table 7: Determination of PPD in hair dye products and henna

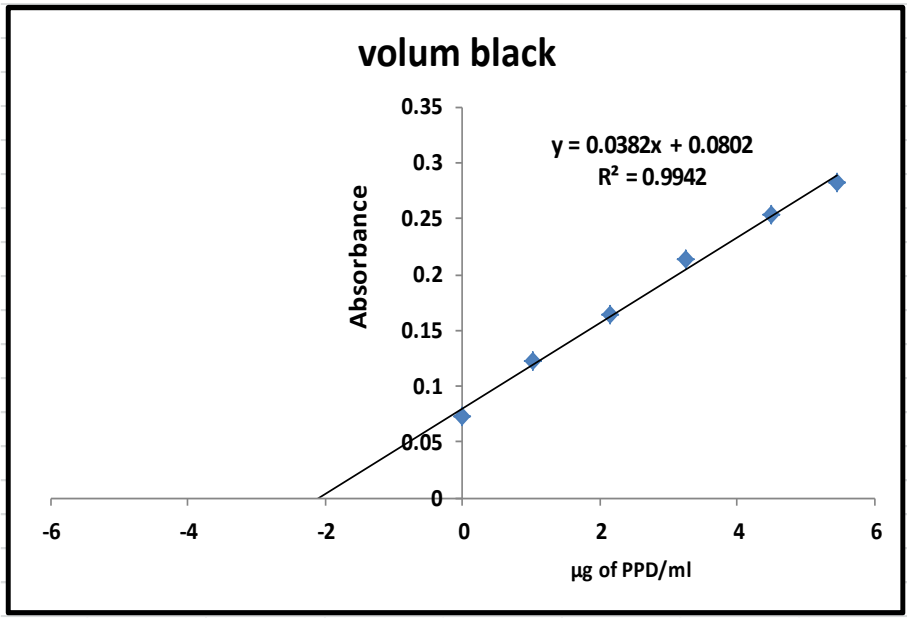
Hair dye	Color	Absorbance / 1ml of the prepared hair dye solution			Average**	Concentration (ppm)	Content in products (mg/0.2 g)
Sea color/ Turkey	Black	0.276	0.272	0.273	0.273	8	0.2
Lana* /Syria	Black	0.278	0.275	0.279	0.277	8.17	0.204
Henna /India	Black	0.854	0.851	0.852	0.852	25	0.625
Volum/Syria	Black	0.071	0.073	0.069	0.071	2.1	0.0525
Seacolor/ Turkey	Brown	0.417	0.419	0.420	0.418	12.3	0.3075
Starplex/ U.K.	Brown	0.081	0.085	0.083	0.083	2.38	0.0595
Garnne / Egypt	Blonde	0.111	0.113	0.115	0.113	3.3	0.0825
Collagen/ Italy	Grey	0.259	0.258	0.255	0.257	7.5	0.1875

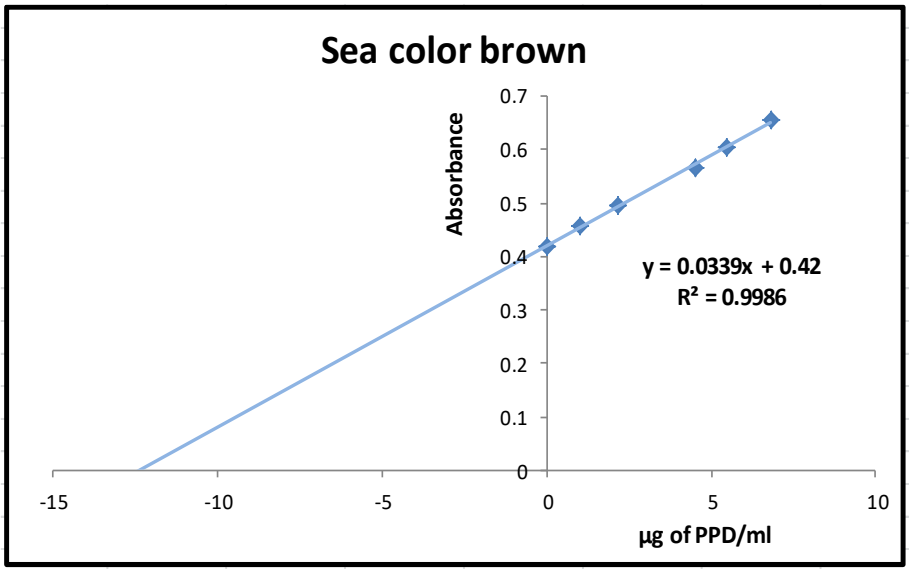
* For men ** Average of three replications

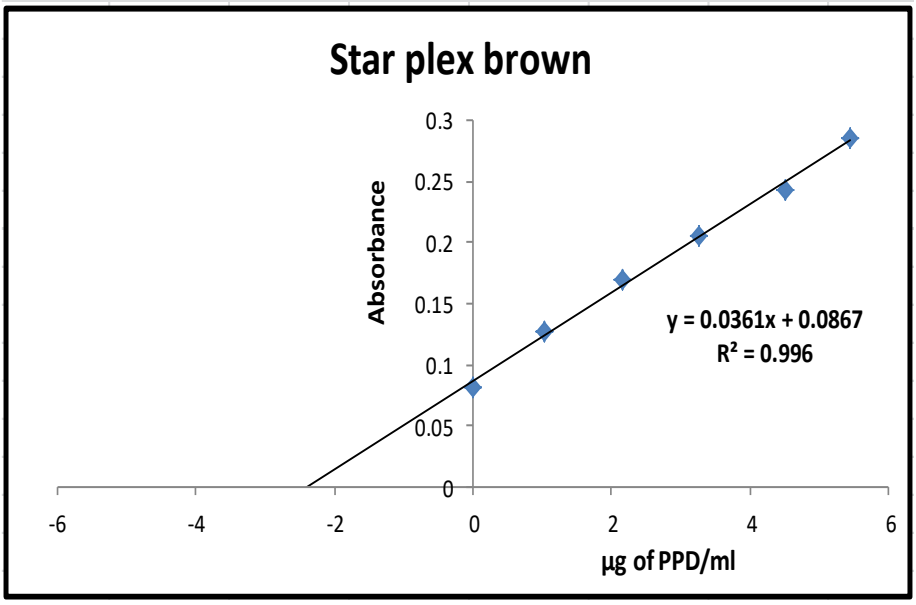

Sea color black
Absorbance
0.5
0.4
0.3
0.2
0.1
0
y = 0.0332x + 0.2711
R² = 0.9991
-10
-5
0
5
10
µg of PPD/ml

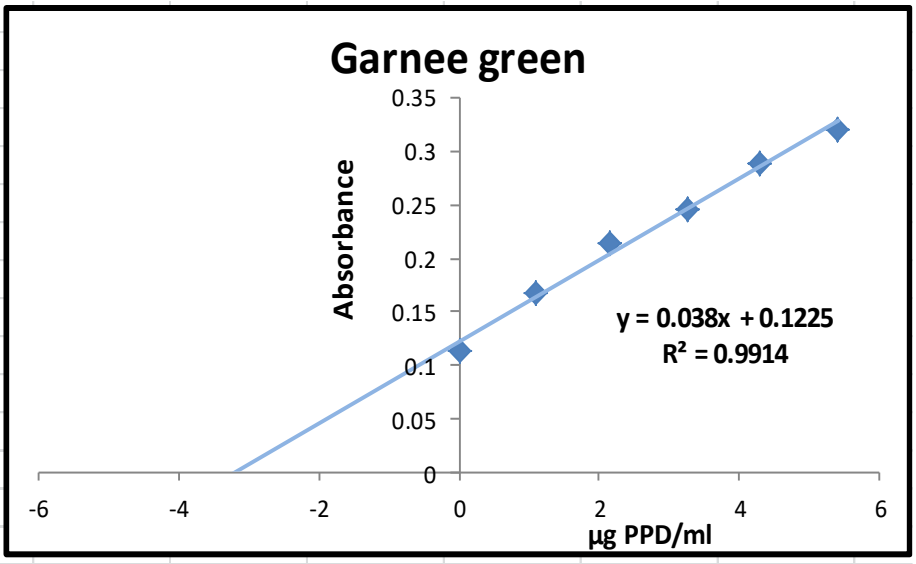

Lana black
Absorbance
0.5
0.4
0.3
0.2
0.1
0
y = 0.0349x + 0.2838
R² = 0.9947
-10
-8
-6
-4
-2
0
2
4
6
8
µg of PPD/ml


Henna black
Absorbance
1.2
1
0.8
0.6
0.4
0.2
0
y = 0.0332x + 0.8542
R² = 0.9913
-30
-25
-20
-15
-10
-5
0
5
10
15
µg of PPD/ml


volum black
y = 0.0382x + 0.0802
R² = 0.9942
Absorbance
µg of PPD/ml


Sea color brown
y = 0.0339x + 0.42
R² = 0.9986
Absorbance
µg of PPD/ml


Star plex brown
y = 0.0361x + 0.0867
R² = 0.996
Absorbance
µg of PPD/ml



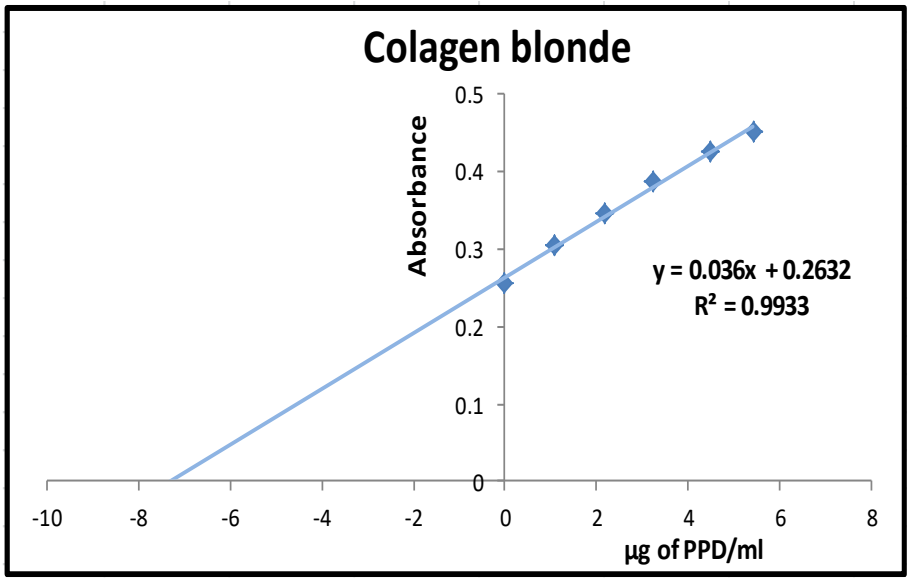


Fig. 7. The standard addition method for determination of PPD in the selected henna and hair dyes products.

Table 8: The information about the standard addition method

Hair dye	Absorbance	Taken (ppm)	Intercept	Slope	found	Recovery*%		RSD*%	
Sea color black	0.273	8	0.2711	0.0332	8.16	101		0.71	
Lana black	0.277	8.17	0.2838	0.0349	8.13	99	Average 99	0.72	Average 1.198
Henna black	0.852	25	0.8542	0.0332	25.7	102		0.14	
Star plex brown	0.083	2.38	0.0867	0.0361	2.4	101		2.4	
Sea color brown	0.418	12.3	0.4200	0.0339	12.38	97		0.4	
Volum black	0.071	2.1	0.0802	0.0382	2.09	99		2.8	
Garnee grey	0.113	3.3	0.1229	0.038	3.23	97		1.6	
Golagen blonde	0.257	7.5	0.2632	0.036	7.1	97		0.82	

* Three replications

6. Acknowledgements

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Conflict of Interest

The authors whose names are listed declare that there are no conflicts of interest (financial and/or non-financial) regarding the publication and/or funding of this manuscript.

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