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Analysis, construction, characterization, and application of copper nanowires loaded on activated carbon for removal of bromophenol blue in water samples

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### ABSTRACT

In the present work, a copper nanowire loaded on activated carbon (Cu-NW-AC) was fabricated and applied as an effective adsorbent for the removal of bromophenol blue (BPB) dye from aqueous solutions and then the percentage of removal was evaluated by UV-Vis spectrophotometer. The synthesized adsorbent was characterized and identified using techniques like Transmission electron microscopy and Brunauer-Emmett-Teller (BET). The effective (TEM) parameters of the removal process were investigated and optimized by experimental design methodology (EDM) based on response surface methodology (RSM) as a powerful optimization method. EDM is a unique method for following the effects of different factors on the removal process simultaneously. Analysis of variance (ANOVA) was used based on p-values and F-tests to investigate the accuracy and reliability of the used method. The optimized parameters were obtained as BPB concentration of 15 mg L<sup>-1</sup>, ultrasonic irradiation time of 14 min, adsorbent dosage of 0.018 g and pH= 5.5 under the desirability function. To evaluate the adsorption mechanism and calculation of maximum adsorption capacity, different adsorption isotherms were studied and according to the results, the Langmuir isotherm model showed the highest compatibility due to its higher R<sup>2</sup> (0.9905). Also, the proposed adsorbent represented good adsorption capacity (123.45 mg g<sup>-1</sup>). Moreover, kinetic studies proved the applicability of the pseudo-second-order model (R<sup>2</sup>=0.993) compared to other models. The achieved results confirmed the applicability of Cu-NW-AC as a versatile adsorbent for the removal of dye molecules from aqueous solutions using the adsorption method as a simple, easy, available and versatile method.

# 1. Introduction

Despite the broad applications of dye molecules in different areas and industries like plastic, textile, cosmetics, paper, and so on, the entrance of these materials into the environment, particularly water sources, has become a global

\*Corresponding Author: Saeid Khodadoust Email: khodadoust@bkatu.ac.ir https://doi.org/10.24200/amecj.v7.i01.253 concern [1]. Some of the dye molecules are hazardous and toxic for humans and animals and can cause irrecoverable and progressed diseases like cancer, mutation, and skin problems[2]. Some dye molecules have restricted and complex structures and are stable at high temperatures. So, they can remain in the environment for a long time and introduce some main problems[3]. Amongst different dye molecules, bromophenol blue (BPB) is considered an anionic dye that has been extensively applied in different fields such as additives, foods, textiles, and papers [4]. Although it is beneficial, it also can cause main environmental problems and hazardous diseases like carcinogenic activities[5]. These adverse effects of BPB have been exciting for researchers and scientists to deal with and design and use appropriate methods for its removal and treatment from the environment, especially water sources. One of the popular methods for the removal of dye molecules is the adsorption process (AP)[6]. AP is not only easy, cheap, available, and efficient but also can be used at large and industrial scales[7]. These exclusive attributes of AP make it a prevalent method for the removal of different pollutants. AP is based on the application of diverse materials as adsorbents, which can adsorb target analytes (dye molecules, organic molecules, etc...) via different adsorption mechanisms[8]. The proposed adsorbents for dye removal should have some main properties in terms of high chemical, physical, and thermal stability, large number of interaction sites, high adsorption capacity, suitable reusability, and fast adsorption rate[9, 10]. To date, various materials have been used as adsorbents. Activated carbon (AC), as one of the famous and applicable adsorbents, has been extensively used for the removal of dye molecules. This fact is due to exceptional properties of AC like simple and easy preparation, cost-effectiveness, sound surface area, and good adsorption capacity [11, 12]. To improve the adsorption capacity and the removal performance of AC, preparing its composites can be an ideal method. The goal of the presented work was the synthesis, characterization, and utilization of AC as an effective adsorbent for the removal of dyes from aqueous solutions. One of the innovations of this work has been the production of AC using disposable cellulose materials, and this has led to the production of a cheap absorbent. The sponginess of the produced AC also helps to absorb organic chemicals in

its pores. Nano-based adsorbents (NBAs) have attracted much attention in recent years. NBAs have the features of a versatile adsorbent like high stability, high surface area, high adsorption capacity, and, most importantly, the ability for modification and production of composition materials [13, 14]. Among different NBAs, NWs are very interesting due to their unique properties like high surface area, high adsorption capacity, and electrical features [15]. Cu-NWs have been broadly used in different fields because they have the properties of suitable adsorbent. A combination of AC and NADS like NW, can produce new types of adsorbents that have unique features, like high stability, high surface area, and high adsorption capacity. Investigation and optimization of the influential factors in the removal process is a vital concept. To this end, different affords have been used till now to use an appropriate approach. One-factor at a time (OFAT) is a traditional optimization approach in which the effect of one factor is followed while the other factors are fixed [16, 17]. In spite that OFAT is simple and easy, it needs to use a high volume of solvents and reagents, and it is also time-consuming. Most importantly, OFAT is not able to evaluate the effects of different parameters simultaneously and is not able to predict valid optimum values. To address these critical disadvantages, experimental design methodology based response surface methodology (RSM) is considered a powerful method and has been extensively applied in removal processes. EDM not only can address the drawbacks of OFAT but also can assess the interactions of effective factors concurrently. The aim of this study was the synthesis, characterization, and application of Cu-NW-AC as an efficient adsorbent for the removal of BPB from aqueous solutions. Moreover, to investigate the mechanism of interactions and

adsorption rate, different isotherm and kinetic models were used and assessed at obtained optimum conditions. The fabricated Cu-NW-AC showed a high capacity for the removal of BPB, which made it an excellent candidate for the removal of other pollutants, especially dye molecules.

# 2. Experimental

#### 2.1. Reagents and Instruments

In this study, all materials and reagents like BPB dye, acetone, distillation water, HCl, and NaOH were prepared at their highest purity form from Merck Company (Darmstadt, Germany). All instruments and laboratory equipment, including pH measurement (BP3001 model, Singapore), UV-Vis spectrophotometer (Perkin Elmer Lambda 25, USA), an ultrasonic bath with heating system (XUBA3 model, England), centrifuge system (Germany), and laboratory balance (XB22OA Precisa model, Switzerland) were prepared and used for calculation of removal percentage and adsorption capacity. For characterization and identification of prepared adsorbent, different techniques like TEM (Hitachi H-800 at 200 kV, Japan), and BET (Microtrac Bel Corp, USA) were used.

#### 2.2. Fabrication of Cu-NW-AC

To synthesize Cu-NW, 150 mL of NaOH solution (7.0 molL<sup>-1</sup>) and 7.5 mL of  $Cu(NO_3)_2$ solution (0.1 molL<sup>-1</sup>) were added to a 250 mL flask. Then, 1.95 mL ethylene diamine 95% and 0.75 mL hydrazine  $(N_2H_4)$  solution were added to the flask, respectively. Then, the obtained solution was heated at 60 °C for 45 min in an ultrasonic bath. After 36 min, the color of the solution was changed from blue to bronze. This color change confirmed the formation of Cu-NW. For the preparation of AC, at first, 10 g waste newspaper (as cellulosic source) was fragmented into 5 mm pieces and then dispersed in KOH solution at 80 °C for two h. The (W/W) ratio of newspaper, salt, and distillation water was selected as 1:3:4. Then, the obtained mixture was placed in an oven at 80 °C for 48 h to dry newspaper pieces. In the next step, the proposed newspaper pieces were crushed by mortar and transferred to an open container. The

target container was then placed in an oven for two hours at 500 °C. After cooling, the obtained ashes were washed with distillation water for neutralization (to get pH=7.5). In the final step, the achieved ACs were dried for 24 h at 120 °C and used for further steps. To prepare Cu-NW-AC, 150 mL of prepared Cu-NW solution was added to 30 g of AC in a 250 mL flask and stirred using a magnetic stirrer for four h. The obtained Cu-NW-AC was filtered, washed with water, and then dried at 110 °C for 12 h.

#### 2.3. Preparation of BPB standard solutions

In the preparation of the BPB stock solution, at first, 0.005 g of BPB dye was added to a 100 mL flask and then 100 mL of distillation water was added to the flask (50 mg  $L^{-1}$ ). For preparation of other solutions, the stock solution was diluted appropriately and then used for further experiments.

#### 2.4. Adsorption experiments

The ultrasonic-assisted adsorption of BPB by Cu-NW-AC was carried out as follows: 15 mg L<sup>-1</sup> of BPB solution was prepared from the stock solution and then adjusted at pH 5.5. In the next step,0.018 g of adsorbent was added to the BPB solution and sonicated for 14 min. Then, the solution was centrifuged to separate the adsorbent from the dye solution. In the final step, the remaining concentration of BPB was determined by UV–Vis spectrophotometer and removal percentage and adsorption capacity were calculated by the following Equations I and II.

$$R\% = C_0 - C_0 / C_0 * 100$$
 (Eq. 1)

$$Q_{e} = ((C_{0} - C_{e})V)/m$$
 (Eq.2)

Where  $C_0$  is the initial BPB concentration (mg L<sup>-1</sup>),  $C_e$  is the equilibrium BPB concentration (mg L<sup>-1</sup>), V is the volume of BPB solution (L), and m is the mass of adsorbent (g). The steps of adsorption and measurement of BPB are shown schematically in Figure 1.



Fig. 1. The steps of adsorption and measurement of BPB

# 3. Results and discussion

# 3.1. Characterization of adsorbent

The characterization of the proposed adsorbent, TEM and SEM analyses were used. Using TEM analysis, the morphology and size of the prepared adsorbent can be investigated. According to TEM images (Figure 2A), there are some black areas with high density, which are related to Cu-NW that were distributed on white areas (AC) with low density. According to SEM images of the AC (Figure 2B), it had a porous structure in which the pores had a spherical shape. Moreover, to evaluate the specific surface area and type of adsorbent, BET analysis is proper. So, based on the BET analysis, the fabricated adsorbent had a surface area of 200 g m<sup>-2</sup>.



In this work, to investigate the influential factors, and their interactions and achieve valid optimum values, RSM was applied. The effects of different variables such as BPB concentration  $(X_1)$  (10-30 mgL<sup>-1</sup>), pH  $(X_2)$  (2-10), adsorbent dosage  $(X_3)$ (4-20 mg), and sonication time  $(X_4)$  (4-16 min) on removal efficiency were optimized using 4 factors at 5 level CCD. The design matrix and the responses for the BPB dye removal using Cu-NW-AC adsorbent are shown in Table 1.

In addition, the accuracy and reliability of the applied method were investigated by ANOVA at a certain confidence level ( $\alpha = 0.05$ )(Table 2)[18].To assess the adequacy and reliability of the proposed model, different parameters such as R<sup>2</sup>, adjusted R<sup>2</sup>, F test, and lack-of-fit test were used. According to the ANOVA analysis, the factors that had p-values less than 0.05 and higher F-values were significant and had significant contributions to the removal process. On the other hand, the parameters with p-values more than 0.05 and lower F-values were not significant and didn't have significant contributions to the removal process[19]. The higher values of R<sup>2</sup> and Adj R<sup>2</sup> proved the accuracy of the used model. Moreover, the lack of a value of more than 0.05 (0.143769) proved the precision and dependability of the recommended model.

Factors				Levels		Star p	oint α=2
			Low (-1)	Central(0)	High(+1)	-α	$+\alpha$
(X <sub>1</sub> ) BPB concentra (X <sub>2</sub> ) pH value (X <sub>3</sub> ) Adsorbent dos (X <sub>4</sub> ) Sonication tim	ation (mg L <sup>-1</sup> ) age (mg) e (min)		15 4 8 7	20 6 12 10	25 8 16 13	10 2 4 4	30 10 20 16
Run	$(X_1)$	(X <sub>2</sub> )		(X <sub>3</sub> )	$(X_4)$	Res	ponse (BPB ER%)
1	15	4		8	7		37.47
2	15	4		8	13		52.07
3	15	4		16	7		61.13
4	15	4		16	13		76.79
5	15	8		8	7		39.13
6	15	8		8	13		50.26
7	15	8		16	7		59.86
8	15	8		16	13		79.76
9	25	4		8	7		31.23
10	25	4		8	13		35.88
11	25	4		16	7		34.84
12	25	4		16	13		44.84
13	25	8		8	7		23.40
14	25	8		8	13		30.33
15	25	8		16	7		22.01
16	25	8		16	13		37.79
17	10	6		12	10		78.92
18	30	6		12	10		25.59
19	20	2		12	10		51.17
20	20	10		12	10		31.65
21	20	6		4	10		22.81
22	20	6		20	10		58.97
23	20	6		12	4		25.94
24	20	6		12	16		60.83
25 (C)	20	6		12	10		56.37
26 (C)	20	6		12	10		55.46

Table 1. The design matrix and the responses for the BPB dye removal using Cu-NW-AC adsorbent

(C): Center point

Adj  $R^2 = 0.965$ 

 $R^2 = 0.984$ 

Source of variation	Sum of square	Dfa	Mean square	F-value <sup>b</sup>	P value
X	3820.579	1	3820.579	9227.337	0.006627
$\mathbf{X}_2$	208.565	1	208.565	503.720	0.028346
X <sub>3</sub>	1497.366	1	1497.366	3616.389	0.010585
$X_4$	1182.028	1	1182.028	2854.795	0.011914
$X_{1}^{2}$	12.988	1	12.988	31.367	0.112483
$X_{2}^{2}$	222.937	1	222.937	538.430	0.027419
$X_{3}^{2}$	239.451	1	239.451	578.314	0.026457
$X_{4}^{2}$	165.592	1	165.592	399.932	0.031807
$\mathbf{X}_{1} \mathbf{X}_{2}$	75.734	1	75.734	182.909	0.046986
$\mathbf{X}_{1} \mathbf{X}_{3}$	399.700	1	399.700	965.342	0.020483
$X_1 X_4$	35.790	1	35.790	86.440	0.068211
$X_2 X_3$	1.351	1	1.351	3.264	0.321838
$X_2 X_4$	4.873	1	4.873	11.769	0.180566
$X_{3}X_{4}$	36.090	1	36.090	87.164	0.067930
Lack of Fit	119.795	10	11.980	28.933	0.143769
Pure Error	0.414	1	0.414		
Total SS	10965.01	25			

Table 2. Analysis of variance (ANOVA) for removal of BPB dye using Cu-NW-AC adsorbent.

<sup>a</sup>Df: Degrees of freedom

<sup>b</sup> Test for comparing model variance with residual (error) variance

The relationship between influential factors and response (R%BPB) is presented in Equation 3. Equation 3 confirmed that the effective factors with positive signs had a direct effect on the removal percentage and can increase it. In contrast, the factors with negative signs had the opposite effect and could reduce the removal percentage[20, 21].

 $\begin{array}{c} \text{RE\%}{=}55.915{\text{-}}12.617\text{X}_{1}{\text{-}}\\ 2.948\text{X}_{2}{\text{+}}7.899\text{X}_{3}{\text{+}}7.018\text{X}_{4}{\text{-}}3.574\text{X}_{2}{\text{-}}\\ 3.704\text{X}_{3}{\text{-}}3.080\text{X}_{4}{\text{-}}2.176\text{X}_{1}\text{X}_{2}{\text{-}}4.998\text{X}_{1}\text{X}_{3}\\ \text{(Eq.3)} \end{array}$ 

# 3.3. Response surface methodology for investigating effective variables

RSM uses 3-dimensional (3D) response surface plots to investigate the effects of different factors on the removal process (Figure 3) [22]. According to these plots, increasing BPB concentration can reduce the removal percentage. At low BPB concentrations, the target adsorbent can interact with more dye molecules and remove them. Increasing adsorbent dosage enhanced the removal percentage<sup>[23]</sup>. In this manner, the functional groups, the specific surface area, and the large reactive surface center of the adsorbent are high and more available to interact with molecule dyes. So, decreasing the adsorbent dosage can decrease the removal percentage. The maximum removal was obtained at a lower pH. BPB is an anionic dye. On the other hand, at higher pH values, since the concentration of hydroxide (OH) is high, the adsorbent can get a negative charge that can produce a repulsive force between the adsorbent and dye molecules and subsequently reduce the removal percentage. As can be seen from 3D plots, enhancing ultrasonic time increased the removal percentage. With increasing time, the possible interactions between adsorbent and dye molecules are increased, which subsequently can raise the removal percentage. After investigating the effective factors, the optimum values were achieved under the desirability function (DF). DF can convert the predicted and experimental response of each factor into a desirability score, while its values between 0.0 and 1.0 show the completely undesirable and fully desired response, respectively. According to the desirability score, maximum recovery (87.455%) was achieved with a BPB concentration of 15 mg  $L^{-1}$ , ultrasonic irradiation time (14 min), adsorbent dosage (0.018 g), and pH= 5.5 (Figure 4).



Fig. 3. 3D response surface plots for investigation of the effects of different factors on removal of BPB dye removal using Cu-NW-AC adsorbent



Fig.4. The obtained optimum values based on DF

#### 3.4. Adsorption isotherm models

To assess the adsorption mechanism and achieve maximum adsorption capacity (q\_), various isotherms models like Langmuir, Freundlich, Temkin, and Dubinin-Radushkevich (DR) were investigated (Table 3). According to the results of different models, Langmuir was more suitable than others which were due to its higher  $R^2$  (0.9905). The Langmuir model explains that the adsorption of dyes onto adsorbent is monolayer and on the homogeneous surface of the adsorbent. The calculated  $\boldsymbol{q}_{\boldsymbol{m}}\!,$  according to the Langmuir model, was 123.45 mg g<sup>-1</sup>. In the Freundlich model, then values of more than 1 proved the favourable adsorption condition. In the Dubinin-Radushkevich model, the values of mean energy (E) less than 8 kJ mol<sup>-1</sup> represented that the adsorption process of BPB dye was probably controlled physically.

#### 3.5. Kinetic models

In the evaluation adsorption process, the adsorption kinetics can be considered an essential characteristic. To this end, pseudo-first order, pseudo-second order, Elovich, and intraparticle diffusion kinetic models were used to discuss the adsorption behaviour (Table 4). Results proved that the adsorption of BPB by Cu-NW-AC was followed by pseudo-second-order which was due to its high  $R^2$  (0.993).

Recently, many techniques based on nanoadsorbents have been used for the removal of dye and other organic material (BTEX) in water samples [24-31]. To further demonstrate the superiority of our proposed method, a comparison of the important features of the proposed method with those reported in the literature [32-35] is given in Table 5.

Isotherm	Equation	Factors	Adsorbent (g)	Time (min)		
models	Equation	i uctor ș	0.018	Adsorbent (g)         Time (min)           0.018         14           123.45		
		$q_m(mgg^{-1})$	123.4	5		
Langmuir	$C_e/q_e = 1/(K_aQ_m) + C_e/Q_m$	$k_a(l mg^{-1})$	3.681			
		$R^2$	0.990	5		
		<u>1</u> n	0.295	;		
Freundlich	$Ln q_e = ln K_F + (1/n) ln C_e$	$K_f(lmg^{-1})$	83.21			
		$R^2$	0.224	5		
		$\beta_1$				
Temkin	$q_e = B_l ln K_T + B_l ln C_e$	$K_T(l mg^{-1})$	100.3	7		
		$R^2$	0.186	3		
5 1		β				
Dubinin and Radushkevich	$Ln q_e = lnQ_m - K\epsilon^2$	q <sub>s</sub>	100.8	3		
		$R^2$	0.435	6		

Table 3. Different isotherm models for removal of BPB by Cu-NW-AC.

Kinetic	Equation	Factors	Adsorbent (g)	Concentration (mg L <sup>-1</sup> )		
models	Lyunton	1 actors	0.018	15		
		$k_1(\min^{-1})$	(	).224		
Pseudo-first order	$\log(q_{e}-q_{t}) = \log q_{e} - \frac{K_{1} t}{2/303}$	$q_e(mgg^{-1})$	1	19.31		
		$\mathbb{R}^2$	(	0.967		
D 1 1		$k_2(\min^{-1})$				
Pseudo-second- order-kinetic	$t/q_t = 1/k_2 q_e^2 + (1/q_e)t$	$q_e(mg g^{-1})$				
		$R^2$	0.993			
T 1		$k\left(\text{mg g}^{-1}\text{min}^{-\frac{1}{2}}\right)$	-			
Intraparticle diffusion	$q_{e} = k_{3}t^{\frac{1}{2}} + c$	$c(mg g^{-1})$	-			
		$R^2$	0.997			
Elovich		$\beta \left(g m g^{-1}\right)$	-			
	$q_t = \ln \frac{(\alpha \beta)}{\beta} + (\frac{1}{\beta}) \ln t$	$\alpha \left( mg \ g^{-1}min^{-1} \right)$	-			
	н , н,	$R^2$	(	0.987		

Table 4. Different kinetic models for removal of BPB by Cu-NW-AC

Table 5. Comparison of the proposed method with some adsorbent reported in the literature

Sorptioncapacity (mg g <sup>-1</sup> )	рН	Contact times (min)	References
123.45	9	120	[32]
129.60	3	80	[33]
101	4	24 h	[34]
27.72		120	[35]
123.45	5.5	14	Thisstudy
	Sorptioncapacity (mg g <sup>-1</sup> ) 123.45 129.60 101 27.72 123.45	Sorptioncapacity (mg g <sup>-1</sup> )         pH           123.45         9           129.60         3           101         4           27.72         123.45           123.45         5.5	Sorptioncapacity (mg g <sup>-1</sup> )         pH         Contact times (min)           123.45         9         120           129.60         3         80           101         4         24 h           27.72         120           123.45         5.5

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# 4. Conclusion

The aim of the presented work was the fabrication and application of Cu-NW-AC as an adsorbent for the removal of BPB dye from water samples. After synthesis, the proposed adsorbent was characterized by TEM and BET analyses. To evaluate effective factors, and their interactions, and obtain valid optimum values, EDM-based RSM was used. Moreover, to judge the accuracy and adequacy of the applied model, ANOVA was incorporated. The adsorption kinetic models and equilibrium parameters, such as kinetic rate constants, the Langmuir, Freundlich, Temkin, D-R constants, and maximum capacity of adsorption, were obtained from the adsorption experiments. These parameters are very important for scale-up batch experiments. A comparison of kinetic models on the overall adsorption rate showed that the dye adsorbent system was best described by the pseudosecond-order rate model. The adsorption data fitted well with the Langmuir isotherm.

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# 6. References

- D. Akın, A. Yakar, U. Gündüz, Synthesis of magnetic Fe3O4-chitosan nanoparticles by ionic gelation and their dye removal ability, Water Environ. Res., 87 (2015) 425-436. https:// doi.org/10.2175/106143014x14062131178673
- [2] S. Gita, S.P. Shukla, G. Deshmukhe, A.R. Singh, T.G. Choudhury, A.K. Singh, Adsorption– biodegradation coupled remediation process for the efficient removal of a textile dye through chemically functionalized sugarcane bagasse, Water Environ. Res., 93 (2021) 2223-2236. https://doi.org/10.1002/wer.1595
- [3] A.J. Ibrahim, H.A. Wahid Dwesh, A.R.Y. Al-Sawad, Adsorption of methylene blue dye onto bentonite clay: Characterization, adsorption isotherms, and thermodynamics study by using UV-Vis technique, Anal. Methods

Environ. Chem. J., 6 (3) (2023) 5-18. https:// doi.org/10.24200/amecj.v6.i03.243

- [4] M. Rastgordani, J. Zolgharnein, V. Mahdavi, Derivative spectrophotometry and multivariate optimization for simultaneous removal of Titan yellow and bromophenol blue dyes using polyaniline@SiO2 nanocomposite, Microchem. J., 155 (2020) 104717. https:// doi.org/10.1016/j.microc.2020.104717
- [5] D. Pandey, A. Daverey, K. Dutta, K. Arunachalam, Dye removal from simulated and real textile effluent using laccase immobilized on pine needle biochar, J. Water Proc. Eng., 53 (2023) 103710. https://doi. org/10.1016/j.jwpe.2023.103710
- [6] J. Zhao, L. Xu, Y. Su, H. Yu, H. Liu, S. Qian, W. Zheng, Y. Zhao, Zr-MOFs loaded on polyurethane foam by polydopamine for enhanced dye adsorption, J. Envirn. Sci., 101 (2021) 177-188. https://doi.org/10.1016/j. jes.2020.08.021
- [7] S. Khodadoust, N. Cham Kouri, Preconcentration of Sn (II) using the methylene blue on the activated carbon and its determination by spectrophotometry method, Spectrochim. Acta A Mol. Biomol., 123 (2014) 85-88. https://doi. org/10.1016/j.saa.2013.12.058
- [8] C. Jamshidzadeh, H. Shirkhanloo, A new analytical method based on bismuth oxidefullerene nanoparticles and photocatalytic oxidation technique for toluene removal from workplace air, Anal. Methods Environ. Chem. J. 2 (2019) 73-86. https://doi.org/10.24200/ amecj.v2.i01.55
- B.A. Gizaw, N.G. Habtu, Catalytic wet air oxidation of azo dye (reactive red 2) over copper oxide loaded activated carbon catalyst, J. Water Proc. Eng., 48 (2022)102797. https:// doi.org/10.1016/j.jwpe.2022.102797
- [10] S. Bahah, Analytical study on lead elimination by anionic clays: Characterization, adsorption kinetics, isotherm, thermodynamic, mechanism and adsorption, Anal. Methods Environ. Chem. J., 3 (2023) 67-88. https://doi.org/10.24200/ amecj.v6.i03.248

- [11] S. Khodadoust, M. Ghaedi, R. Sahraei, A. Daneshfar, Application of experimental design for removal of sunset yellow by copper sulfide nanoparticles loaded on activated carbon, J. Ind. Eng. Chem., 20 (2014) 2663-2670. https://doi.org/10.1016/j. jiec.2013.10.053
- [12] A. J. Ibrahim, ZnO nanostructure synthesis for the photocatalytic degradation of azo dye methyl orange from aqueous solutions utilizing activated carbon, Anal. Methods Environ. Chem. J., 5 (4) (2022) 5-19. https:// doi.org/10.24200/amecj.v5.i04.200
- [13] A. Amiri Pebdani, S. Dadfarnia, A.M. Haji Shabani, S. Khodadoust, Application of Ni:ZnS nanoparticles loaded on magnetic multi-walled carbon nanotubes as a sorbent for dispersive micro-solid phase extraction of phenobarbital and phenytoin prior to HPLC analysis: experimental design, RSC Adv., 6 (2016) 89250-89258. https://doi.org/10.1039/ C6RA15981H
- [14] F. Momtazan, S. Khodadoust, F. Zeraatpisheh, M. Behbahani, Synthesis of mesoporous silica for adsorption of chlordiazepoxide and its determination by HPLC: Experimental design, J. Sep. Sci., 42 (2019) 3253-3260. https://doi.org/10.1002/jssc.201900373
- [15] R. Ravindren, S. Mondal, K. Nath, N.C. Das, Prediction of electrical conductivity, double percolationlimitandelectromagnetic interference shielding effectiveness of copper nanowire filled flexible polymer blend nanocomposites, Compos. B: Eng., 164 (2019) 559-569. https:// doi.org/10.1016/j.compositesb.2019.01.066
- [16] Ehsan Zolfonoun, Spectrofluorometric determination of L-tryptophan after preconcentration using multi-walled carbon nanotubes, Anal. Methods Environ. Chem. J., 2 (2019) 43-48, https://doi.org/10.24200/ amecj.v2.i01.43
- [17] N. Chamkouri, S. Khodadoust, F. Ghalavandi, Solid-phase extraction coupled with HPLC-DAD for determination of B vitamin concentrations in halophytes, J. Chromatogr.

Sci., 53 (2015) 1720-1724. https://doi. org/10.1093/chromsci/bmv080

- [18] M. Kaabipour, S. Khodadoust, F. Zeraatpisheh, Preparation of magnetic molecularly imprinted polymer for dispersive solid-phase extraction of valsartan and its determination by high-performance liquid chromatography: Box-Behnken design, J. Sep. Sci., 43 (2020) 912-919. https://doi.org/10.1002/jssc.201901058
- [19] P. Thomas, N.P. Rumjit, C.W. Lai, M.R.B. Johan, EDTA functionalised cocoa pod carbon encapsulated SPIONs via green synthesis route to ameliorate textile dyes-Kinetics, isotherms, central composite design and artificial neural network, Sustain. Chem. Pharm., 19 (2021) 100349. https://doi. org/10.1016/j.scp.2020.100349
- [20] A. Amiri Pebdani, A.M. Haji Shabani, S. Dadfarnia, M.S. Talebianpoor, Saeid Khodadoust, Preconcentration of valsartan by dispersive liquid–liquid microextraction based on solidification of floating organic drop and its determination in urine sample: Central composite design, J. Sep. Sci., 39 (10) 1935-1944. https://doi.org/10.1002/ jssc.201600010
- [21] S. Khodadoust, T. Nasiriani, F. Zeraatpisheh, Preparation of a magnetic molecularly imprinted polymer for the selective adsorption of chlordiazepoxide and its determination by central composite design optimized HPLC, New J. Chem., 42 (2018) 14444-14452. https://doi.org/10.1039/C8NJ02643B
- [22] G. Swain, S. Singh, R. Sonwani, R. Singh, R.P. Jaiswal, B. Rai, Removal of Acid Orange 7 dye in a packed bed bioreactor: Process optimization using response surface methodology and kinetic study, Bioresour. Technol. Rep., 13 (2021) 100620. https://doi. org/10.1016/j.biteb.2020.100620
- [23] M. Dolatabadi, S. Ahmadzadeh, Catalytic ozonation process using modified activated carbon as a cataly s t for the removal of sarafloxacin antibiotic from aqueous

solutions, Anal. Methods Environ. Chem. J., 6 (2) (2023) 31-41. https://doi.org/10.24200/ amecj.v6.i02.236

- [24] M Arjomandi, A review: analytical methods for heavy metals determination in environment and human samples, Anal. Methods Environ. Chem. J., 2 (2019) 97-126. https://doi. org/10.24200/amecj.v2.i03.73
- [25] N. Esmaeili, J. Rakhtshah, Ultrasound assisted-dispersive-modification solidphase extraction using task-specific ionic liquid immobilized on multiwall carbon nanotubes for speciation and determination mercury in water samples, Microchem. J., 154 (2020) 104632. https://doi.org/10.1016/j. microc.2020.104632
- [26] R. Ashouri, Dynamic and static removal of benzene from air based on task-specific ionic liquid coated on MWCNTs by sorbent tubeheadspace solid-phase extraction procedure, Int. J. Environ. Sci. Technol., 18 (2021) 2377-2390. https://doi.org/10.1007/s13762-020-02995-4
- [27] J. Rakhtshah, A rapid extraction of toxic styrene from water and wastewater samples based on hydroxyethyl methylimidazolium tetrafluoroborate immobilized on MWCNTs by ultra-assisted dispersive cyclic conjugation-micro-solid phase extraction, Microchem. J., 170 (2021) 106759. https:// doi.org/10.1016/j.microc.2021.106759
- [28] P. Paydar, A novel method based on functionalized bimodal mesoporous silica nanoparticles for efficient removal of lead aerosols pollution from air by solid-liquid gas-phase extraction, J. Environ. Health Sci. Eng., 18 (2020) 177–188. https://doi. org/10.1007/s40201-020-00450-7
- [29] S. Teimoori, An immobilization of aminopropyl trimethoxysilane-phenanthrene carbaldehyde on graphene oxide for toluene extraction and separation in water samples, Chemosphere, 316 (2023) 137800. https:// doi.org/10.1016/j.chemosphere.2023.137800
- [30] S. Teimoori, A.H. Hassani, M. Panahi, N.

Mansouri, Rapid extraction of BTEX in water and milk samples based on functionalized multi-walled carbon nanotubes by dispersive homogenized-micro-solid phase extraction, Food Chem., 421 (2023) 136229. https://doi. org/10.1016/j.foodchem.2023.136229

- [31] S. Teimoori, A.H. Hassani, New extraction of toluene from water samples based on nano-carbon structure before determination by gas chromatography, Int. J. Environ. Sci. Technol., 20 (2023) 6589–6608. https://doi. org/10.1007/s13762-023-04906-9
- [32] A.Q. Alorabi, M.S. Hassan, M. Fe3O4-CuO-activated Azizi, carbon composite as an efficient adsorbent for bromophenol blue dye removal from aqueous solutions, Arab. J. Chem., 13 (2020) 8080-8091. https://doi.org/10.1016/j. arabjc.2020.09.039
- [33] M. Rastgordani, J. Zolgharnein, V. Mahdavi, spectrophotometry Derivative and multivariate optimization for simultaneous removal of Titan yellow and Bromophenol blue dyes using polyaniline@ SiO2 nanocomposite, Microchem. J., 155 (2020) 104717. https://doi.org/10.1016/j. microc.2020.104717
- [34] M. Essandoh, Rafael A. Garcia, Efficient removal of dyes from aqueous solutions using a novel hemoglobin/iron oxide composite, Chemosphere, 206 (2018) 502-512. https:// doi.org/10.1016/j.chemosphere.2018.04.182
- [35] S. Dhananasekaran, R. Palanivel, S. Pappu, Adsorption of methylene blue, bromophenol blue, and coomassie brilliant blue by α-chitin nanoparticles, J. Adv. Res., 7 (2016) 113-124, https://doi.org/10.1016/j.jare.2015.03.003



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# Determine methylene blue based on carbon paste electrode modified with nanoparticles of nickel oxide-nitrogen carbon quantum dots and carbon structures by cyclic voltammetry

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#### ABSTRACT

This paper deals with an electrochemical method for the determination of methylene blue (MB) by fabrication of an electrode based on a carbon paste modified with nano-nickel oxide and nitrogen carbon quantum dots (NiO-NCQD), graphene-carbon nitride (G-C<sub>3</sub>N<sub>4</sub>), reduced graphene oxide (rGO), and graphite powder and paraffin oil are as a plasticizer. This electrode is used as a working electrode. The analytical method used is cyclic voltammetry (CV), The oxidationreduction curve of methylene blue was shown using this electrode. It is a quasi-reversible curve, and it works at (pH = 1) and the best acid used is HCl a concentration of (0.1M). It was also found that the linear range is within the range of  $(7.99-31.98 \text{ mg L}^{-1})$ , where the oxidation equation for it can be described by the equation  $I_{OX} = 1.508C_{MB} + 229.5$ and while the redaction equation is  $I_{Red}^{-2.236C} = -2.236C_{MB}^{-2.232.5}$  where is the correlation coefficient (R<sup>2</sup>=0.9823) and (R<sup>2</sup>=0.9722) for both oxidation and reduction respectively. The standard deviation (SD) and relative standard deviation (RSD%) were obtained at (0.361 mg  $L^{-1}$  and 0.294 mg  $L^{-1}$ ) and (4.52% and 3.68%) for both oxidation and reduction respectively. Retrospective, the limit of quantitative (LOQ) and limit of detection (LOD) were achieved at (99.65%; 99.70%), (0.24 mg L<sup>-1</sup>; 0.13 mg L<sup>-1</sup>), and (0.071 mg L<sup>-1</sup>; 0.039 mg L<sup>-1</sup>) for both oxidation and reduction respectively. Methylene blue was analyzed by UV-Vis spectrophotometry at (663 nm).

# 1. Introduction

Dyes are colored aromatic organic compounds (VOCs) that absorb light in the visible [1]. Many organic dyes leak into water due to industrial facilities, thus causing harm to the environment [2]. One of these organic dyes is methylene blue (MB) with the chemical formula shown in Figure 1 [3]. The Methylene blue (MB) with a formula of  $C_{16}H_{18}CIN_3S$  [4] is also called Methylthioninium chloride [5]. MB is from the phenothiazine family, and it is called 3,7

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-Bis(dimethylamino) phenothiazine [6], heterocyclic dye [7], cationic dye [8], and it is a basic dye [9]. Methylene blue is used in many applications, for example: in the dyeing of cotton, wool, paper [9], and tattoo [11]. It seeps from industrial facilities into the aquatic environment [12] and has a toxic effect [13]. MB has harmful effects on humans/ environment, and exposure to it causes encephalopathy in humans [14]. Moreover, MB has beneficial usage, such as the treatment of high hemoglobin in the case of high levels of more than 20%, which causes a problem for patients [15], the detection of Alzheimer's [16], the treatment of COVID-19 [17], useful in surgery [18], can be biologically utilized as a drug [19], and

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other uses. The toxicity of methylene blue can be removed from aquatic media using many catalysts and adsorbents such as biomass [20], bacteria [21], activated charcoal [22], treated or mixed nanometal oxides [23], reduced graphene oxide, metal oxides [24], nano carbon structure [25], and other nanomaterials such as functionalized Nano graphene with aminopropyl trimethoxysilane-phenanthrene-4-carbaldehyde (NGO@APTMS-PNTCA)[26]. MB is affected by pH (acidic or alkaline) and methylene blue has several forms in acidic and alkaline mediums (Fig. 1 and 2) [27].





Fig.1. Chemical structure of methylene blue





Methylene blue is affected by oxidation and reduction and gives two electrons in a highly acidic medium [28], and it can also be analyzed by chromatographic methods using high-performance liquid chromatography (HPLC) [29]. Cyclic voltammetry(CV) can provide kinetic and mechanistic information, the behavior of methylene blue in the electrochemical cell was studied using CV, where several parameters were determined, namely: such as diffusion coefficient (D) Equation 1 [30-32], mass transport, Equation 2 electrochemical reversibility ( $\Lambda$ ) Equation 3, Gibbs free energy ( $\Delta$ G) Equation 4, interface trap density (Dit) Equation 5, constant (K<sup>0</sup>) Equation 6, highest Occupied Molecular Orbital (HOMO) Equation 7, Lowest Unoccupied Molecular Orbital (LUMO) Equation 8 [33], thermodynamic equilibrium constants ( $K_{th}$ ) expresses the extent to which the studied substance is affected by temperature and Gibbs free energy ( $\Delta G$ ) Equation 9 [34], Electronegativity ( $\chi$ ) Equation 10, Electronic Chemical ( $\mu$ ) Equation 11, Chemical hardness () Equation 12, the electrophilicity index ( $\omega$ ) Equation 13 [35], the maximum transferred charge capacity ( $\Delta Nmax$ ), Equation 14 [34], Softness ( $\sigma$ ) Equation 15 [36] electron affinity (A), Equation 16 and The ionization energy (I) Equation 17 [37]. All Equations are shown in below Table 1.

Equations	Equations	Parameters	Parameters
$I_{p,f}^{quasi} = 0.436 \text{ n. F. A. C.} \left(\frac{nFDv}{RT}\right)^{\frac{1}{2}}$ (Eq. 1)	$\chi = \frac{-(E_{HOMO} + E_{LUMO})}{2}$ (Eq.10)	i <sub>p</sub> : Peak current (A)	A: the gate area,
$m_{\text{trans}} = \left(\frac{\pi n FDv}{RT}\right)^{\frac{1}{2}}$ (Eq.2)	$\mu = \frac{(E_{HOMO} + E_{LUMO})}{2}$ (Eq.11)	F: Faraday's constant (C. mol <sup>-1</sup> )	Cox: accumulation capacitance
$\Lambda = \frac{k^0}{\left(\frac{\pi n D F v}{RT}\right)^{\frac{1}{2}}}$ (Eq.3)	$\eta = \frac{(E_{LUMO} - E_{HOMO})}{2}$ (Eq.12)	C: Concentration (mol. cm <sup>-3</sup> )	n: Number of electrons in the redox reaction
$\Delta G = Eox - Ered - Eg + C$ (Eq.4)	$\omega = \frac{\mu^2}{2\eta}$ (Eq.13)	T: Temperature (K)	A: Electrode area (cm <sup>2</sup> )
$Dit = \frac{C_{OX} * \Delta V}{q A * Eg}$ (Eq.5)	$\Delta Nmax = -\frac{\mu}{\eta}$ (Eq.14)	v: Scan rate (V s <sup>-1</sup> )	R: Gas constant (J mol <sup>-1</sup> K <sup>-1</sup> )
$K^0 = \Lambda \cdot m_{trans}$ (Eq. 6)	$\sigma = \frac{1}{\eta}$ (Eq.15)	K <sup>0</sup> : constant	D: Diffusion coefficient (cm <sup>2</sup> . s <sup>-1</sup> )
$E_{HOMO} eV = [E_{ox} - E_{1/2} + 4.8]$ (Eq.7)	A=-ELUMO (Eq.16)	E <sub>ox</sub> : Oxidation potential	Λ: Electrochemical reversibility
$E_{LUMO} = (E_{HOMO} - Eg)$	$I = -E_{HOMO}$	Eg: Optical Bandgap	i <sub>0</sub> : exchange current density
(Eq. 8)	(Eq.17)	E <sub>red</sub> : Redaction potential	$(A m^{-2})$
$K_{th} = \exp\left(-\frac{\Delta Gad}{RT}\right)$ (Eq.9)		E <sub>1/2</sub> : Half-oxidation potential for peak	q: electron charge ΔV: flat-band voltage shift

**Table1**. The equations and parameters of kinetic and mechanistic information of Cyclic voltammetry(CV)

It is defined by the ratio of standard rate constant  $(k^0)$  to mass transfer. (K0=i0/F, C: It is the electrostatic interaction energy for the initially formed ion pair, generally, this symbol refers to the energy gap law).

This research aims to determine the concentration of methylene blue (MB) in aqueous media and its electrochemical behavior in a volt-ampere cell by CV by manufacturing an electrode from carbon paste modified with nanomaterials (NiO-NCQD, rGO,  $G-C_3N_4$ ). Many parameters related to the behavior of this pollutant were calculated using cyclic voltammetry, such as diffusion coefficient (D), mass transport (mTrans), electrochemical reversibility (A), Gibbs free energy ( $\Delta G$ ), interface trap density (Dit), constant (K<sup>0</sup>), highest Occupied Molecular Orbital (HOMO), Lowest Unoccupied Molecular Orbital (LUMO), thermodynamic equilibrium constants  $(K_{\alpha})$ , Gibbs free energy ( $\Delta G$ ), Electronegativity ( $\gamma$ ), Electronic Chemical  $(\mu)$ , Chemical hardness (), the electrophilicity index  $(\omega)$ , the maximum transferred charge capacity ( $\Delta$ Nmax), Softness ( $\sigma$ ), electron affinity (A), and the ionization energy (I). In addition, a comparison was made between the two methods using carbon paste modified by CV and UV-Vis spectrometer. The results showed that there is no difference between the two methods in terms of accuracy.

### 2. Experimental

#### 2.1. Instrumental

The voltammetry system is used for trace analysis and education. The accessories with VA computer software and all electrodes for a complete measurement system: Multi-Mode Electrode pro (MME pro), Ag/AgCl reference electrode, and Pt as auxiliary electrode were used. In this study, a modern voltammetric was connected to a PC based on a USB port (Metrohm797; volt-amperometry analyzer with analyzer cell, USA). A spectrophotometric device D-Lab model SP-UV1000 was used. Sartorius pH meter type PB-11 was used from Data Weighing System Company (pH meter and mV meter; DWS Inc.,).

#### 2.2. Reagents and Materials

Methylene Blue (MB) was purchased from Merk (CAS N.: CAS 61-73-4, Germany). The concentrated hydrochloric acid purity 37% (CAS N.: 7647-01-0, Sigma, Germany), monosodium phosphate (CAS N.: 7558-80-7, Sigma), sodium hydroxide (CAS N.: 1310-73-2), sulfuric acid with purity is 98% (CAS N.: 7664-93-9), acetic acid CH<sub>3</sub>COOH its purity is 99.5% (CAS N.: 64-19-7, Merck, Germany), the pure and solid H<sub>3</sub>BO<sub>3</sub> (CAS N.: 10043-35-3), phosphorous

acid with purity is 85% (CAS N.: 13598-36-2, Sigma) and potassium fur cyanide  $K_3$ [Fe(CN)<sub>6</sub>]. 3H2o as solid and high purity (CAS N.: 13746-66-2, Sigma) were prepared from Sigma. The quartz cells 1cm inner diameter, copper wire, small glass tubes with an inner diameter of 8 mm and a length of about 70 mm, a scale, and a micropipette were prepared.

#### 2.3. Synthesis and Characterization

Several materials were manufactured and described in previous research such as reduced graphene oxide (rGO) [38], graphene-carbon nitride (G-C<sub>3</sub>N<sub>4</sub>) [39], and nickel oxide nitrogen carbon quantum dots (NiO-NCQD) [40].

#### 2.4. Sample Preparation and Procedure

The preparation of electrode and standard solutions is as follows:

#### 2.4.1.Manufacture of the electrode body

The electrode body was made of a glass tube cut to a length of  $50\pm0.5$  mm so that it is open from both ends. Inside it is a copper wire whose lower end is connected with the modified carbon paste, and its upper end is connected to the device, the modified carbon paste consists of nickel oxide nitrogen carbon quantum dots (NiO-NCQD) of (10%) graphenecarbon nitride G-C<sub>3</sub>N<sub>4</sub> powder (10%), reduced graphene oxide (10%), graphite powder (30%), paraffin oil (40%) as a plasticizer. This electrode is used as a working electrode, it is symbolized by the symbol (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME) the measuring cell is a platinum auxiliary electrode and the comparison electrode is silver chloride silver with a constant potential of 0.222V.

#### 2.4.2. Preparation of standard solutions

First, the standard solution of MB was prepared based on 0.032 g of MB powder which was transferred completely into a volumetric flask of 100 mL distilled water to the capacity mark solution with a concentration of 10 mM. A solution of monosodium phosphate modified with phosphorous acid based on a buffer solution of (0.1M) monosodium phosphate NaH<sub>2</sub>PO<sub>4</sub> was prepared and a solution of phosphorous acid (H<sub>3</sub>PO<sub>4</sub>; 1.08 M) was added to it until the PH changed to pH=1. For the preparation of the sulphur acid solution, a solution of (0.1M) sulphur acid is prepared by taking 0.2776 mL of concentrated sulfuric acid with a purity of 98% and a density of 1.84 g cm<sup>-3</sup> for a volume of 50 mL distilled water (DW) up to the capacity mark, and we get the pH of

1. For modified Britton-Robinson Buffer solution (BRB), a buffer solution is prepared from BRB, which consists of  $H_{3}PO_{4}$  (0.2076M) with acetic acid (CH,COOH; 0.04M) and boron acid (H,BO,; 0.04M) concentrations on the pH value of 1. For the preparation of phosphorous acid solution, a solution of 1.08M of phosphorous acid was prepared by taking 3.64 mL of concentrated phosphorous acid with a purity is 85% and density of 1.71 g cm<sup>-3</sup> for a volume of 50 mL and it was supplemented with distilled water (DW) until the capacity mark reached to the pH value 1. Finally, a solution of hydrochloric acid 0.1M was prepared by taking 0.414 mL of concentrated hydrochloric acid based on a purity of 37% and density of 1.19 g cm<sup>-3</sup> in 50 mL of distilled water (DW) up to the capacity mark to get a pH value 1.

#### 2.4.3. Test solution for electrode

Weigh approximately 0.0329 g of  $K_3[Fe(CN)_6]$  and 0.186 g of KCl. Then, transfer completely to a 25mL of DW to a concentration solution of  $K_3[Fe(CN)_6]$  (5Mm) and 0.1 M of KCl.

# 2.4.4.Preparation of solution for

# spectrophotometric measuring

First, 0.025 g of MB powder was transferred completely to a volumetric flask of 25 mL capacity and the volume was completed with distilled water (DW), so the concentration is 1000 mg L<sup>-1</sup> (1.0 g L<sup>-1</sup>) was made then, the different standard solution samples of 1, 2, 4, 6, 8, 10 mgL<sup>-1</sup> was prepared from it by dilution with DW.

# 3. Results and Discussion

The prepared electrode was tested by test solution, to ensure the work of the prepared electrode before starting the measurement procedure, as shown in Figure 3A. It was found that the electrode worked and was reliable, identified the contaminant methylene blue, and determined the optimal conditions for work, including pH, scanning speed, and scanning rate.

# 3.1. The effect of pH

The effect of the pH value was studied within the pH range from 1 to 10, where the scanning was done within a potential scanning range (-1 to +1) V. At a range of pH (5-10) has no peak, while a redaction peak appeared within the pH range of 1 to 4. Also, an oxidation peak has appeared within the range of pH (1-2). The results of pH based on NiO-NCQD/g- $C_3N_4$ /rGO/CPME electrode are shown in Figures 3A-3M.











C) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 9



D) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 8



E) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 7



F) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 6



G) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 5



H) Oxidation curve of methylene blue oxidation and reduction of 0.5 mM methylene blue at pH = 4



I) Oxidation curve of methylene blue oxidation and reduction of 0.5m M methylene blue at pH = 3



J) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 2



K) Effect of pH on the value of the oxidation current strength and the return of a 0.5 mM MB



L) Oxidation curve of methylene blue oxidation and reduction of 0.5mM methylene blue at pH = 1



M) Effect of the type and nature of the medium used at pH = 1 on the oxidation and redaction current of a 0.25 mM methylene blue solution where, A: monosodium phosphate with phosphorous acid, B: 0.1M sulfur acid, C: modified (BRB), D: Phosphorous acid, E: Hydrochloric acid 0.1M.

**Fig. 3.**Curves by electrode (NiO-NCQD/g-C3N4/rGO/CPME) from A to M.

The previous curve shows the oxidation peak at a potential value of 0.585V and a redaction peak at a potential value of -0.21V, so the practical potential difference is  $\Delta E=0.795V$ , which is a value greater than  $\Delta E=0.059/2=0.0295V$  using the manufactured electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME), it is a semi-reversible curve subjected to oxidation equation in the middle (pH=1), according to the following oxidation and redaction Equation. The effect of the type of medium was evaluated. Several media were used at pH 1, which are hydrochloric acid, sulfuric acid, phosphorous acid, modified peritoneal, and monosodium phosphate modified with phosphorous acid which is shown in Figures 3L and 3M. It was found from the previous curve that the best acid used was 0.1M hydrochloric acid.

#### 3.2. The effect of scan rate

The scan rate was studied within the range of 10, 30, 50, 70, and 100 mv sec<sup>-1</sup> on the peak current  $I_{(p)}$  as shown in Figures 4A and 4B, and it was observed that it is the best scan rate is100 mv sec<sup>-1</sup>.



**Fig. 4.** Effect of scan rate (10-30-50-70-100) mv/sec on a 0.25Mm methylene blue solution at pH=1 at 0.1M hydrochloric acid by electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME)

From the scanning rate in Figures 3 and 4, it is concluded that the relationship between the potential scanning rate  $(mv \ sec^{-1})$  and the peak current intensity I ( $\mu$ A) for both oxidation and reduction is linear. Also, it showed a directly proportional to the value of the oxidation peak current intensity, and inversely proportional to the value of the reduction peak current intensity. Whereas the value of the correlation coefficient was obtained at R<sup>2</sup>=0.9938 and R<sup>2</sup>=0.9019 for both oxidation and reduction, respectively. Also, the equation of the line for each of them is y=5.0082x+80.574 and y=-3.116x-194.37 for both oxidation and reduction, respectively.

#### 3.3. Analytical application

A standard solution was prepared from Methylene blue within the range (25-100)  $\mu$ M or (7.99-31.98) mg L<sup>-1</sup> (Fig. 5A), which was studied at (pH=1) of (0.1M) hydrochloric acid in water samples. All samples were measured using an electrochemical method by Cyclic voltammetry (CV) method based on the electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME). The standard curve for the oxidation and reduction of methylene blue in terms of concentration and the current strength of the oxidation and reduction peaks is shown in Figure 5B. Due to Figure 5B, it was found that the concentration of methylene blue can be determined within the linear range (25-100)  $\mu$ M or (7.99-31.98) mg L<sup>-1</sup>, where the oxidation equation was  $I_{Ox} = 1.508C_{MB} + 229.5$  and the correlation coefficient (R<sup>2</sup>=0.9823), while the redaction equation is  $I_{Red} = -2.236C_{MB} - 232.5$  and the correlation coefficient (R<sup>2</sup>=0.9722).

# 3.4. Studying the behavior of methylene blue in aqueous solutions

The behavior of methylene blue has been studied in aqueous solutions for a range of concentrations of methylene blue (25-50-75-100)  $\mu$ M using the proposed electrode and at (pH=1) using (0.1M) hydrochloric acid. In this study, diffusion coefficient (D), mass transport (m<sub>trans</sub>), constant (K<sup>0</sup>), electrochemical reversibility (), and interface trap density (D<sub>it</sub>) were shown in Table 2. The values in Table 2 are plotted and shown in graphical curves in Figure 6. Effect of the concentration of methylene blue using electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/ CPME) on Diffusion coefficient (D), Mass transport (m<sub>trans</sub>), Constant (K<sup>0</sup>), Electrochemical reversibility (A), and interface trap density (D<sub>it</sub>) were shown in Figure 6.



Fig. 5. A standard series of methylene blue (25-50-75-100)μM at PH=1 using 0.1M hydrochloric acid by electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME)

СµМ	Form	$D * 10^9$ $(m^2 \cdot s^{-1})$	m <sub>trans</sub>	K <sup>0</sup>	Λ * 1000	Dit eV <sup>-1</sup> cm <sup>-2</sup>
		(m · s )				
25	_	177.4497	0.0021	2.1537E-06	0.9895	3.1609E+15
50	atior	63.8003	0.0013	2.5828E-06	1.9791	1.8953E+15
75	Oxid	34.4500	0.0009	2.8468E-06	2.9687	1.3927E+15
100	·	23.0170	0.0007	3.1026E-06	3.9583	1.1384E+15
25	ч	199.8728	0.0023	-2.2857E-06	-0.9895	3.3547E+15
50	Iction	83.46421	0.0014	-2.9541E-06	-1.9792	2.1678E+15
75	Sedu	47.94469	0.0011	-3.3584E-06	-2.9688	1.6431E+15
100	4	32.53039	0.0009	-3.6885E-06	-3.9584	1.3534E+15

 Table 2. The values of the diffusion constant, the transmitted mass, the velocity constant, and the reflection coefficient of the oxidation and reflux peaks, Methylene blue in aqueous by electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME)



Fig.6. Effect of the concentration of methylene blue using the electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME) on different parameters. A)Diffusion coefficient (D), B) Mass transport (m<sub>trans</sub>), C) Constant (K<sup>0</sup>),
 D) Electrochemical reversibility, E) Interface trap density (D<sub>i</sub>)

The diffusion coefficient and the mass transport to the electrode surface decreased with the increase in the concentration of methylene blue in aqueous media. Also, some physical and chemical properties were calculated in Table 3.

It is concluded from the value of the chemical voltage that the electrons in the molecule (MB) need an energy of (3.8842 eV) in order to exit the equilibrium system, and the molecule receives an additional electronic charge from its surroundings within its studied system of (-12.905 eV) from the value of the electrophilicity index, the molecule has a resistance to charge transfer of (-1.7107 eV) from the value of chemical hardness, and the compound is an electron acceptor since the value of the maximum charge capacity is positive, and the compound has an energy difference between the vacuum energy level and the minimum level of the conduction band, its value is (-2.17351 eV) of electronic affinity, and the reaction is spontaneous from the free energy of gypsum, this indicates that the oxidation of (MB) Spontaneous oxidation in the studied solution in the presence of an electric current (generated by the auxiliary stream), and the solution of this pollutant is thermodynamically balanced at Laboratory temperature and normal pressure, knowing that the energy gap value is equal to (3.42149 eV).

# 3.5. Comparison of methylene blue analysis using the proposed electrode with the Cyclic voltammetry method and the spectrophotometric method

It is studied spectroscopically in the visible field (VIS), where standard solutions of methylene blue solutions including 1, 2, 4, 6, 8, and 10 mg  $L^{-1}$  were prepared and then scanned spectroscopically as in Figure 7A. It appears from the previous curve that the maximum absorption value of Methylene blue was observed at 663nm, which has a high colour intensity, and the absorption curve in terms of concentration was shown in Figure 7B.

**Table 3**. Some properties and constants of physical chemistry of the electrochemical behavior of methylene blue organic pollutant (MB) within the electrochemical cell based on cyclic amperometric voltage using a probe (NiO-NCQD/g- $C_3N_4$  / rGO/ MCPE)

(HOMO)	5.595eV	(K <sub>th</sub> )	1.001
(LUMO)	2.1732ev	$(\Delta N_{max})$	2.271eV
( <b>Eg</b> )	3.421eV	(σ)	-0.585eV
$(\Delta \boldsymbol{G})$	-2.626 KJ mol <sup>-1</sup>	<b>(I</b> )	-5.595eV
(χ)	-3.884eV	(A)	-2.173eV
(ω) (η)	-12.905eV -1.7107eV	(μ)	3.884eV



Fig. 7. Curve for methylene blue (1,2,4,6,8,10) mg L-1A) Scanning spectroscopy of wavelength and absorbanceB) concentration vs absorbance

Metho	ods	Standard concentration	$Xn = 3$ $(mgL^{-1})$	SD (mgL <sup>-1</sup> )	RSD (%)	R (%)	LOQ (mgL <sup>-1</sup> )	LOD (mgL <sup>-1</sup> )
A	Ox	7.00	7.97	0.36	4.52	99.64	0.24	0.07
	Red	(mgL <sup>-1</sup> )	7.97	0.29	3.69	99.70	0.13	0.04
UV-Vis	SIV		8.22	0.25	3.07	102.87		

**Table 4.** Comparison of the determination of Methylene blue using the electrode (NiO-NCQD/g-C<sub>3</sub>N<sub>4</sub>/rGO/CPME) with the spectrophotometric method in the visual field at 663nm

Proposed Method: (NiO-NCQD/g-C $_3N_4$ /rGO/CPME) with CV Spectrophotometry: UV-Vis

From the previous curve, it appears that Methylene Blue has an Equation of  $C_{MB}$ =6.305×A+0.3467 and a correlation coefficient of R<sup>2</sup>=99.2712.

The proposed method was applied to a standard sample of 7.99 mg L<sup>-1</sup> concentration in two ways, comparing of spectroscopic method with the CV method based on NiO-NCQD/G-C<sub>3</sub>N<sub>4</sub>/ rGO/ MCPE, to ensure the validity and accuracy of the proposed method by calculating the statistical parameters which was shown in Table 4. It was also calculated by the value of  $F_{ex}$ . So, the value of  $F_{ex}$  was 2.04 for oxidation and 1.35 for reduction which is smaller than the value of  $F_{tab}$ =19.0 as a confidence level of 95% with  $\alpha = 0.05$  and n = 3. Therefore, there is no significant difference between the two methods.

## 4. Conclusions

This paper deals with an electrochemical method for the determination of methylene blue (MB) by fabrication of an electrode (NiO-NCQD/G-C<sub>2</sub>N<sub>4</sub>/ rGO/CPME), This electrode is used as a working electrode. The oxidation-reduction curve of methylene blue was shown using this electrode. It is a quick-reversible curve, and it works at (pH=1) and the best acid used is HCl a concentration of (0.1M). It is found that the linear range is within the range of 25-100 µM. Several constants were studied to determine the behavior of methylene blue within the electrochemical cell. The CV method was compared to spectrophotometry at 663nm and there is no difference between the determination of MB by the CV and spectrophotometry methods.

#### **5. References**

 I. Khan, K. Saeed, I. Zekker, B. Zhang, A.H. Hendi, A. Ahmad, I. Khan, Review on methylene blue: its properties, uses, toxicity and photodegradation, Water, 14 (2022) 242.

#### https://doi.org/10.3390/w14020242

- [2] H.N. Hamad, S. Idrus, Recent developments in the application of bio-waste-derived adsorbents for the removal of methylene blue from wastewater: A review, Polymers, 14 (2022) 783. https://doi.org/10.3390/polym14040783
- [3] T. Lupascu, M. Ciobanu, E. Culighin, Absorption of methylene blue from aqueous Solutions on activated coal CAN-9: kinetics and equilibrium studies, Roman. J. Ecol. Environ. Chem., 4 (2022) 22-28. https://doi. org/10.21698/rjeec.2022.102
- [4] A. Miclescu, L. Wiklund, Methylene blue, an old drug with new indications. J. Rom. Anest. Terap. Int., 17 (2010) 35-41. http:// www.jurnalul-anestezie.ro/2010/h-Miclescu\_ Methylene%20blue.pdf
- [5] E.M. McDonagh, J.M. Bautista, I. Youngster, R.B. Altman, T.E. Klein, Pharm GKB summary: methylene blue pathway. Pharmacogenet. Genom., 23 (2013)498. https://doi.org/10.1097/ FPC.0b013e32836498f4
- [6] C. Stack, S. Jainuddin, C. Elipenahli, M. Gerges, N. Starkova, A.A. Starkov, M. Dumont, Methylene blue upregulates Nrf2/ARE genes and prevents tau-related neurotoxicity, Hum. Mol. Genet., 23 (2014) 3716-3732. https://doi.org/10.1093/hmg/ddu080
- M. Hassanpour, H. Safardoust-Hojaghan, M. Salavati-Niasari, Degradation of methylene blue and Rhodamine B as water pollutants via green synthesized Co<sub>3</sub>O<sub>4</sub>/ZnO nanocomposite, J. Mol. Liq., 229 (2017) 293-299. https://doi.org/10.1016/j.molliq.2016.12.090
- [8] V.K. Yadav, N. Choudhary, D. Ali, G. Kumar, G. Gnanamoorthy, A.U. Khan, B.Z. Tizazu, Determination of adsorption of methylene blue dye by incense stick ash waste and its toxicity on RTG-2 cells, Adsorp. Sci.

Technol., 2022 (2022) 8565151. https://doi. org/10.1155/2022/8565151

- [9] Z. Reçber, Adsorption of methylene blue onto spent Alchemilla vulgaris leaves: Characterization, isotherms, kinetic and thermodynamic studies. Inter. J. Environ. Sci. Technol., 19 (2022) 4803–4814. https://doi. org/10.1007/s13762-022-04053-7
- [10] S.M. Merdas, W. Al-Graiti, A. Al-Ameer, Using PVA@ WNS composite as adsorbent for methylene blue dye from aqueous solutions, J. Med. Chem. Sci., 5 (2022) 1289-1298. https:// doi.org/10.26655/JMCHEMSCI.2022.7.18
- [11] A.E. Manlove, A.M. Linnebur, Primary bilateral cleft lip repair using the modified millard technique, Atlas Oral Maxillofac. Surg. Clin., 30 (2022) 19-25. https://doi.org/10.1016/j. cxom.2021.11.005
- [12] C. Luo, S. Wang, D. Wu, X., Cheng, H. Ren, UV/Nitrate photocatalysis for degradation of Methylene blue in wastewater: Kinetics, transformation products, and toxicity assessment, Environ. Technol. Innov., 25 (2022) 102198. https://doi.org/10.1016/j. eti.2021.102198
- [13] K. Duan, T. Que, S. Koppala, R. Balan, B. Lokesh, R. Pillai, S. Munusamy, A facile route to synthesize n-SnO2/p-CuFe<sub>2</sub>O<sub>4</sub> to rapidly degrade toxic methylene blue dye under natural sunlight, RSC Adv., 12 (2022) 16544-16553. https://doi.org/10.1039/D2RA01690G
- [14] P.K. Gillman, CNS toxicity involving methylene blue: the exemplar for understanding and predicting drug interactions that precipitate serotonin toxicity, J. Psychopharm., 25 (2011) 429-436. https://doi. org/10.1177/0269881109359098
- [15] S.H. Mun, G.J. Park, J.H. Lee, Y.M. Kim, H.S. Chai, S.C. Kim, Two cases of fatal methemoglobinemia caused by self-poisoning with sodium nitrite: A case report, J. Med., 101 (2022) e28810. https://doi.org/10.1097/ MD.000000000028810
- [16] M. Oz, D.E. Lorke, G.A. Petroianu, Methylene blue and Alzheimer's disease, Biochem. Pharmacol., 78 (2009) 927-932. https://doi. org/10.1016/j.bcp.2009.04.034
- [17] A. Alemany, P. Millat-Martinez, M. Corbacho-Monné, P. Malchair, D. Ouchi, A. Ruiz-Comellas, E. Grau, High-titre methylene blue-treated convalescent plasma as an early treatment for outpatients with COVID-19: a randomised, placebo-controlled trial, Lancet

Respir. Med., 10 (2022) 278-288. https://doi. org/10.1016/S2213-2600(21)00545-2

- [18] C. Jamshidzadeh, A new analytical method based on bismuth oxide-fullerene nanoparticles and photocatalytic oxidation technique for toluene removal from workplace air, Anal. Methods Environ. Chem. J. 2 (2019) 73-86. https://doi.org/10.24200/amecj.v2.i01.55
- [19] J.P. Tardivo, A. Del Giglio, C.S. De Oliveira, D.S. Gabrielli, H.C. Junqueira, D.B. Tada, M.S. Baptista, Methylene blue in photodynamic therapy: From basic mechanisms to clinical applications, Photodiagnosis Photodyn. Ther., 2 (2005) 175-191. https://doi.org/10.1016/ S1572-1000(05)00097-9
- [20] M.T. Carneiro, A.Z. Barros, A.I. Morais, A.L. Carvalho Melo, R.D. Bezerra, J.A. Osajima, E.C. Silva-Filho, Application of water hyacinth biomass (Eichhornia crassipes) as an adsorbent for methylene blue dye from aqueous medium: Kinetic and isothermal study, Polymers, 14 (2022) 2732. https://doi.org/10.3390/ polym14132732
- [21] M.M. Asl, N. Mansouri, Functionalized graphene oxide with bismuth and titanium oxide nanoparticles for efficiently removing formaldehyde from the air by photocatalytic degradation-adsorption process, J. Anal. Test., 7 (2023) 444-458. https://doi.org/10.1007/ s41664-023-00272-0
- [22] A. El-Monaem, M. Eman, A.M. Omer, G.M. El-Subruiti, M.S. Mohy-Eldin, A.S. Eltaweil, Zero-valent iron supported-lemon derived biochar for ultra-fast adsorption of methylene blue, Biomass Convers. Bior., 14 (2024) 1697– 1709. https://doi.org/10.1007/s13399-022-02362-y
- [23] V.Perumal, C. Inmozhi, R. Uthrakumar, R. Robert, M. Chandrasekar, S.B. Mohamed, K. Kaviyarasu, Enhancing the photocatalytic performance of surface-Treated SnO<sub>2</sub> hierarchical nanorods against methylene blue dye under solar irradiation and biological degradation, Environ. Res., 209 (2022) 112821. https://doi.org/10.1016/j.envres.2022.112821
- [24] A. Nisar, M. Saeed, M. Muneer, M. Usman, I. Khan, Synthesis and characterization of ZnO decorated reduced graphene oxide (ZnO-rGO) and evaluation of its photocatalytic activity toward photodegradation of methylene blue, Environ. Sci. Pollut. Res., 29 (2022) 418-430. https://doi.org/10.1007/s11356-021-13520-6
- [25] S. Teimoori, New extraction of toluene from

water samples based on nano-carbon structure before determination by gas chromatography, Int. J. Environ. Sci. Technol., 20 (2023) 6589-6608. https://doi.org/10.1007/s13762-023-04906-9

- [26] A.H. Hassani, M. Panahi, N. Mansouri, An immobilization of aminopropyl trimethoxysilane-phenanthrene carbaldehyde on graphene oxide for toluene extraction and separation in water samples, Chemosphere, 316 (2023)137800. https://doi.org/10.1016/j. chemosphere.2023.137800
- [27] O. Blank, E. Davioud-Charvet, M. Elhabiri, Interactions of the antimalarial drug methylene blue with methemoglobin and heme targets in Plasmodium falciparum: a physico-biochemical study, Antioxid. Redox Signal., 17 (2012) 544-554. https://doi.org/10.1089/ars.2011.4239
- [28] S. Roldán, M. Granda, R. Menéndez, R. Santamaría, C. Blanco, C. Supercapacitor modified with methylene blue as redox active electrolyte, Electrochim. Acta, 83 (2012) 241-246. https://doi.org/10.1016/j. electacta.2012.08.026
- [29] N. Martin, Y. Leprince-Wang, HPLC-MS and UV–Visible coupled analysis of methylene blue photodegradation by hydrothermally grown ZnO nanowires, Phys. Status Solidi A, 218 (2021) 2100532. https://doi.org/10.1002/ pssa.202100532
- [30] M.C. Henstridge, E. Laborda, E.J. Dickinson, R.G. Compton, Redox systems obeying Marcus–Hush–Chidsey electrode kinetics do not obey the Randles–Ševčík equation for linear sweep voltammetry, J. Electroanal. Chem., 664 (2012) 73-79. https://doi.org/10.1016/j. jelechem.2011.10.015
- [31] J.E.A. Randles, Cathode ray polarograph, Part II, the current-voltage curves, Trans. Faraday Soc., 44 (1948) 327-338. https://doi. org/10.1039/TF9484400327
- [32] A. García-Miranda Ferrari, C.W. Foster, P.J. Kelly, D.A. Brownson, C.E. Banks, Determination of the electrochemical area of screen-printed electrochemical sensing platforms, Biosensors, 8 (2018) 53. https://doi. org/10.3390/bios8020053
- [33] K.I. Alabid, H.N. Nasser, Study of the behavior and determination of phenol Based on modified carbon paste electrode with nickel oxidenitrogen carbon quantum dots using cyclic voltammetry, Anal. Methods Environ. Chem. J., 6 (2023) 58-68. https://doi.org/10.24200/

amecj.v6.i01.227

- [34] M.R. Jalali Sarvestani, R. Ahmadi, B. Farhang Rik, Procarbazine adsorption on the surface of single walled carbon nanotube: DFT studies, Chem. Rev. Lett., 3 (2020) 175-179. https://doi.org/10.22034/CRL.2020.110451
- [35] O.M.E.R. Rebaz, P. Koparir, L. Ahmad, M. Koparir, Computational determination the reactivity of salbutamol and propranolol drugs, Turk. Comput. Theor. Chem., 4 (2020) 67-75. https://doi.org/10.33435/tcandtc.768758
- [36] M.J. Sani, Spin-orbit coupling effect on the electrophilicity index, chemical potential, hardness and softness of neutral gold clusters: A relativistic ab-initio study, High Tech Innov. J., 2 (2021) 38-50. https://doi.org/10.28991/ HIJ-2021-02-01-05
- [37] S. Erkan, Structural, spectroscopic and anticancer properties of hydroxy-and sulfonamideazobenzene platinum (II) complexes: DFT and molecular docking studies, Cumhuriyet Sci. J., 39 (2018) 1036-1051. https://doi. org/10.17776/csj.421027
- [38] K.I. Alabid, H.N. Nasser, H.K. Maleh, Reduction of graphene oxide by new chemical and green methods, J. Ultrafine Grained Nanostruct. Mater., 55 (2022) 172-185. https:// doi.org/10.22059/jufgnsm.2022.02.09
- [39] K.I. Alabid, H.N. Nasser, Synthesis and charcteriztion grapheme-carbon nitride nanostructure in one step, Ibn Al-haitham j. Pure Appl. Sci., 36 (2023) 260-272. https://doi. org/10.30526/36.3.3103.
- [40] K.I. Alabid, H.N. Nasser, Synthesis and characterization of Nickel Oxide with Nitrogen quantum carbon dots as nanoadsorbent (NiO-NCQD) nanocomposite. Int. J. Nano Dimension, 14 (2023) 227-237. https://doi. org/10.22034/ijnd.2023.1984570.2217



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# Chromium desalinization using novel chitosan functionalized iron oxide- biochar composites: Analysis, synthesis, characterization and adsorption performance

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### A B S T R A C T

In the study, chitosan functionalized iron oxide incorporated with peanut shell biomass was prepared for potential adsorption of chromium (VI) from an aqueous media. The prepared material was characterized by modern spectroscopic methods for confirming the successful embedding. The adsorption experiments were conducted in batch systems. The experimental data showed robust removal of chromium supported by kinetic and equilibrium studies. The sorption data exhibited a strong agreement with the pseudo-second-order kinetics model, further confirming conformity with the Langmuir isotherm model. Adsorption studies were taken to find the effects of pH and time, reusability, ionic strength and presence of coexisting ions. The maximum sorption capacity was achieved as 14.28 mg g<sup>-1</sup> at pH 4 and the optimum contact time was 40 minutes. The background electrolytes have much less effect on uptake efficiency and this green adsorbent can be utilized for up to four cycles. Additionally, a systematic approach was employed to ensure the precision and accuracy of the spectroscopic method. Calibration was linear in the range from 0.5 to 6.0  $\mu$ g L<sup>-1</sup> (R<sup>2</sup> > 0.99). The limits of detection (LOD) and quantification (LOQ) were 0.65  $\mu$ g L<sup>-1</sup> and 2.16  $\mu$ g L<sup>-1</sup>, respectively. The relative standard deviation (RSD) was 7.62 % (n=7). The method accuracy was verified by analyzing recovery (92.8  $\pm$  0.5% to 103.09  $\pm$  0.5 %.) studies on water samples. In conclusion, novel chitosan functionalized Iron oxide-biochar composites find unbeatable, recyclable and highly efficient adsorption properties for chromium desalinization.

# **1. Introduction**

Chromium, a versatile and widely used metal, has found its way into various industrial processes, from steel production to electroplating. However, industrial waste often contains chromium heavy metal ions as a byproduct of these activities. These ions, primarily in the form of hexavalent chromium (Cr (VI), pose significant toxicity to aquatic life and are harmful to human health. Understanding the sources, characteristics, and

\*Corresponding Author: Anurag Choudhary Email: anurag051981@gmail.com https://doi.org/10.24200/amecj.v6.i04.281 potential hazards of chromium heavy metal ions from industrial waste is crucial for effectively managing and remedying these pollutants. Effective disposal of chromium from wastewater is essential to protect the environment, and human health, and ensure compliance with global regulations while promoting sustainable water management. Ecological elimination [1], chemical precipitation [2], redox [3] and adsorption [4] are only some of the methods that have been used for Cr (VI) removal so far. Adsorption is considered the best and most widely used approach for eliminating toxic contaminants due to its inexpensive, effortless, and highly influential capability. Experiments have

used a wide range of adsorbing materials, including biochar, chitosan, and iron oxide, to remove Cr (VI). The peanut (Arachis hypogaea L.) is an ancient crop that is widely cultivated in tropical and subtropical climates. India holds the position of being the second-largest producer of peanuts globally, following China. Peanut shells are the remainder of the initial processing of peanuts [5]. The peanut shell is a substantial byproduct of the peanut industries, constituting around 25% to 30% of the legume's total weight. These shells are discarded as such as waste after the removal of groundnut seed from its pod at the last phase of peanut processing. These are the abundant agro-industrial waste product which has a very slow degradation rate under natural conditions [6]. The peanut shell is a lignocellulosic material mainly composed of cellulose (44.8%), hemicelluloses (5.8%), and lignin (36.1%), which are complex organic polymers. To successfully manage and control this extremely troublesome detrimental species, it may be unique and enticing to convert shells into biochar and utilize them in eliminating cationic toxicants. Biochar produced from waste biomass finds varied applications in diversified realms. In agriculture, it stimulates soil fertility, water retention, and nutrient efficiency. As a stable carbon sink, biochar contributes to climate change mitigation by sequestering carbon in the soil for many years. It also acts as a water filtration medium, removing contaminants from wastewater. Its adsorption properties make it useful for environmental remediation efforts, and it can even be used as a renewable energy source [7]. Additionally, recent research has shown that biochar may be utilized as a thrifty sorbent to remove a variety of toxins from water. The biochar possesses several desirable characteristics, including a substantial specific surface area, a porous structure, an abundance of surface functional groups, and mineral components. These properties render biochar a suitable adsorbent for the removal of pollutants from aqueous solutions. Enhancements and treatment methods applied to biochar customization result in improved adsorption capabilities compared to its pristine form. By modifying, its surface properties can be tailored to target specific contaminants, making it even more effective for adsorption purposes. Consequently, а range of techniques, including chemical functionalization, physical activation, impregnation, coating, and surface oxidization, have been employed to enhance the efficacy of these materials in the context of environmental remediation [8]. Chitosan, a biopolymer derived from chitin, has gained remarkable attention and intensively studied for its potential application in heavy metal removal due to its unique properties such as non-toxic nature, abundant, remarkable adsorption capacity, biocompatibility, and budget-friendly substance [9]. The existence of abundantly functioning groups, specifically amine and hydroxyl, in the polymer's scaffold facilitates active receptor sites to adsorb metal ions more proficiently. Chitosan possesses a unique structure that includes high-functioning groups, such as amino (-NH<sub>2</sub>) and hydroxyl (-OH) groups, in its backbone. These functional groups serve as active sites for adsorption. The amino groups can act as Lewis bases, capable of forming coordination complexes with metal ions through ion exchange or chelation processes. The hydroxyl groups, on the other hand, contribute to the adsorption through electrostatic interactions, hydrogen bonding, and surface complexation [10]. Although chitosan-modified biochar has a higher adsorption capacity, it is necessary to perform time-consuming post-treatment activities such as sedimentation, centrifugation, and filtering to collect the solid particles that were filtered out of the treated water. Since it takes a long time for the lowdensity particles to completely settle, the recovery of these tiny particles from the huge industrial waste treatment tank becomes difficult. Therefore, the introduction of magnetic characteristics to the biochar-based adsorbent may further alleviate the challenging recovery of the biochar-based adsorbents from the water bodies. Magnetic impregnation of the biochar would increase the composite's reusability after modification. Many recent investigations have synthesized flexible composite adsorbents by combining magnetic particles and chitosan with biomass. As a bonus, the introduction of magnetic species may attract additional metal ions owing to its superior conductivity, making magnetic separation a win-win technique for recovering adsorbent from an aqueous medium. Magnetic biochar and chitosan, when combined, provide an excellent composite for cleaning up secondary pollution by removing a wide range of metals [11]. The current research on the utilization of chitosan for surface modification of biochars to improve their ability to apprehend heavy metals is still limited. This combination shows enormous potential as it effectively utilizes the perks of biochar and chitosan, resulting in significant benefits for binding Cr (VI). Fe<sub>2</sub>O<sub>2</sub> particles are very useful in the treatment of wastewater due to their accessible cost and ease of separation. They have been used to effectively remove heavy metals and organic pollutants [12].

Biomass adsorbents are widely applied as secret weapons for the sustainable adsorption of environmental toxicants. The green sorbents prepared by biomass are invaluable tools for unlocking the power of waste by reducing the cost of remediation techniques for safe water. This research explores the efficacy of removing Cr (VI) using crosslinked chitosan hydrogel immobilized with magnetic biochars created from peanut shells by pyrolytic technique. Scanning electron microscopy (SEM), Fourier transform infrared (FTIR) analysis, X-ray photoelectron spectroscopy (XPS), and (Brunauer- Emmett-Teller) BET analysis were all collectively used to characterize the generated material. The adsorption property of chitosan-modified magnetic biochar for Cr (VI) in aqueous solution was also studied by examining the effects of pH, kinetics, and sorption isotherms.

## 2. Material and methods

#### 2.1. Materials

Chitosan powder (85 % acetylation degree) was purchased from Alfa Aesar suppliers in, the USA. (Lot No A2041604). Peanut shells were collected from locally available fields to make pristine biochar. Ferric chloride hexahydrate, Glutaraldehyde (50 % V/V), acetic acid, Sodium hydroxide, potassium dichromate and 1,5 diphenyl carbazide (DPC) were purchased from Fisher Scientific, India. The solution was prepared using double-distilled water, and all of the chemicals utilized in the experiment were of the analytical reagent grade.

# **2.2.** Incorporation procedure of $Fe_2O_3$ -biochar composite

The collection of peanut shells and synthesis of biochar pyrolytically was referred to in our previously published literature [13], herein, slight modifications were applied to obtain a magnetic biochar composite. According to the standard procedure, the dried peanut shells were crushed into micron-sized particles in a domestic mixer grinder before being sieved via a sieved shaker machine to produce a 70-80 mesh screen. The next step included soaking 25 grams of totally dried and powdered biomass in 80 mL of a 2M FeCl, solution. For 0.5 hours, the solution was continuously stirred using a magnetic stirrer at room temperature. After that, the mixture was aged for an additional 0.5 hours at 70 °C to speed up hydrolysis and Fe<sup>3+</sup> precipitation. After that, the pre-treated biomass sample was placed in an alumina crucible and heated in a vacuum-applied muffle furnace at temperatures of 250, 350, 450, and 550 °C with rises

of 10 °C per minute and retention times of 10 minutes at each level while  $N_2$  was continuously pumped in at a rate of 20 mL min<sup>-1</sup>. The sample was kept at the same temperature after reaching 550 °C for two hours in an oxygen-free atmosphere. After finishing the process, the furnace's temperature naturally dropped to room temperature. The dried material was then ground and sieved to produce magnetic biochar that had a particle size of less than 0.5 mm. The dedicator held the prepared material. The finished product is known as a PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>2</sub> composite (yield 38.5%).

# 2.3. Preparation of chitosan functionalized Fe<sub>2</sub>O<sub>3</sub>-biochar composite

Chitosan powder (8.0 g) was first dissolved in 2% (v/v) acetic acid (500 mL) to obtain a homogenized viscous chitosan solution under continuous stirring for 2 h at 50 °C. Afterwards, 4 g of PSB/y-Fe<sub>2</sub>O<sub>2</sub> composite was added to 50 ml of chitosan solution and the compounds were reacted in the ultrasonic bath for 30 min at 40 °C. 10 mL glutaral dehyde (25 %in H<sub>2</sub>O) was then injected dropwise very slowly into the reaction system. After 30 min, keeping at 40 °C, NaOH solution (0.1 M) was added dropwise into the mixtures until the pH value reached 9. The solution was then allowed to stand as such in a Petri dish and undisturbed for the next 24 hours. A soft cross-linked three-dimensional network structure of chitosancoated biochar magnetic hydrogel was formed and the solution turned light yellow to brownish yellow. The prepared chitosan hydrogel was then thoroughly washed with milli-Q water several times to remove any unreacted monomer. The final product was named CH@PSB- $\gamma$ -Fe<sub>2</sub>O<sub>2</sub> composite. The detailed preparation process is shown in Figure 1.

#### 2.4. Characterization methods

Standard spectroscopic methods were leveraged to explore the structural peculiarities of the synthesized material. Fourier-transform infrared spectroscopy (FTIR) (Perkin Elmer) with a working range of 4000-400 cm<sup>-1</sup> provides verified existence of abundant functional groups and some morphological alteration resulting from the incorporation of chitosan onto the surface of magnetic biochar. Exterior aesthetics and microstructure were analyzed by Scanning electron microscope (Carl Zeiss, Germany, model: Zeiss Gemini 300). The crystalline structure was examined by an X-ray diffractometer (Panalytical XPERT-PRO, UK) with  $2\Theta$  range from 5° to 98° at a wavelength of 1.54059 A<sup>0</sup>. Energy dispersive X-ray spectrum (EDS) was obtained for elemental compositions as a crucial tool for unravelling materials' composition



Fig. 1. Schematic presentation for synthesizing CH@PSB/ γ-Fe<sub>2</sub>O<sub>3</sub>

and structural characteristics, shedding light on their chemical makeup and potential interactions. XPS spectra (Scienta omicron, Germany,) furnished with Al K<sub>a</sub> X-ray source (1486.7 eV) in ultra-high vacuum were used to determine the surface composition, relative abundance of these compositions and surface chemical structure of the prepared adsorbent. The point of zero charges (PZC) is the location where the total amount of positive and negative charges on the surface of a substance is equalized. The surface exterior gets negatively charged above the PZC and positively charged below it. The surface area and pore size distributions of PSB/y-Fe<sub>2</sub>O<sub>2</sub> and CH@PSB/y-Fe<sub>2</sub>O<sub>2</sub> were calculated using the Barrett-Joyner-Halenda (BJH) model and the Brunauer-Emmett-Teller (BET) equation, respectively (quantachrome V5.21 adsorption analyzer). The samples were degassed for 5.3 hours at 200 °C and weighed around 0.0442 mg. To get pore size distributions and BET surface area, BET and BJH interpretations were then applied using the N<sub>2</sub> adsorption-desorption isotherm at 77.35 K.

# 2.5. Chromium Adsorption study through batch system

Chromium (VI) reference solutions (100 mL) of varied quantities were imparted with a predetermined amount of adsorbent in Erlenmeyer flasks of 250 mL

for every single batch experiment. Adsorption activity was then assessed at a predetermined contact time at room temperature using an orbital agitator with a 140 rpm shaking speed. Once the solutions had reached equilibrium, they were all collected from the flask, traversed through a micron filter, and the quantity of remanent chromium content in the mixture was quantified using a spectrophotometer (Perkin Elmer, Lambda 850). In the experimental method, calibration standards were meticulously prepared by determining absorbance peak by scanning through wavelength range from 190 to 800 nm for known samples of chromium ranging from 0.2 to 8.0 mg L<sup>-1</sup>. DPC (Diphenyl Carbazide) method was used to determine the absorbance peak calorimetrically by the reaction of 1,5 Diphenyl Carbazide with dissolved hexavalent chromium solution to produce red-violet complex in an acidic medium (pH= $2.0 \pm 0.5$ ) [14]. The Maximum absorbances were measured at 543 nm against the reagent blank, Hence, this wavelength was selected to determine the absorption peak for equilibrium chromium content during all the experiments. The procedure was carried out in standard 10 mm quartz cuvettes in triplicate. The average absorbance was calculated, and a calibration curve was plotted. Validation of the analytical method was performed by finding the linearity range, detection limit (LOD), quantification limit (LOQ), relative standard

deviation (%RSD) and recovery percentage to verify the absence of interference as per recommendations of IUPAC [15].

Batch systems were set in a way to evaluate the impact of contact durations (5, 10, 20, 30, 60, 90, and 120 min), pH (ranging from 2-12), the effect of coexisting ions, ionic strength and reusability of the prepared material. Sorption results for hexavalent chromium onto CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> were expressed in terms of dismissal efficacy (%) and uptake capacity (mg g<sup>-1</sup>) using Equations (1 and 2).

Removal efficiency (%) = 
$$\frac{[c_i - c_t]}{c_t} \times 100$$
  
(Eq.1)

Adsorption capacity  $q_e (mg g^{-1}) = \frac{[C_i - C_t]}{m} \times V$ (Eq.2)

In the given formula, m represents adsorbent mass (gram),  $C_i$  and  $C_t$  signify the initial and residual chromium concentrations (mg L<sup>-1</sup>) observed at time t (min), and V stands for volume (L) in a batch study.

#### 2.6. Adsorption isotherm

Adsorption isotherms reflect the correlation between the concentration of a substance in either a gas or liquid phase and the quantity becomes adsorbed onto a solid surface when equilibrium is reached. The Langmuir and Freundlich isotherm models were utilized for calculating removal capacity. These models aid in comprehending the equilibrium connection between the quantity of chromium adsorbed onto the adsorbent at a certain temperature and the initial concentration of chromium in the solution. Isotherm equilibriums were studied for the varied concentrations of 100 mL chromium solution tested for 150 minutes stirring at ambient temperature with an optimum pH level of 4 by introducing 0.5 g adsorbent. The equilibrium concentration of chromium was determined spectrophotometrically. The Langmuir isotherm is often used to describe monolayer adsorption on a homogenous surface. It was adopted for adsorption to occur at specific sites on the adsorbent surface, and each site can adsorb only one sorbate ion [16]. The Freundlich isotherm is an empirical model focused on adsorption likely to occur on heterogeneous surfaces with varying adsorption energies [17,18].

Linear from for Langmuir and Freundlich isotherm equations were employed to describe the equilibrium adsorption parameters using Equations (3) and (4), respectively:

Langmuir Equation: 
$$\frac{1}{q_e} = \frac{1}{q_m} + \frac{1}{q_m K_L} \frac{1}{c_e}$$
  
(Eq. 3)

Freundlich Equation:  $logq_e = logK_f + \frac{1}{n}logC_e$ (Eq. 4)

Where  $q_e$  is the maximum adsorption capacity (mg g<sup>-1</sup>) of the prepared adsorbent,  $C_e$  is the equilibrium concentration of chromium (VI) (mg L<sup>-1</sup>). K<sub>L</sub> and K<sub>f</sub> are Langmuir and Freundlich constants, respectively.

# 2.7. Adsorption kinetic study

The field of kinetic investigation pertains to examining the temporal evolution of chemical reactions or processes. The study utilized various kinetic frameworks to analyze the mechanisms of mass transfer and chemical reactions associated with adsorption. Adsorption occurs when an adsorbate molecule interacts with an active site on the surface of an adsorbent molecule. The study employed to estimate pseudo-first and pseudo-second-order models. Through examining adsorption kinetics, valuable insights associated with the efficacy, ideal circumstances, and fundamental mechanisms of the aforementioned process were explored [19]. To comprehend the process of sorption on CH@PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>2</sub>, the experimental data was subjected to fitting using Lagergren's pseudo-first and pseudo-second order kinetics [20]. The Concentration of collected Chromium (VI) mixed with adsorbent material was determined spectrophotometrically at different time intervals of 2-120 min. The adsorption capacities provided at different times served as input for pseudofirst and pseudo-second order kinetics expressed as Equations (5) and (6), respectively:

$$Log(q_e - q_t) = Logq_e - \frac{K_1 t}{2.303}$$
(Eq. 5)
$$\frac{t}{q_t} = \frac{1}{K_2 q_e^2} + \frac{t}{q_e}$$
(Eq.6)

Where, and are adsorbed quantity (mg g<sup>-1</sup>) of chromium (VI) at equilibrium and at time t, respectively.  $K_1(\min^{-1})$  and  $K_2(g \text{ mg}^{-1}\min^{-1})$  are

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adsorption rate constants for pseudo-first and pseudosecond kinetics. Traditional linear plots will be obtained for both the mechanism and to predict the reliability of the study, A linear regression algorithm was applied for optimization analysis.

#### 2.8. Linear Regression analysis

The most frequent statistical technique for evaluating the model's validity is the linear regression coefficient. It statically measures the average functional relationship between variables. Also, it measures the degree of dependence of one variable on the other(s). It can be represented as blow Equations 7 and 8.

$$R^{2} = 1 - \frac{SS_{Resudal}}{SS_{Total}}$$
(Eq.7)

$$SS_{resudal} = \sum_{i=0}^{n} (q_{e,i} - q_{model,i})^{2}$$
(Fig. 8a)

$$SS_{total} = \sum_{i=0}^{n} (q_{e,i} - \overline{q_{e,i}})^{2}$$
(Fig. 8b)  

$$\overline{q_{e,i}} = \frac{1}{n} \sum_{i=0}^{n} q_{e,i}$$
(Fig. 8c)

Where,  $q_i$  and  $q_{model, i}$  have experimentally obtained data and isotherm model prediction, respectively. For ith data point where n in the number of experimental data point.

#### 2.9. Regeneration of adsorbent material

To evaluate the reusability of the synthesized adsorbent, six repeated cycles of sorption-desorption assays were tested. 100 mg of adsorbent was added to 100 ml Cr (VI) (100 mg L<sup>-1</sup>) for agitating at 140 RPM at 27°C for 12 hrs. After each agitation, adsorbent particles were separated from the suspension by a micron filter and washed thoroughly with desorption reagent (0.1 HNO<sub>3</sub>) followed by double deionized water and then dried at 80°C. The regenerated adsorbent was reused for the next cycle of the sorption process.

#### 2.10. Influence of co-existing ions

The adsorption effectiveness may be considerably impacted by the presence of other ions in the solution. Since the ultimate goal is the treatment of industrial effluents, which will most probably contain other ions (like Cl<sup>-</sup>, NO<sub>3</sub><sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, Na<sup>+</sup>, Mg<sup>2+</sup> etc.) along with heavy metal ions, an efficient adsorbent is expected to retain its high adsorption capacity even in the presence of other ions. For determining the effect of the presence of other ions, the adsorption efficiency with the prepared adsorbent was checked at two different concentrations (1 ppm and 10 ppm) of NaCl, and NaNO<sub>3</sub>. Na<sub>2</sub>SO<sub>4</sub>, Na<sub>2</sub>(PO<sub>4</sub>)<sub>3</sub> along with chromium ions.

# 3. Result and Discussion

#### 3.1. Characterization of Prepared Composite

X-ray diffraction (XRD) can provide а comprehensive understanding of the phase structural features of CH@PSB/y-Fe<sub>2</sub>O<sub>3</sub>. As illustrated in Figure 2 (a) the XRD patterns of pristine chitosan (CH) (85 % DDA) showed one strong and a weak reflection at  $2\Theta = 10.08^{\circ}$  and  $2\Theta = 20.22^{\circ}$  respectively which reflects about amorphous crystal structure. The XRD spectrum patterns of PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> exhibit several pairs of diffraction peaks in a range of 24<sup>0</sup> ~  $65^{\circ}$  as Figure 2(b). Based on the JCPDS Card No. 19-0629, the distinct peaks at certain angles located at 24.38°, 33.38°, 35.38°, 41.14°, 49.14°,  $54.26^{\circ}$  and  $62.60^{\circ}$  corresponding to the (012), (104), (110), (113), (024), (116), and (214) crystal planes respectively, are the characteristics for Fe<sub>2</sub>O<sub>2</sub> deposition on the PSB surface [21]. However, no apparent differences were observed for  $PSB/\gamma$ -Fe<sub>2</sub>O<sub>2</sub> and CH@PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> (Fig. 2c). The resulting XRD patterns were analyzed with peak shifting and lower peak intensity suggesting reduced crystallinity in  $CH@PSB/\gamma$ -Fe<sub>2</sub>O<sub>2</sub> due to interaction of chitosan polymer. The weaker diffraction signals compared to the well-ordered crystalline for  $PSB/\gamma$ -Fe<sub>2</sub>O<sub>2</sub> composite are due to the coating of chitosan on the Fe<sub>2</sub>O<sub>2</sub>-biochar surface. Chitosan typically contains amorphous regions, which might contribute to low intense diffraction peaks in the diffractograms. These amorphous regions could mask or dampen the intensity of the crystalline peaks from the magnetic biochar. Additionally, the peak shifts indicate alterations in the lattice parameters or crystallographic arrangement caused by the introduction of chitosan. These results suggest that chitosan was successfully immobilized with PSB/y-Fe<sub>2</sub>O<sub>2</sub> surface. FT-IR spectra for the pristine chitosan, PSB/y-Fe<sub>2</sub>O<sub>3</sub> and CH@PSB/ y-Fe<sub>2</sub>O<sub>3</sub> are shown in



Fig. 2. XRD patterns for CH (a);  $PSB/\gamma$ -Fe<sub>2</sub>O<sub>3</sub> (b); and CH@PSB/\gamma-Fe<sub>2</sub>O<sub>3</sub> (c)

Figure 3. The absorption band at 3200~3600 cm<sup>-1</sup> is attributed to the -OH and N-H groups stretching vibration present in chitosan (Fig. 3a) but the intensity of these bands reduced after coating due to interaction of these functional groups with biochar surface (Fig. 3c). The band at 2931 and 2850 cm<sup>-1</sup> (Fig. 3c) was assigned for starching vibration of aliphatic CH bond present in magnetic biochar [22]. The appearance of characteristic peaks around 1650-1550 cm<sup>-1</sup>, attributed to the amide I and II bands corresponding to N-H bending and C-N stretching vibrations and the presence of a band at around 1027 cm<sup>-1</sup> related to the C-O stretching vibration are strong evidence of successful chitosan coating on the magnetic biochar surface. These peaks are specific to chitosan's polysaccharide structure and are not typically observed in the FTIR spectra of magnetic biochar. The peak at 1365 cm<sup>-1</sup> is often attributed to the bending vibration of the methyl (CH<sub>2</sub>) groups. It indicates the presence of methyl groups in the composite, which could be associated

with either the chitosan component or the magnetic biochar. The peak at 1407 cm<sup>-1</sup> might be due to the presence of stretching vibration of the C-O bond in the -COOH group. In addition, a peak at 1653 cm<sup>-1</sup> is related to the amide I band, which arises from the C=O stretching vibration of the amide (CONH) bonds in chitosan. It confirms the presence of chitosan in the coated biochar sample. The peak at 1605cm<sup>-1</sup> in Fe<sub>2</sub>O<sub>2</sub>-biochar composite (Fig. 3b) was attributed to vibrations involving metal-oxygen bonds which disappeared after reacting with chitosan suggesting a significant change in the composition or structure of the material due to the chemical interaction with composite and surface coverage. The detection of the aforementioned peaks confirms the successful incorporation of chitosan onto the biochar surface, validating the formation of the chitosan-magnetic biochar composite. Additionally, the disappearance of peaks at 1064 and 1150 cm<sup>-1</sup> in chitosan spectra (Fig. 3a) reveals that coating onto the magnetic biochar surface suggests significant



Fig. 3. FTIR spectra for Chitosan (a);  $PSB/\gamma$ -Fe<sub>2</sub>O<sub>3</sub> (b); and CH@PSB/\gamma-Fe<sub>2</sub>O<sub>3</sub>(c)

changes in the chitosan's chemical environment which involves chemical interactions, cross-linking, hydrogen bonding, conformational changes in the chitosan molecules. Furthermore, compared with chitosan polymer, in the spectrum of CH@PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> a new strong peak located at 527 cm<sup>-1</sup> was attributed to the Fe-O lattice vibrations of Fe<sub>2</sub>O<sub>4</sub>. This indicates the presence of Fe<sub>3</sub>O<sub>4</sub> particles in the prepared sample [23]. Notably, the functional groups of CH@PSB/ y-Fe<sub>2</sub>O<sub>2</sub> were different from those of pristine chitosan and magnetic biochar, which impacted its subsequent adsorption ability. Magnetic biochar made from ferric chloride has distinctive characteristics in its surface morphology. As shown in Figure 4 (b), it generally has a porous structure with a variety of pore sizes and internal mesopore distributions, which increases its surface area and adsorption capacity. The breakdown of lignocellulosic material at high temperatures was linked to the production of a sequence of cracks on the surface of biochar, which caused volatile chemicals to evaporate from the newly created pores. Some shimmering developed on the surface of PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>, indicating the existence of thick surface layering particles, which are composed of magnetic nanoparticles inserted during manufacturing. These magnificent particles might be incorporated into the porous surface of the biochar or scattered throughout it. Due to the carbonization process and the development of cracks and fissures, the surface seems uneven and rough. Figure 4 (d, e, and f) shows that chitosan coating on magnetic biochar may result in a reduction in pore volume or quantity when compared to uncoated magnetic biochar. This is mainly because the magnetic biochar's surface develops a layer of chitosan coating, which partly fills in the pores already present and decreases the volume of those pores. However, CH@PSB/y-Fe<sub>2</sub>O<sub>2</sub> had a higher sorption capacity than PSB/γ- $Fe_2O_3$ , indicating that the adsorption process was predominantly dependent on the functional groups rather than the surface pore structure.



Fig. 4. SEM images of Pristine chitosan (a);  $PSB/\gamma$ -Fe<sub>2</sub>O<sub>3</sub>(b, c); and CH@PSB/\gamma-Fe<sub>2</sub>O<sub>3</sub>(d, e, f)

EDS results for Chitosan (CH), magnetic Biochar (PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>2</sub>) and chitosan-coated magnetic biochar (CH@) PSB/  $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>) offer a glimpse into their composition and potential structural modifications. In this study, we delve into the variations in C, N, O and Fe contents across Fe<sub>2</sub>O<sub>2</sub>/biochar, chitosan-coated Fe<sub>2</sub>O<sub>2</sub> biochar, and pure chitosan. From Figure 5 (a, b, and c) EDS of the prepared material showed remarkable transformation after coating of chitosan onto magnetic biochar. The CH@PSB/y-Fe<sub>2</sub>O<sub>3</sub> displays a significant increase in carbon content, reaching 57.7% as compared to  $PSB/\gamma$ - Fe<sub>2</sub>O<sub>2</sub> (25.8%) This enhancement is indicative of the successful incorporation of chitosan onto the biochar surface. Observed carbon content in pure chitosan of 42.3%. The distinctive increase in carbon content observed in the CH@PSB/y- Fe2O3 sample corroborates the successful interaction between chitosan and the biochar matrix. The decline in nitrogen content in the CH@PSB/y-Fe<sub>2</sub>O<sub>3</sub> sample (0.9 %) compared with PSB/ $\gamma$ - Fe<sub>2</sub>O<sub>2</sub> (3.3 %) suggests that the chitosan coating process has led to modifications in the nitrogen composition of the composite material. The results for BET surface analysis are shown in Table 1 the results were compared with BET analysis of pristine biochar made from peanut shells with  $Fe_2O_3$  and crosslinked chitosan-loaded samples showed a significant reduction in surface area as well as pore volume and pore size. The reduction is due to the aggregation of  $Fe_2O_3$  nanoparticles and chitosan on to surface. While the reduction in surface area, pore size, and volume might seem unfavourable, the addition of chitosan onto the magnetic biochar surface might enhance specific functionalities of the material's behavior for targeted adsorption.

With the intent of deeper insight into the surface chemical and elemental composition of the prepared adsorbent, XPS survey spectra were recorded and analyzed according to Figure 6 (a to d). As illustrated in Figure 6a, XPS survey spectra of CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> find four significant peaks which revealed the presence of C 1s (283.5 eV), N 1s (396.5 eV), O 1s (531 eV) and Fe 2p (711.5 eV). It was further analyzed that the elemental composition of CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> contains C (61.89 %), O (21.78 %), N (2.83 %) and Fe (8.08 %). The high-resolution spectrum for C 1s is deconvoluted in Figure 6b, revealing the existence



Fig. 5. EDS analysis of Chitosan (a); PSB/γ-Fe<sub>2</sub>O<sub>3</sub> (b); and CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub> (c)

Sample	$S_{BET} (m^2 g^{-1})$	V <sub>micro</sub> (cm <sup>3</sup> g <sup>-1</sup> )	Pore diameter (nm)
PS Biochar [13]	479.569	0.124	1.426
$PSB/\gamma$ - $Fe_2O_3$	340.252	0.958	1.053
CH@PSB/γ-Fe <sub>2</sub> O <sub>3</sub>	98.788	0.875	0.756

Table 1. BET and BJH analysis results for prepared samples

PSB: Peanut shell biochar

of carbon in three different functional groups: C-N (283.84 eV), C-C (284.64 eV), C-O (286.72 eV). The high-resolution spectrum of O 1s [Figure 6c] finds two deconvoluted peaks with binding energies at 531.59 (eV) and 529.24 (eV) corresponding to C=O and C-O functional groups, respectively. The high-resolution XPS spectra of Fe 2p of CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> exhibited four peaks: 723.77 eV could be assigned for Fe 2P<sub>1/2</sub>; 709.71 for Fe 2P<sub>3/2</sub>; and peaks corresponding to binding energies at 718.02 and 713.71 may be associated with

shake-up or satellite peaks which are characteristics of Fe in Fe<sub>2</sub>O<sub>3</sub>. The XPS results of chitosan-coated magnetic biochar indicated the potential appearance of the iron oxides, reduced iron species, interacting with chitosan polymer. Figure 7a illustrates the zero point charge of CH@PSB/ $\gamma$ - Fe<sub>2</sub>O<sub>3</sub> is located at 4.5 pH. As a result of the protonation of the amine group in chitosan, the adsorbent's surface acquires a positive charge below this pH, which is favorable for the adsorption of negatively charged entities.



Fig. 6. XPS Survey Spectra of CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> (**a**); High resolution XPS spectra of C 1s (**b**); O 1s (**c**); Fe 2p (**d**)

#### 3.2. Adsorption studies for chromium

#### 3.2.1. Consequences of changing solution pH

The adsorption of chromium by CH@PSB/y-Fe<sub>2</sub>O<sub>2</sub> can be significantly influenced by the pH of the solution. When pH is less than 6.8, HCrO<sub>4</sub>- becomes major speciation but at pH more than 7.2, Cr (VI) ions can form negatively charged oxyanions such as chromate  $(CrO_4^{2-})$  and dichromate  $(Cr_2O_7^{2-})$ . The point of zero charge for CH@PSB/y-Fe<sub>2</sub>O<sub>2</sub> was located at 4.5 pH (Fig. 7a). Adsorption studies are typically conducted at varied ranges of pH to determine the optimal conditions for efficient chromium removal. Sorption capacity for Cr (VI) notably declined from 85.2 mg g<sup>-1</sup> to 24.6 mg g<sup>-1</sup> with the change in pH value from 2 to 8 as depicted in Figure 7b. Sorption performance for the produced composites was found to be boosted at lower pH value, mainly due to enhanced electrostatic attraction created by protonated amino functional groups on chitosan. At higher pH, the negatively charged surface of adsorbent material impeded chromium sorption due to electrostatic repulsion among (CrO<sub>4</sub><sup>2-</sup>) and OH-. Figure 8 illustrates the process of chromium adsorption by sorbent material.

#### 3.2.2. Influence of Ionic Strength on Adsorption

The sorption behaviour of chromium by CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> was investigated in the presence of a varied concentration of NaNO<sub>3</sub> and results were exhibited as decreasing adsorption capacity from 83.6 to 30.1 mg L<sup>-1</sup> (Fig. 9). With increasing NaNO<sub>3</sub> strength from 0.05 M to 2M maintaining pH at 4.0, suggesting supersession in Cr (VI) sorption at elevated ionic strength. This was primarily due to the competitive sorption of nitrate ions onto CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub>.

#### 3.3. Adsorption Equilibrium Analysis

The adsorption capacity for the prepared material was evaluated using the two most universally adopted Langmuir and Freundlich isotherm models. The pertinent reports are provided in Table 2 and Figure 10 (a and b) are illustrated for validity of the models. The linear fitting of the observed data based on origin pro 8.5 software revealed that the performance of CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> was better fitted by the Langmuir equation with a correlation factor (R<sup>2</sup>) of 0.9998. These findings demonstrated the monolayer adsorption process and Equation 3 was used to determine the maximum adsorption capacity (Q<sub>max</sub>) as 14.285 mg g<sup>-1</sup>.


Fig. 7. Zero-point charge (a); and effect oZero-point-sorption of chromium onto CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub>(b)



Fig. 8. Strategic Representation of Chromium Adsorption onto CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub>



Fig. 9. Effect of Ionic Strength



Fig. 10. Isothermal equilibrium study for chromium adsorption onto  $CH@PSB/\gamma$ -Fe<sub>2</sub>O<sub>3</sub> Langmuir (a); and Freundlich model (b)

#### 3.4. Adsorption kinetics

Sorption rate is another consideration for studying adsorption capacity associated with contact duration. The sorption kinetic was investigated for 0 to 90 min of contact time. As shown in Figure 11 sorption occurred rapidly for an initial 20 min of contact and then this trend slowly grew until it reached a plateau. Sorption equilibrium attained in just 40 min. The elevated abundance of adsorption surfaces may be assigned for the speedy progress in the initial adsorption rate of Cr (VI) [24]. Adsorption rates accordingly decreased as the number of accessible adsorption sites decreased over time. As a result, 40 minutes was shown to be an ideal contact time for the adsorption process. The sorption kinetics for pseudo-first and pseudo-second-order models were applied for chromium adsorption onto CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> and corresponding results were evaluated in Figure 12 (a and b) and Table 2. The experimental findings of calculated parameters revealed that the data were best signified by the pseudo-second-order model (Table 2), with a correlation coefficient (R<sup>2</sup>) of 0.9994 with an equilibrium sorption capacity of 83.33 mg g<sup>-1</sup>. Kinetics consideration is suggested for surface adsorption and chemisorption mechanism in the ratecontrolling step.



**Fig. 11.** Effect of time on adsorption of chromium onto CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub>



Fig. 12.Adsorption Kinetics for Chromium sorption onto CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub>: Pseudo-first (a); and Pseudo-second-order model (b) and Pseudo-second-order model (b)

Langmuir constants				Freundlich	n constants	
KL	Q <sub>max</sub> (mg g <sup>-1</sup> )	R <sup>2</sup>	K <sub>F</sub>	n		R <sup>2</sup>
0.1170	14.285	0.9998	0.616	2.56	(	0.9871
Experimental value	Pseudo first order			Pseudo second order		
Qe,exp.	$K_1$	Qe, cal	$\mathbb{R}^2$	K <sub>2</sub>	Qe, cal	R <sup>2</sup>
(mg g <sup>-1</sup> )	(min <sup>-1</sup> )	(mg g <sup>-1</sup> )		(g mg <sup>-1</sup> min <sup>-1</sup> )	(mg g <sup>-1</sup> )	
0.168	0.0161	14.79	0.744	0.089	83.33	0.9994

Table 2. Adsorption isotherm and kinetic parameters for chromium adsorption onto CH@PSB/y-Fe2O3

Initial Chromium content: 2 mg L-1; pH:4.0; Temperature:28 0C; Time:0-90 min@140RPM; Adsorbent dose: 0.5g; Solution volume: 100 mL

# 3.5. Reutilization of the adsorbent

Six consecutive iterations of sorption-desorption were tested for synthesized adsorbent to execute reusability. As demonstrated in Figure 13, removal efficiency gradually declined with every next cycle, and it dropped from 85 % to 26 % in the sixth round. The material has excellent reusability for up to four cycles. Overall, the results of these recycled trials provide compelling evidence that the prepared CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> demonstrated excellent reusability and holds significant promise for practical applications.



**Fig. 13**. Six sorption-desorption cycles for Cr (VI) adsorption onto CH@PSB/γ-Fe<sub>2</sub>O<sub>3</sub>

# 3.6. Influence of coexisting ions on removal efficiency for real water samples

Drinking water can contain multiple ions as water sources often come into contact with geological formations, industrial activities, or other environmental factors. The synchronous presence of other ions can significantly influence the removal of chromium during water treatment. The interactions between these coexisting ions and chromium can affect the adsorption capacity, speciation, and overall efficiency of the removal process. Experiments were therefore carried out to learn how coexisting ions affect chromium removal in the presence of four ions such as Cl<sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, NO<sub>3</sub><sup>-</sup>, and PO<sub>4</sub><sup>2-</sup> (for 1 ppm or 1 mgL<sup>-1</sup> and 10 ppm concentrations). The experimental observations (Fig.14) suggesting for consistent efficiency at approximately 80 % chromium removal by CH@PSB/ $\gamma$ -Fe<sub>2</sub>O<sub>3</sub> at 1 ppm solution. Except for PO<sub>4</sub><sup>2-</sup> removal efficiency by coexisting ions was only minimally hampered. It is because of competitive interactions of anionic species with functional groups on modified chitosan. However, there is a weaker interaction for Cl<sup>-</sup>, SO<sub>4</sub><sup>2-</sup>, NO<sub>3</sub><sup>-</sup> but due to the higher affinity of PO<sub>4</sub><sup>2-</sup> ion with iron oxide its effect is high. In addition, as the concentration of coexisting ions increased to 10 ppm, this influence was higher.



#### 3.7. Validation of analytical method

The analytical data acquired for the spectrophotometric validity of the method are presented in Table 3. Using 100 mL solutions with increasing analyte concentrations  $(0.2 - 8 \ \mu g \ L^{-1})$ , the absorbance peaks and calibration curves were produced, as shown in Figures 15 (a) and 15 (b), respectively. All points of the calibration curve were subjected to the preconcentration procedure for the determination of Cr (VI) via the carbazide method as mentioned in the experimental part of this work. The calibration curve exhibits linearity in the range from 0.5 to  $6 \,\mu g \, L^{-1}$  with a regression coefficient ( $R^2$ ) equal to 0.9976. Using seven consecutive measurements, the analytical approach presented in this research demonstrated high accuracy with a relative standard deviation (RDS) of 7.62%. The limit of detection (LOD), defined as the smallest amount of analyte that a method can detect, was

% Relative Standard Deviation (RSD)

% Recovery Percentage (R)

calculated to be 3 times the standard deviation obtained by 7 readings divided by the slope of the calibration curve. The quantification limit (LOQ), which expresses the actual amount of analyte in a sample with considerable precision and accuracy, was determined to be 10 times the standard deviation obtained by 7 readings divided by the slope of the calibration curve. The LOD was 0.65  $\mu$ g L<sup>-1</sup> and the LOQ was 2.16  $\mu$ g L<sup>-1</sup>. The accuracy of the method was tested from three known concentrations of standard solution, and the results revealed good percent recovery (% R) of 92.8  $\pm 0.5\%$  to  $103.09 \pm 0.5$ %. A comparison of proposed methods and some of the published microextraction methods for extraction and determination of Cr ions are summarized in Table 4. The accuracy and LOD values of the present method, for the determination of chromium ions, are comparable to those reported in the literature [25-32].

7.62

to  $103.1 \pm 0.5 \ 0.5 \pm 92.8$ 



Fig. 15. UV Spectra for calibration standards (a); and calibration curve (b)

by proposed 6 v-vio spectrophotometer				
Characteristics	Validation Parameters			
Calibration Curve	Values			
Concentration Range (µg L <sup>-1</sup> )	0.2-8.0			
Absorbance Range	0.09-4.75			
Wavelength Range (nm)	190-800			
Slope	0.6583			
Intercept	0.172			
Regression Coefficient	0.9976			
Regression Equation	Y= 0.6583X + 0.172			
Validation Appearances	Values			
Measurement wavelength (nm)	543			
Linear Range (µg L <sup>-1</sup> )	6.0 - 0.5			
Limit of Detection (LOD) µg L <sup>-1</sup>	0.65			
Limit of Quantification (LOQ) µg L <sup>-1</sup>	2.16			

 Table 3. Analytical validation parameters for Cr (VI) determination

 by proposed UV-VIS spectrophotometer

Detection method	LOD	Separation	RSD	Linear range	Recovery	Rof
Detection method	(µg L-1)	Technique	(%)	(µg L <sup>-1</sup> )	(%)	
SPE-FAAS		SPE	5 >	0.4-15	97 <	[25]
ETAAS	0.005	TSIL-DLLME	3.8	0.32 -0.02	105 -95	[26]
ETAAS	0.005	IICDET/LLME	3.8	0.02-1.75	98	[27]
ETAAS*	8.0	IL-DLLBME	4.3	0.03-4.4	104 -97	[28]
FAAS	7.1	SPE	2.7	20-700	88-110	[31]
Spectrophotometric	0.023			0.03-6.0		[32]
UV-Vis	0.65	DPC	7.62	6-0.5	92-103	This work

**Table 4.** Comparison of UV-Vis spectrophotometry using CH@PSB-γ-Fe<sub>2</sub>O<sub>3</sub> to determine Cr (VI) in water with other studies of the literature

FAAS: Flame Atomic Absorption spectrometry

SPE: Solid Phase Extraction

ET-AAS: Electro-Thermal Atomic Absorption Spectrometer

**UV-Vis Spectrometry** 

\*LOD=0.8 ng L<sup>-1</sup>

# 4. Conclusion

In this study, a new adsorbent material was fabricated by immobilizing chitosan-coated iron oxide biochar for the effective desalinization of chromium ions from aqueous solution. Various spectroscopic studies confirmed the synthesis of a composite material comprising Fe<sub>2</sub>O<sub>2</sub>-biochar coated further with crosslinked chitosan. This hybrid material combines the magnetic properties of Fe<sub>2</sub>O<sub>3</sub> with the porous structure of biochar, enhanced by the presence of chitosan. The successive coating of chitosan onto the Fe<sub>2</sub>O<sub>2</sub>-biochar surface adds a layer of complexity the material's composition and properties, to potentially impacting its surface area, porosity, and adsorption capabilities. This synthesized material holds promise for applications in the adsorption of chromium ions and the spectroscopic method used in the determination of chromium showed a LOD of 0.65 µg L<sup>-1</sup> with a recovery percentage of more than 92 %. The optimized pH for the highest removal capacity  $(0.4 \pm 84.9 \text{ mg g}^{-1})$  is in the range of 3-5 pH. Additionally, there is a slight decrease in capacity with an increase in ionic strength. Langmuir isotherm equation fits well with adsorption data and the obtained removal capacity for CH@PSB/y-Fe<sub>2</sub>O<sub>2</sub> exhibits 14.285 mg g<sup>-1</sup> in batch system. The kinetic study suggests that pseudo-second-order is the best description of chromium adsorption onto CH@ PSB/y-Fe<sub>2</sub>O<sub>3</sub> suggesting a chemisorption pathway. The a slight decrease in removal efficiency in the presence of some other anions except for PO<sub>4</sub><sup>2-</sup> ion. A reusability assay indicates for excellent utility of the prepared material for toxicant removal and more

than 50 % sorption capacity is retained for up to four cycles. In conclusion, the synthesized biosorbent is a novel green material for effective holds significant potential for wide-ranging applications in the field of Cr (VI) removal.

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#### 6. Reference

- L. Liu, W. Li, W. Song, M. Guo, Remediation techniques for heavy metal-contaminated soils: Principles and applicability, Sci. Total Environ., 633 (2018) 206-219. https://doi.org/10.1016/j. scitotenv.2018.03.161
- [2] X. Zhou, T. Korenaga, T. Takahashi, T. Moriwake, S. Shinoda, A Process monitoring controlling system for the treatment of wastewater containing chromium (VI), Water Res., 27 (1993) 1049-1054. https://doi.org/10.1016/0043-1354(93)90069-T
- [3] Y. Pang, Zeng, G.M. Zeng, L. Tang, Y. Zhang, Y. Y. Liu, X.X. Lei, M.S. Wu, Z. Li, C. Liu, Cr(VI) reduction by Pseudomonas aeruginosa immobilized in a polyvinyl alcohol/sodium alginate matrix containing multi-walled carbon nanotubes, Bioresour. Technol., 102

(2011) 10733-10736. https://doi.org/10.1016/j. biortech.2011.08.078

- [4] D. Mohan, C.U. Pittman, Activated carbons and low cost adsorbents for remediation of tri- and hexavalent chromium from water, J. Hazard. Mater., 137 (2006) 762-811. https:// doi.org/10.1016/j.jhazmat.2006.06.060
- [5] X. Guo, X. Lü, The need for biofuels in the context of energy consumption, Advances in 2<sup>nd</sup> generation of bioethanol production, Woodhead Publishing Series in Energy, CRC press, New York, pp 9-30, 2021. https://doi.org/10.1016/ B978-0-12-818862-0.00004-2
- [6] W. Zheng, K. Phoungthong, F. Lü, L.M. Shao, P.J. He, Evaluation of a classification method for biodegradable solid wastes using anaerobic degradation parameters, Waste Manage., 33 (2013) 2632–2640. https://doi.org/10.1016/j. wasman.2013.08.015
- [7] I.G. Coronilla, L. M. Barrera, E. C. Urbina, Kinetic, isotherm and thermodynamic studies of amaranth dye biosorption from aqueous solution onto water hyacinth leaves, J. Environ. Manage., 152 (2015) 99-108. https://doi. org/10.1016/j.jenvman.2015.01.026
- [8] A. Zhang, X. Li, J. Xing, G. Xu, Adsorption of potentially toxic elements in water by modified biochar: A review, J. Environ. Chem. Eng., 8 (2020) 104196. https://doi.org/10.1016/j. jece.2020.104196
- [9] U. Upadhyay, I. Sreedhar, S.A. Singh, C.M. Patel, K. L. Anitha, Recent advances in heavy metal removal by chitosan based adsorbents, Carbohydr. Polym., 251(2021) 117000. https://doi.org/10.1016/j.carbpol.2020.117000
- [10] E. Guibal, Interactions of metal ions with chitosan-based sorbents: A review, Sep. Purif. Technol., 38 (2004) 43-74. https://doi. org/10.1016/j.seppur.2003.10.004
- [11] W.C. Chong, J.F. Chin, Z.W. Heng, H.C. Teoh, Y.L. Pang, Recent development of magnetic biochar crosslinked chitosan on heavy metal removal from wastewater - Modification, application and mechanism, Chemosphere, 291 (2022)133035. https://doi.org/10.1016/j. chemosphere.2021.133035
- [12] J. Cheng, F. Xiao, W. Cao, C. Yang, J. Chen, Z. Luo, Removal of heavy metals from aqueous solution using chitosan-combined magnetic biochars, J. Colloid Interface Sci., 540 (2019) 579-584. https://doi.org/10.1016/j. jcis.2019.01.068
- [13] A. Choudhary, A. Kadawasara, S.S Poonia,

P. kumar, V.K. Janu, Pyrolytic preparation of active carbons from peanut shell biomass for adsorptive elimination of fluoride from groundwater of Shekhawati region, Orient. J. Chem., 38 (2022) 1338-1350. http://dx.doi. org/10.13005/ojc/380602

- [14] APHA, Standard methods for the examination of water and wastewater. 21st edition, american public health association/American water works wssociation/water environment federation, Washington DC, 1288 pages, 2005. www.apha.org
- [15] M. Thompson, SLR. Ellison, R. Wood, Harmonized guidelines for single-laboratory validation of methods of analysis (IUPAC Technical Report). Pure Appl. Chem., 74 (2002) 835–855. https://doi.org/10.1351/ pac200274050835 35.
- [16] I. Langmuir, The constitution and fundamental properties of solids and liquids, J. Am. Chem. Soc., 183 (1917) 102–105. https://doi. org/10.1021/ja02268a002
- H. Freundlich, Uber die adsorption in lösungen, Zeitschrift für Physikalische Chem., 57 A (1907) 385–470. https://doi.org/10.1515/zpch-1907-5723
- S. Ahmed and S. Ikram, Chitosan and its derivatives: A Review in recent innovations, Int. J. Pharm. Res., 6 (2015) 14-30. http://dx.doi. org/10.13040/IJPSR.0975-8232.6(1).14-30
- [19] Y. S. Ho, G. McKay, The kinetics of sorption of divalent metal ions onto sphagnum moss peat, Water Res., 34 (2000) 735–742. https://doi. org/10.1016/S0043-1354(99)00232-8
- [20] S. Lagergren, On the theory of the so-called adsorption of dissolved substances, solubility (Theoretical chemistry), Sven. Vetenskapsakad. Handingarl, 24 (1898) 1–39. https://cir.nii. ac.jp/crid/1570009750361875328
- [21] X. Duan, D. Wang, G. Qian, J.C. Walmsley, A. Holmen, D. Chen, X. Zhou, Fabrication of K-promoted iron/carbon nanotubes composite catalysts for the Fischer–Tropsch synthesis of lower olefins, J. Energy Chem., 22 (2016) 13-20. https://doi.org/10.1016/j.jechem.2016.01.003
- [22] J. Deng, Y. Liu, S. Liu, G. Zeng, X. Tan, B. Huang, X. Tang, S. Wang, Q. Hua, Z. Yan, Competitive adsorption of Pb(II), Cd(II) and Cu(II) onto chitosan-pyromellitic dianhydride modified biochar, J. Colloid. Interface. Sci., 506 (2017) 355-364. https://doi.org/10.1016/j. jcis.2017.07.069
- [23] R. Li, W. Liang, H. Huang, S. Jiang, D. Guo, M. Li, Z. Zhang, A. Ali, J.J. Wang, Removal of

cadmium (II) cations from an aqueous solution with aminothiourea chitosan strengthened magnetic biochar, J. Appl. Polym. Sci., 135 (2018) 46239. https://doi.org/10.1002/ app.46239

- [24] W. Zhang, S. Mao, H. Chen, L. Huang, R. Qiu, Pb(II) and Cr(VI) sorption by biochars pyrolyzed from the municipal wastewater sludge under different heating conditions, Bioresour. Technol., 147 (2013) 545–552. https://doi.org/10.1016/j.biortech.2013.08.082
- [25] F. Golbabaei, Z. Sadeghi, A. Vahid, A. Rashidi, On-line micro column preconcentration system based on amino bimodal mesoporous silica nanoparticles as a novel adsorbent for removal and speciation of chromium (III, VI), J. Environ. Health Sc. Eng., 13 (2015) 1-12. https://doi.org/10.1186/s40201-015-0205-z
- [26] M.M. Eskandari, B. Kalantari, Dispersive liquid-liquid microextraction based on taskspecific ionic liquids for determination and speciation of chromium in human blood, J. Anal. Chem., 70 (2015) 1448-1455. https://doi. org/10.1134/S1061934815120072
- [27] H. Shirkhanloo, M. Ghazaghi, M.M. Eskandari, Cloud point assisted dispersive ionic liquidliquid microextraction for chromium speciation in human blood samples based on isopropyl 2-[(isopropoxycarbothiolyl) disulfanyl, Anal. Chem. Res., 10 (2016)18-27. https://doi. org/10.1016/j.ancr.2016.10.002
- [28] H. Shirkhanloo, M. Ghazaghi, H.Z. Mousavi ,Chromium speciation in human blood samples based on acetyl cysteine by dispersive liquid–liquid biomicroextraction and in-vitro evaluation of acetyl cysteine/cysteine, J. Pharm. Biomed. Anal., 118 (2016) 1-8. http://dx.doi.org/10.1016/j.jpba.2015.10.018
- [29] S. A. H. Mirzahosseini, the evaluation and determination of heavy metals pollution in edible vegetables, water and soil in the south of Tehran province by GIS, Arch. Environ. Prot., 41 (2015) 63-72. https://doi.org/10.1515/aep-2015-0020
- [30] M Arjomandi, A review: analytical methods for heavy metals determination in environment and human samples, Anal. Methods Environ. Chem. J., 2 (2019) 97-126. https://doi.org/10.24200/ amecj.v2.i03.73
- [31] V. Leitea, B. Jesisa, V. Duartea, V. Constantinob, C. Izumic, J. Trontoa, F. Pintoa, Determination of chromium (VI) by dispersive solid-phase extraction using dissolvable Zn-Al layered double hydroxide intercalated with L-Alanine as

adsorbent, Microchem. J., 146 (2019) 650–657. https://doi.org/10.1016/j.microc.2019.01.063

[32] A. Lace, D. Rayan, M. Bowkett, J. Cleary, Chromium Monitoring in water by colorimetry using optimised 1,5-diphenylcarbazide method, Int. J. Environ. Res. Public Health, 16 (2019) 1803; https://doi.org/10.3390/ijerph16101803



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Solid phase-fabrication of multi-walled carbon nanotubes and their derivatives for efficient extraction and analysis of Bismarck Brown-Y Dye from aqueous solution

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# ABSTRACT

This investigation used efficient multi-walled carbon nanotubes (MWCNTs) and their derivatives; MWCNT-Tris, MWCNT-H, MWCNT-Tetra and MWCNT-G, for extraction and removing the Bismarck Brown-Y (BB-Y) by solid phase extraction (SPE). The concentration of BB-Y was measured by UV-Vis spectrophotometer after the SPE technique. The solid phases were analyzed and characterized by utilizing several techniques, including Fourier Transform Infrared Spectroscopy (FTIR), Field Emission Scanning Electron Microscopy (FESE), zeta potential measurement, and X-ray Diffraction (XRD). At optimization conditions, the optimum concentration of the BB-y was obtained at 200 mgL<sup>-1</sup> and 300 mg L<sup>-1</sup> for MWCNT and MWCNT-Tris, whereas 400 mg L<sup>-1</sup> for MWCNT-H, MWCNT-Tetra, and MWCNT-G. Additionally, the optimal pH value was 6.0 for MWCNT-Tris, and it was 10 for MWCNT, MWCNT-H, MWCNT-Tetra, and MWCNT-G. However, the volume of samples was achieved at 25 mL. Furthermore, it was found that the most effective flow rate for the eluting solvent was 0.5 ml min<sup>-1</sup>. Besides the type and volume of eluents were examined and evaluated. Finally, the present work involved the determination of adsorption capacity using Langmuir and Freundlich isotherm models under ideal conditions. The Langmuir model revealed that the  $q_{max}$  for the MWCNT, MWCNTtris, MWCNT-H, MWCNT-Tetra, and MWCNT-G was obtained 862.07, 1075.27, 1282.05, 1298.70, and 1333.33 mg g<sup>-1</sup>, respectively.

# 1. Introduction

A living ecosystem requires water as a fundamental element. Although the water covers 1.386 billion square kilometers of the world, approximately 97 percent is made up of salt-water-containing seas and oceans, and over two percent is in the form of ice caps and glaciers. Still, approximately one percent is distributed as rivers, lakes, underground water, and water vapor can be drinking water [1]. Nowadays, one of the most critical problems in the world today is water contamination by numerous hazardous substances. Multiple harmful pollutants are released into the environment due to the rapid rise of human industrialization, unplanned activities such as urbanization, and careless use of water resources from nature, as well as enormous population growth [2]. Polluted waters include a variety of contaminants, including both inorganic and organic, which include dyes, pesticides, pharmaceuticals, degraded organic waste, and organic pollution. Besides, harmful heavy elements represent inorganic pollutants [3]. The dyes are colored organic materials used in various industries including clothing, textiles, leather, plastic, paper, medicine, beauty products, and food. They also color hair, fur, petroleum products, and lubricants.

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Moreover, dyes represent one of the pollutants with the most hazardous chemicals of all those productions. According to the report, around 200,000 tons of dyes are utilized in industries annually, and their chromophorecontaining chemical effluent is improperly treated before being drained into water [4] The effect of dyes and other pollutants starts when discharged into the environment, particularly those that are harmful to living creatures [5]. Typically, dyestuffs offer outstanding coverage when used at relatively low concentrations as a result of their high intensity of color which lead to unwilling color changes in the waste brought on by the dyes and can prevent sunlight from penetrating due to the effect on consumption of dissolved oxygen, hence inhibiting photosynthesis process. Besides, the colored textile effluent also makes the water more mutagenic, carcinogenic, and gene-toxic, all of which pose serious risks to the environment as well as flora and fauna [6-8]. In 1856, Perkin produced the first manufactured dye, mauveine, an aniline dye [9] Natural and artificial dyes are the two main groups that are based on the raw ingredients used. Natural dyes are produced by using trees, plants, and other elements of nature. However, synthetic dyes, are produced by using chemicals and oil [10]. There is another classification of dyes depending on either their structure or their application, based on their structural characteristics, the following dyes are categorized: azo, nitro, phthalein, triphenyl methane, indigo, and anthraquinone while, the classification of dyes based on their application, including acid dye, basic dye, direct dye, ingrain dye, dispersed dye, mild dye, vat dye, and reactive dyes [11]. Nevertheless, due to their intricate chemical compositions, dyes are very impervious to decay. Consequently, they often remain stable in a variety of circumstances, including aerobic digestion, heat, light, and oxidizing factors [12]. Therefore, different ways to remove dyestuffs from wastewater before discharging into the environment are membrane filtration, coagulation, flocculation, adsorption, modified oxidation processes, photocatalytic degradation, and biological therapy which beyond one of three basic techniques which are physical, biological and chemical [13,14]. Traditional physical techniques, such as adsorption and separating membranes. The adsorption method is attractive for the removal of colors even though dyes are resistant to oxidation, and biodegradation, as well as being stable to light and heat. Furthermore, the adsorption method is effective and doesn't need any further preprocessing before being used. Besides, it is regarded cheap and optimal method [15,16]. Solid phase extraction (SPE) is a conventional approach to acquire adequate samples for analysis. The partition coefficient between the mobile/aqueous

phase and the adsorbent is what determines how the analyte is separated. There have been reports of various modified SPEs. The most popular types of SPE include column SPE, pipette tip SPE (PT-SPE), magnetic solid phase extraction (MSPE), and dispersive solid phase extraction (DSPE) [17]. SPE for preconditioning samples has several benefits, including quick separation, low prices, little solvent use, high enrichment efficiency and recovery rate, rapid processing times, no emulsion creation, and the flexibility to be used with various analytical detection techniques [18]. As adsorbent materials carbon nanoparticles (CNs) represented by graphene, graphene oxide, carbon nano-disks, carbon nanotube rings, either single or multiwall, and carbon nanocones, are commonly employed in SPE [19]. As opposed to typical adsorbent materials, CNTs offer great chemical stability, higher surface area, smaller pore size, hollow structure, and ease of modifying; these benefits make CNTs a more favorable adsorbent than traditional adsorbent for preparing samples techniques in addition to their purity, surface area, functional groups on the surface, adsorption sites, and experimental settings, CNTs' adsorption effectiveness also depends on these factors [20]. Also, dves and other organic compounds (BTEX) can be extracted by nanostructures such as CNTs, graphene (G), graphene oxide(GO), silica (SiO<sub>2</sub>), and Metal-organic frameworks (MOFs) by SPE methods before being determined by the liquid or gas chromatography (GC-FID/ GC-MS/ HPLC) in liquid phases [21-27]. This paper aims to examine the feasibility of utilizing a multi-walled carbon nanotube and its derivatives as a solid phase in solid-phase extraction (SPE) for the purification of water contaminated with organic hazardous dye Bismarck Brown-y, 1,3-Benzenediamine, -[1,3-phenylenebis (2,1-diazenediyl)] 4,40 bis-. hydrochloride (1:2) (Fig.1).

#### 2. Experimental

#### 2.1. Chemicals and Materials

Bismarck-Brown Y has been purchased from Merck Company with molecular formula and weight of  $C_{18}H_{20}Cl_2N_8$  and Mw 419.31 g mol<sup>-1</sup>, respectively (CAS Number: 10114-58-6). In addition, graphite fine powder and DCC (N, N'- Dicyclohexylcarbodiimide, CAS Number: 538-75-0) were from Merck company. Stock solutions from all dyes was prepared by dissolving (1000 mg L<sup>-1</sup>) in distilled water. PH was adjusted by 0.1- 0.01 mol L<sup>-1</sup> from two solutions NaOH (CAS Number:1310-73-2, Merck) and HCl (GCC). Potassium chlorate was purchased from Sigma-Aldrich. According to each manufacturing company, the liquid solvents have been listed as follows: pure ethanol (J.T. Baker), pure methanol (Himedia),



Fig. 1. Chemical Structure of Bismarck Brown-Y

dimethylsulfoxide (Himedia), dimethylformamide (GCC), nitric acid (GCC) and sulfuric acid (J.T. Baker). The identification of multi-walled carbon nanotubes (MWCNTs) and their derivatives was achieved through the utilization of various analytical techniques, including Fourier-transform infrared spectroscopy (FT-IR), X-ray diffraction (XRD), transmission electron microscopy (TEM), and scanning electron microscopy (SEM). The absorbance measurement for the BB-Y dye solution was conducted at a specific wavelength of 455.5 nm, utilizing a UVvisible spectrophotometer (PG Instrument T80 + UV-Vis). The dye removal efficiency (R%) is expressed as a percentage and the quantity of BB-y dye absorbed per unit weight of absorbent at a given time. Also, q  $(mg g^{-1})$  was determined using Equation 1.

$$q_{a} = (C_{0} - C_{a}) \times V/m \qquad (Eq.1)$$

 $C_{e}$  referred to the concentration of Bismarck Brown-y in (mg L<sup>-1</sup>) at time t, whereas  $C_{0}$  pointed out the initial concentration in (mgL<sup>-1</sup>) of the same dye. However, V represents the volume of dye in a (L), while m is the weight of the solid phase in (g).

# 2.2. Preparation of Multi-Walled Carbon Nanotube

The modified Staudenmaier approach was used in this investigation to create MWCNTs. The graphite powder was added based on stirring to the mixture which consisted of concentration of H<sub>2</sub>SO<sub>4</sub> and HNO<sub>2</sub> in (1.0 g: 10v: 5v) ratio (mass: volume: volume) for 30 min, then cooling the solution at  $5C^0$  in the ice bath with forcefully stirring. KClO<sub>2</sub> with ratio to graphite (5: 1) (mass: mass) was slowly added to the solution and gradually increased the heat until 75 C<sup>0</sup> and stayed overnight. After that, the solution was left in dry air for three days. Most of the graphite precipitated on the bottom. However, some reacting carbons were floating. One liter of DI water was added to the floating carbon particles after collecting them with one hour of vigorous stirring, and then the solution was rapidly filtered and dried [28].



Fig. 2. Step of preparation of multi-walled carbon nanotube

#### 2.3. Purification of MWCNTs

A 0.05 g of impure MWCNTs was heated at 350  $^{\circ}$  for two hours to eliminate amorphous carbon and catalyst impurities. After being cooled to room temperature for more emphasis to eliminate additional impurities, the heat-treated MWCNTs were ultrasonically for 4 hours in 20 mL of concentrated HCl then repeatedly rinsed with water until the pH became neutral, and then dried at 100 C° in an air oven.

#### 2.4. Functionalization of MWCNTs

Nitric acid and sulfuric acid were mixed to perform chemical oxidation, where 0.1 g of pristine MWCNTs was added to a mixture consisting of 4.0 M and 10.0 M, respectively, with an ultimate volume of 20 mL in the ratio (1:3) to reduce the damage of the nanotubes. The mixture was stirred magnetically at room temperature for 18 hours.

# **2.5.** Modification of MWCNT-COOH with

### Amino compound

Carboxyl groups, which allotrope in the structure of MWCNTs, interacted with amine groups to form a new group known as an amide group. Consequently, 1.0 g of MWCNT-COOH was taken and placed in a beaker (100 mL), then 1.0 g of DCC was poured into the beaker after being dissolved in 20 mL of DMF solvent. The mixtures were left at room temperature with constant stirring for 30 minutes. In the following step, 60 mL of methanolic solution was added to the mixture above, which already contained 1.0 g of either 4, 4'- Methylenedianiline, tris-(hydroxymethyl) aminomethane and tetraethylenepentamine or 2, 6-diamine pyridine. The solution is kept at 60 C° for 24

hours with continuous stirring. The resultant mixture was centrifuged at 2500 rpm to separate it. After multiple washes with DMF-Methanol solution and DW, the black precipitate was produced after drying it for 10 hours at 70°C [29].

#### 2.6. Solid Phase Extraction

Three basic steps were applied in the solid phase extraction (SPE) technique: column preparation step, loading step, and elution step [30]. The polypropylene tube (injection tube) was utilized in this study as a column (cartridge), as shown in Figure 3. The column was packed with a one mm thickness of permeable film from polypropylene (disc), followed by four layers of glass filter paper sheets. Subsequently, the column was loaded with an amount weight of the prepared solid phase, in this study, which was either MWCNT, MWCNT/G, MWCNT/Tris, MWCNT/ Tetra, or MWCNT/H. The surface of the packed solid phase should be homogeneous and plane with equal height. Moreover, as a last step in the column preparation, 10 ml of ionic water was passed through the column to activate the phase and remove air space. Whereas, in the second step (loading step), the dye solution under the study with neutral  $pH_{nzc}$  was passed through the packed solid phase, where the dye will attach with the solid phase in the column, while the unbounding part of the dye and the rest of matrix will pass through the column with flow rate depends on gravity. Finally, and as the last step (elution step), which involved passing the elution solution through the column to rid out the dye from the packed solid phase, the solution was collected to determine the concentration of dye by UV-visible instrument.



Fig.3. General procedure based on solid phase extraction by MWCNTs, MWCNTs/G, MWCNTs/Tris, MWCNTs/Tetra

# 2.7. Characterization Methods

To evaluate the surface properties of multi-walled carbon nanotubes (MWCNTs) and their derivatives (MWCNT/G, MWCNT/Tris, MWCNT/Tetra, or MWCNT/H), various analytical techniques, including Fourier-transform infrared spectroscopy (FTIR), X-ray diffraction (XRD, Fig. 4a-e), scanning electron microscopy for MWCNT and their derivatives (SEM, Fig. 5a-d), and zeta potential were employed for analysis. Fourier Transform Infrared (FT-IR) spectroscopy was employed to ascertain the sort and characteristics of the functional groups present in the synthesized solid phases (Tables 1 and 2). Including these functional categories enhances diversity and, consequently, the extraction process.



Fig. 4. XRD pattern (a) MWCNT-COOH, (b) MWCNT-Tris, (c) MWCNT-H, (d) MWCNT-Tera and (e) MWCNT-G





Fig. 5c. FE-SEM MWCNT-Tetra



Fig. 5d. FE-SEM of MWCNT-G

	Table 1. FT-IR peaks in (cm <sup>-1</sup> ) of MWCNTs and their Derivatives					
Groups	MWCNTs	<b>MWCNTs-tris</b>	MWCNTs-H	MWCNTs-tera	MWCNTs-G	
-OH	3534	3630				
-NH		3198-3140	3429-3244	3591-3445	3491-3419	
-CH <sub>2</sub>	2940-2847	2904-2862	2940-2847	2940-2847	2940-2847	
C=C	1571	1550	1593	1590	1516	
C=O	1672	1710	1759	1793	1743	
C-N		1109	1128	1392	1645	

Table 2. Zeta Potential for MWCNTs and their Derivatives

Potential	MWCNT	<b>MWCNT-tris</b>	MWCNT-H	MWCNT-tera	MWCNT-G
Zeta	-36mV	-123.2mV	-46mV	-48mV	-36mV

# 3. Results and Discussion

#### 3.1. Optimization of the extraction procedure

The study for identifying the most optimal conditions for a given analytical technique involves manipulating one condition of the experiment while keeping the other factors constant. Consequently, to determine the optimal values for all components and identify the perfect conditions for achieving maximum efficiency in the process of extraction and removal of the dye [31]. The several types of investigations were executed as follows:

#### 3.1.1. Amount of solid phase

The quantity weight of MWCNT, MWCNT/G, MWCNT/Tris, MWCNT/Tetra, or MWCNT/H was examined in this study as solid phase extraction in separation column with different range between 5-50 mg. Meanwhile, the other conditions must be fixed, as mentioned in the general method. Figure 6 shows that increasing the weight of the solid phase augments the retrieval percentage, where the recovery percentage achieves the highest value while employing a weight of 0.01g. Subsequently, the recovery percentages reached a state of stability in the biggest weights, then descending to a minimum of 0.5 g, and this observed behavior was consistent throughout all solid phases during the investigation of the BB-y dye. Consequently, a constant weight of 0.01 g for the solid phase was chosen as the standard weight for all phases in the following tests.

#### 3.1.2. Effect of Dye concentration

The effect of the concentrations of BB-Y dye solution has been studied after passing different concentrations (50-700) mg  $L^{-1}$  from the day on 0.01 g of prepared solid phase, which is either one of MWCNT, MWCNT/G, MWCNT/Tris, MWCNT/Tetra, or MWCNT/H. It is essential to point out that all other experimental parameters, such as pH, dye volume, flow rate, the type and volume of elution solution, were maintained constant, as mentioned in the general procedure, where the perfect concentrations of a dye solution were 200 mg  $L^{\text{-1}}$  and 300 mg  $L^{\text{-1}}$  for both MWCNT and MWCNT-Tris, whereas 400 mg L<sup>-1</sup> for each of MWCNT-H, MWCNT-Terta and MWCNT-G respectively. The recovery percentage of all solid phases of MWCNTCOOH, MWCNT-Tris, MWCNT-H, MWCNT-Tetra, and MWCNT-G was obtained 60%, 63%, 62%, 66%, and 72%, respectively for the dye under the study. The proper utilization of this ratio has significance because the recovery efficiency wasn't at its highest point during this stage, and it is anticipated that it will increase by improving other conditions. Figure 7 demonstrates the effect of concentrations of BB-Y dye solution by proposed procedure.



Fig. 6. Effect of the solid phase amount on the recovery of Bismarck Brown-y



Fig. 7. Effect of Bismarck Brown-Y concentration on the recovery percentage

#### *3.1.3. Effect of pH*

The pH function is an essential factor in the assessment of extraction processes since it has a direct effect on the surface charge of the solid phase and the chemical structure of the dye. The effect of pH on solid phase extraction (SPE) was studied in this investigation from range 2 to12. Figure 8 revealed that increases the recovery percentage of cationic dye (BB-y) at pH=6 in MWCNT-Tris, whereas it was 10 for MWCNT, MWCNT-H, MWCNT-Terta, and MWCNT-G, respectively. The surface of multi-walled carbon nanotubes (MWCNTs) has a positive charge when the pH value is lower than 6.7. However, it becomes a negative charge when the pH exceeds 6.7. Therefore, the recovery ratio increases at pH 10 because of the electrostatic attraction, which accrues between the negative charges of the surface (MWCNTs) and the positive ions of dye BB-Y. On the other hand, the surface of MWCNTs in acid pH becomes the positive charge. The BB-Y converted to positive ions as a result of the protonation process, which leads to a decrease in the percentage of recovery and adsorption capacity because of electrostatic repulsion between the positive charge of the surface with the positive charge of the protonation dye [32]. As for the MWCNTs-Tris, which has three hydroxyl groups at pH 6, becomes a negative charge as zeta potential (-36 mV). Moreover, the protonation of dye leads to a charge in the nitrogen atom by the positive charge; all these reasons are due to an

increase in the adsorption capacity and recovery ratio at pH 6 (Eq. 2) [32].

Dye-NH<sub>2</sub> +H<sup>+</sup> 
$$\longrightarrow$$
 Dye-NH<sub>3</sub><sup>+</sup> ...... (Eq. 2)

However, if the pH is over 6, the dye solution becomes base, which leads to de-protonation of the nitrogen atom, and the MWCNT-tris converted more negatively as Equation 3.

$$-CH_2OH + OH^- \longrightarrow CH_2O^- + H_2O....$$
(Eq. 3)

Consequently, the recovery percentage and adsorption capacity decrease at a pH of more than 6 [33]. The recovery percentages for the three surfaces, MWCN-tetra, MWCN-H, and MWCN-G, are increasing at pH 10, which may be at higher pH levels. The functional groups present on the surface of the three surfaces above, such as N–H and C–H groups, can work as activation agents through negative charges. Consequently, it increased the quantity of negatively charged molecules. Therefore, the adsorption of BB-Y dye can potentially be ascribed to the significant electrostatic attraction between the charge on the three surfaces and the positively charged BB-Y molecules [34]



Fig. 8. Effect of pH on the recovery of Bismarck Brown-Y

# 3.1.4. Effect of dye volume

Investigating the impact of the volume of the solution, including the target material, is crucial in selecting the most favorable conditions for the solid-phase extraction approach [35][36]. Figure 9 illustrates the behavior of cationic dye BB-Y on five prepared surfaces, which were ordered at 74, 77, 79, 84, and 86 recoveries (%) for MWCNTCOOH, MWCNT-Tris, MWCNT-H, MWCNT-Tetra, and MWCNT-G, respectively. This study examined the different volumes of dye 1mL, 5 mL, 10 mL, 25 mL, 50 mL, and 75 mL at optimal conditions that have been obtained in previous experiments 0.01g of all surfaces with 200mg L<sup>-1</sup> and 300mg

L<sup>-1</sup> for MWCNT and MWCNT-Tris as well as 400mg L<sup>-1</sup> for MWCNT-H, MWCNT-Terta, and MWCNT-H, respectively. Besides, the remaining other conditions are without change as the general method. The Percentage of recovery showed a progressive reduction. The observed pattern can be attributed to a logical relationship between the extraction effectiveness and the solution's volume. Specifically, as the volume of the solution increases, the dye concentration falls, resulting in a decrease in extraction efficiency. On this fundamental, the dye will dilute with increasing the volume of the solution. Therefore, the optimal volume is 25mL for the cationic dye Bismarck brown –y.



Fig. 9. Effect of Bismarck Brown-Y solution volume on the recovery percentage



Fig.10. Effect of flow rate on the recovery of Bismarck Brown-Y

#### 3.1.5. Effect of flow rate

One of the most important parameters influencing the efficiency of solid phase extraction is the flow rate of the dye solution. Therefore, achieving a balance in the flow rate is crucial since low flow rates hinder the attainment of high rates of recovery for the target material. This limitation arises from the potential disengagement between the solid phase and the target material. Hence, the effect of flow rate on the percentage of recovery is influenced by the connected time between the dye from one side and the solid phase on the other. The recovery percentage decreases with increasing the flow rate of the dye solution, as demonstrated in Figure 10, where 0.5 ml min<sup>-1</sup> is the perfect flow rate time for the BB-Y solution.

# 3.1.6.Effect of type and volume of eluting solution

The impact of different types of solvents as eluent solvents was investigated in this paper, where these solvents were chosen depending on their polarities. The polarity coefficient reveals the solvent's capacity to interact with the solute [37]. Table 3 illustrates the polarity of eluent solvents in this study. Figure 11 shows the different values of the percentage of recovery and which ordered 93, 95, 96, 98, and 99 for five surfaces under the study: MWCNTCOOH,

MWCNT-Tris, MWCNT-H, MWCNT-Tetra, and MWCNT-G, respectively. Additionally, the investigation revealed a clear correlation between the polarity of the eluent solution and the percentage of the recovery, which means that increasing the polarity of the eluent solvent increases the recovery percentage. Consequently, DMSO recorded the highest percentage since the polarity coefficient is highest at about 7.2. In contrast, other eluent solvents decrease the percentage recovery by decreasing the polarity coefficient, as obvious in Table 3. The strong affinity of BB-Y dyes towards the DMSO solvent can be ascribed to the polar nature of the dye. The dye undergoes a phase transition, then transitions from a liquid to a solid state, and subsequently migrates with the rinse solution that possesses a higher polarity (see Figure 11). Moreover, the volume of elution solvent was investigated in this study where the 5 mL of DMSO represented the perfect volume for disengagement of the cationic dye from the solid phase, as revealed in Figure 12, which illustrated the relationship between the optimal volume of elution solvent and the percentage recovery. Besides, the optimal volume of elution solvent can be calculated, by enrichment factor by Equation 4 [38].

> Enrichment Factor (EF)=Original dye volume/ Elution volume (Eq.4)



Fig 11. Effect of elution type on the recovery of Bismack Brown-Y

Solvent	<b>Polarity Index</b>			
DMSO	7.2			
Methanol	5.1			
Ethanol	4.3			
Toluene	2.4			
n-hexane	0.1			

 Table 3. Polarity of Different Eluents that used in this study



Fig. 12. Effect of DMSO volume on the recovery of Bismarck Brown-Y

#### 3.2. Isotherm study

The SPE techniques are regarded as better than other procedures due to their ease of use, flexibility to remove unwanted matrix ingredients, and capacity to attain high enrichment factors [39]. The adsorption isotherm experiments were conducted using BB-Y solutions of specific concentrations under the indicated optimal experimental conditions. By analyzing the isotherm, it becomes feasible to elucidate the correlation between the solid phases and dye and propose the underlying mechanisms of interaction [40]. Two models were used in this study to explain the adsorption isothermal comportment, which is the Langmuir model and the Freundlich model.

#### 3.2.1. Langmuir isotherm model

The Langmuir model is reliant upon maximum adsorption, which corresponds to the emergence of a saturated monolayer of adsorbate (liquid molecules) on the adsorbent (solid surface). Equation 5 presupposes the presence of homogenous adsorption sites.

$$C_o/qe=1/q_{max} K_L+1/q_{max}$$
(Eq.5)

Figure 13 illustrates the Langmuir model for BB-Y, where the  $(1/q_{max})$  refers to a straight line. However, the cutoff pointed out  $(1/q_{max} \times K_L)$ . Consequently, the maximum adsorption capacity  $(q_{max})$ , Langmuir constant  $(K_L)$ , and correlation coefficient  $(R^2)$  were determined by the graphical representation of the Langmuir equation. This involved charting the relationship between  $C_e/q_e$  on the Y-axis and  $C_e$  on the X-axis, as mentioned in Table 4. The outcomes of the maximum adsorption capacity of BB-Y dye by solid phase (MWCNT-G) is the highest value of the others.

### 3.2.2. Freundlich isotherm model

The equation elucidates the phenomena of adsorption and interference occurring on heterogeneous surfaces, if adsorption occurs at active sites that have varying



Fig. 13. Langmuir isotherm of adsorption of Bismarck Brown-y

Solid Phase	q <sub>max</sub> (mg g <sup>-1</sup> )	K	R <sup>2</sup>
MWCNT	862.068	0.098	0.9997
MWCNT-Tris	1075.268	0.136	0.9999
MWCNT-H	1282.051	0.169	0.9999
MWCNT-T	1298.701	0.275	0.9997
MWCNT-G	1333.333	0.234	0.9941

Table 4. Langmuir isotherm parameters for the adsorption of CR dye

adsorption energies. The Freundlich equation is a mathematical model used to describe the absorption of solutes onto solid surfaces (Eq.6).

$$\ln q_e = \ln K_F + 1/n \times \ln C_e$$
 (Eq.6)

By graphing the Freundlich equation between  $lnq_e$  on the Y-axis and  $lnC_e$  on the X-axis as in the illustration in Figure 14, the values of the Freundlich constant  $K_F$  and correlation coefficient  $R^2$  were determined as elucidated in Table 5, where the slop of straight line refer to 1/n. However, the cutoff refers to  $lnK_F$ . The value of  $K_F$  is highest in MWCNT-G, which refers to the adsorption energy between the prepared solid phase and BB-y [30]. As a result, this outcome is compatible with the  $q_{max}$  value derived from the Langmuir equation.

The values of 1/n provide information about the preference or unfavorable of the adsorption process. A value of 1/n = 0 implies irreversible adsorption, while a value between 0 and 1 indicates preferable adsorption between the solid phase and target material. On the other hand, when 1/n > 1, adsorption may not be favorable. As observed in Table 5 the values of 1/n for all prepared solid phases are more than zero and less than one, which indicates the adsorption of all surfaces is preference. According to the values  $R^2$  which had been obtained from Tables 5 and 4, Langmuir model is higher than the Freundlich model. Therefore, adsorption is a chemisorption mechanism.

#### 4. Conclusion

The optimized method for extraction processes was conducted in this study, where the amount of solid-



Fig. 14. Freundlich isotherm of adsorption of Bismarck Brown-y

<b>Table 5.</b> Freundlich isotherm param	eters for the adsor	ption of BB-y	y at 25 °C
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	*	-	•
Solid Phase	K <sub>F</sub>	1/n	<b>R</b> <sup>2</sup>
MWCNT	335.452377	0.16779	0.8345
MWCNT-Tris	529.3871411	0.12981	0.89398
MWCNT-H	731.8898321	0.10382	0.96162
MWCNT-Tetra	823.6535562	0.08807	0.97272
MWCNT-G	890.3280586	0.07441	0.9385

phase extraction was 0.01g for all surfaces. However, the optimal concentration for Bismarck brown-y was 200 mg L<sup>-1</sup> of MWCNT and MWCN- tris while the optimal concentration was 400 mg L<sup>-1</sup> for each prepared solid phase MWCNT-H, MWCNT-tetra, and MWCNT-G. The perfect pH was 6 for MWCNTtris, and for the rest surfaces the pH was 10. The volume of dye was studied which was 25 mL. In addition, the flow rate was 0.5 ml min<sup>-1</sup> in perfect condition. Furthermore, the type of eluent solution and the optimal volume were DMSO and 5ml from it. Finally, the  $q_{max}$  was obtained at 862.0691, 1075.2689, 1282.0513, 1298.7013, and 1333.33 for the MWCNT, MWCNT-tris, MWCNT-H, MWCNT-Tetra, and MWCNT-G, respectively.

# **5. References**

- [1] A. Balasubramanian, P. A. Balasubramanian, The world's water geochemical modelling of groundwater for prevention of incrustation in the water supply systems of salem district, Tamil Nadu, Indian, View project educational video documentaries in earth, atmospheric and ocean sciences view project, 2017. https:// www.researchgate.net/publication/315123891
- [2] P. Senthil Kumar, A critical review on recent developments in the low-cost adsorption of dyes from wastewater, Desalin. Water Treat., 172 (2019) 395–416. https://doi.org/ 10.5004/ dwt.2019.24613
- [3] J. C. G. Sousa, A. R. Ribeiro, M. O. Barbosa, M. F. R. Pereira, A. M. T. Silva, A review on environmental monitoring of water organic pollutants identified by EU guidelines, J. Hazard. Mater., 344 (2018) 146–162. https:// doi.org/10.1016/j.jhazmat.2017.09.058
- [4] F. F. A. Aziz, A. A. Jalil, S. Triwahyono, M. Mohamed, Controllable structure of fibrous SiO<sub>2</sub> -ZSM<sub>5</sub> support decorated with TiO<sub>2</sub> catalysts for enhanced photodegradation of paracetamol, Appl. Surf. Sci., 455 (2018) 84–95. https://doi.org/10.1016/j.apsusc.2018.05.183
- [5] A. Kumar Samanta, P. Agarwal, Application of natural dyes on textiles, India. J. Fibre Texile Res., 34 (2009) 384-399. https://nopr.niscpr. res.in/bitstream/123456789/6886/1/IJFTR%20 34(4)%20384-399.pdf
- [6] F. Mcyotto, Q. Wei, D. K. Macharia, M. Huang, C. Shen, C. W. K. Chow, Effect of dye structure on color removal efficiency by coagulation, Chem. Eng. J., 405 (2021) 126674. https://doi. org/10.1016/j.cej.2020.126674
- [7] Q. Wei, F. O. Mcyotto, C. W. K. Chow,

Z. Nadeem, Z. Li, J. Liu, Eco-friendly decolorization of cationic dyes by coagulation using natural coagulant Bentonite and biodegradable flocculant sodium Alginate, SDRP J. Earth Sci. Environ. Stud., 5 (2020) 51–60. https://doi.org/ 10.25177/ jeses.5.2.ra.10648

- [8] P. Arulmathi, C. Jeyaprabha, P. Sivasankar, V. Rajkumar, Treatment of textile wastewater by coagulation–flocculation process using gossypiumherbaceumandpolyanilinecoagulants, Clean Soil, Air, Water, 47 (2019) 1800464. https://doi.org/10.1002/clen.201800464
- [9] N. Tara, S. I. Siddiqui, G. Rathi, S. A. Chaudhry, Inamuddin, A. M. Asiri, Nano-engineered adsorbent for the removal of dyes from water: A review, Curr. Anal. Chem., 16 (2019) 14–40. https://doi.org/10.2174/157341101566619011 7124344
- [10] S. P. Keerthana, R. Yuvakkumar, P. S. Kumar, G. Ravi, D. V. N. Vo, D. Velauthapillai, Influence of tin (Sn) doping on Co<sub>3</sub>O<sub>4</sub> for enhanced photocatalytic dye degradation, Chemosphere, 277 (2021) 130325. https://doi.org/10.1016/j. chemosphere.2021.130325
- [11] S. Varjani, P. Rakholiya, H. Y. Ng, S. You, J. A. Teixeira, Microbial degradation of dyes: An overview, Bioresour. Technol., 314(2020) 123728. https://doi.org/10.1016/j.biortech.2020.123728
- [12] G. Crini, Non-conventional low-cost adsorbents for dye removal: A review, Bioresour. Technol., 97 (2006) 1061–1085. https://doi.org/10.1016/j.biortech.2005.05.001
- [13] V. K. Gupta, Suhas, Application of low-cost adsorbents for dyeremoval-Areview, J. Environ. Manage., 90 (2009) 2313–2342 . https://doi. org/10.1016/j.jenvman.2008.11.017.
- [14] S. M. Ghoreishi, R. Haghighi, Chemical catalytic reaction and biological oxidation for treatment of non-biodegradable textile effluent, Chem. Eng. J., 95 (2003) 163–169. https://doi. org/10.1016/S1385-8947(03)00100-1
- [15] M. Rezaei, A. Habibi-Yangjeh, Simple and large scale refluxing method for preparation of Ce-doped ZnO nanostructures as highly efficient photocatalyst, Appl. Surf. Sci., 265 (2013) 591–596. https://doi.org/10.1016/j. apsusc.2012.11.053
- [16] B. Mu, A. Wang, Adsorption of dyes onto palygorskite and its composites: A review, J. Environ. Chem. Eng., 4 (2016) 1274–1294. https://doi.org/10.1016/j.jece.2016.01.036
- [17] X. Hou, S. Tang, J. Wang, J Wang, Recent

advances and applications of graphenebased extraction, Trends Anal. Chem., 119 (2019) 115603. https://doi.org/ 10.1016/j. trac.2019.07.014

- [18] E. Dziurkowska, M. Wesolowski, Solid phase extraction purification of saliva samples for antipsychotic drug quantitation, Molecules., 23 (2018) 2946. https://doi.org/ 10.3390/ molecules23112946
- [19] Y. Wen, L. Chen, J. Li, D. Liu, L. Chen, Recent advances in solid-phase sorbents for sample preparation prior to chromatographic analysis, TrAC Trends Anal. Chem., 59 (2014) 26–41. https://doi.org/10.1016/j.trac.2014.03.011
- [20] S. Di, Recent advances and applications of magnetic nanomaterials in environmental sample analysis, TrAC Trends Anal. Chem., 126 (2020) 115864. https://doi.org/10.1016/j. trac.2020.115864
- [21] C. Jamshidzadeh, A new analytical method based on bismuth oxide-fullerene nanoparticles and photocatalytic oxidation technique for toluene removal from workplace air, Anal. Methods Environ. Chem. J. 2 (2019) 73-86. https://doi.org/10.24200/amecj.v2.i01.55
- [22] M Arjomandi, A review: analytical methods for heavy metals determination in environment and human samples, Anal. Methods Environ. Chem. J., 2 (2019) 97-126. https://doi. org/10.24200/amecj.v2.i03.73
- [23] R. Ashouri, Dynamic and static removal of benzene from air based on task-specific ionic liquid coated on MWCNTs by sorbent tubeheadspace solid-phase extraction procedure, Int. J. Environ. Sci. Technol., 18 (2021) 2377-2390. https://doi.org/10.1007/s13762-020-02995-4
- [24] J. Rakhtshah, A rapid extraction of toxic styrene from water and wastewater samples based on hydroxyethyl methylimidazolium tetrafluoroborate immobilized on MWCNTs by ultra-assisted dispersive cyclic conjugationmicro-solid phase extraction, Microchem. J., 170 (2021) 106759. https://doi.org/10.1016/j. microc.2021.106759
- [25] S. Teimoori, An immobilization of aminopropyl trimethoxysilane-phenanthrene carbaldehyde on graphene oxide for toluene extraction and separation in water samples, Chemosphere, 316 (2023) 137800. https://doi.org/10.1016/j. chemosphere.2023.137800
- [26] S. Teimoori, A.H. Hassani, M. Panahi, N. Mansouri, Rapid extraction of BTEX in water

and milk samples based on functionalized multi-walled carbon nanotubes by dispersive homogenized-micro-solid phase extraction, Food Chem., 421 (2023) 136229. https://doi. org/10.1016/j.foodchem.2023.136229

- [27] S. Teimoori, A.H. Hassani, New extraction of toluene from water samples based on nanocarbon structure before determination by gas chromatography, Int. J. Environ. Sci. Technol., 20 (2023) 6589–6608. https://doi.org/10.1007/ s13762-023-04906-9
- [28] B. Ali, Preparation of carbon nanotubes via chemical technique (modified staudenmaier method), Nanosci. Nanotechnol. Asia, 7 (2017) 113–122. https://doi.org/10.2174/2210681206 666160711161421
- [29] Z. B. Ali Abdulnabi, H. T. Abdulsahib Faris, A J Al-doghachi, Synthesis and characterization of some selenazone complexes and nanoadsorbent surfaces from industrial waste for removing some carcinogenic dyes and heavy metals from water, proposal, 2021. https://faculty.uobasrah. edu.iq/uploads/publications/1643354925.pdf.
- [30] M. H. Abdel-Aziz, DFT and experimental study on adsorption of dyes on activated carbon prepared from apple leaves, Carbon Lett., 31 (2021) 863–878. https://doi.org/10.1007/ s42823-020-00187-1
- [31] T. N. Majid, A. A. Abdulwahid, An efficient cheap source of activated carbon as solid phases for extraction and removal of Congo Red from aqueous solutions, Anal. Methods Environ. Chem. J., 5 (2022) 40–54. https://doi. org/10.24200/amecj.v5.i03.205
- [32] M. I. Mohammed, S. Baytak, Synthesis of bentonite–carbon nanotube nanocomposite and its adsorption of rhodamine dye From water, Arab. J. Sci. Eng., 41 (2016) 4775–4785. https://doi.org/ 10.1007/s13369-016-2190-7
- [33] G. Z. Kyzas, N. K. Lazaridis, Reactive and basic dyes removal by sorption onto chitosan derivatives, J. Colloid Interface Sci., 331 (2009) 32–39. https://doi.org/10.1016/j.jcis.2008.11.003
- [34] G.Z.Kyzas, D.N.Bikiaris, N.K. Lazaridis, Lowswelling chitosan derivatives as biosorbents for basic dyes, Langmuir., 24 (2008) 4791–4799. https://doi.org/10.1021/la7039064
- [35] A. A. Gouda, W. A. Zordok, Solid-phase extraction method for preconcentration of cadmium and lead in environmental samples using multiwalled carbon nanotubes, Turk. J. Chem., 42 (2018) 1018–1031. https://doi. org/10.3906/kim-1711-90

- [36] M. Hossein, N. Dalali, A. Karimi, K. Dastanra, Solid phase extraction of copper, nickel, and cobalt in water samples after extraction using surfactant coated alumina modified with indane-1, 2, 3-trione 1, 2-dioxime and determination by flame atomic absorption spectrometry, Turk. J. Chem., 34 (2010) 805– 814. https://doi.org/10.3906/kim-1002-22
- [37] L. R. Snyder, Classification off the solvent properties of common liquids, J. Chromatogr. Sci., 16 (1978) 223–234. https://doi. org/10.1093/chromsci/16.6.223
- [38] Y. Li, W. Zhang, R. G. Wang, P. L. Wang, X. O. Su, Development of a efficient and sensitive dispersive liquid-liquid microextraction technique for extraction and preconcentration of 10 β2-Agonists in animal urine, PLOS One, 10 (2015) e0137194. https://doi.org/10.1371/journal. pone.0137194
- [39] G. Daneshvar Tarigh, F. Shemirani, Simultaneous in situ derivatization and ultrasound-assisted dispersive magnetic solid phase extraction for thiamine determination by spectrofluorimetry, Talanta., 123 (2014) 71–77. https://doi. org/10.1016/j.talanta.2014.01.045
- [40] E. Yilmaz, G. Guzel Kaya, H. Deveci, Removal of methylene blue dye from aqueous solution by semi-interpenetrating polymer network hybrid hydrogel: Optimization through Taguchi method, J. Polymer Sci., Part A: Polymer Chem., 57 (2019) 1070–1078. https://doi.org/ 10.1002/pola.29361



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# Analytical Method: Determination of famotidine drug using chemiluminescence method

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#### **ABSTRACT**

This study involved the development of a novel, cost-effective, fast, and highly sensitive analytical technique for quantifying minimal amounts of the drug famotidine through chemiluminescence. The method is centred around the measurement of energy emitted as a result of the interaction between the drug and Luminol in an alkaline solution; this interaction generates an electronically excited intermediate state, releasing a portion of the system's energy as photons. The method was sensitive for the analysis of famotidine. The linear calibration curve (LR) is obtained in the range 2-12 mg mL<sup>-</sup> <sup>1</sup>, with a high correlation coefficient ( $\mathbb{R}^2$ ) of 0.9929. The molecular absorption coefficient ( $\epsilon$ ) was calculated at 2621×10<sup>4</sup> L mol<sup>-1</sup> cm<sup>-1</sup>. The method displayed excellent sensitivity with a Sandell's sensitivity of  $1.287 \times 10^{-5}$  mg cm<sup>-2</sup>, the detection limit (LOD) was found to be 0.0314 mg mL<sup>-1,</sup> and the limit of quantification (LOQ) was 0.0952 mg mL<sup>-1.</sup> This study found that recovery was obtained at 104 - 96.5 %, and the relative standard deviation (RSD%) was below 1.981%. The results showed that the proposed technique has efficient recovery for measuring famotidine in pharmaceutical preparations.



# C8H15N7O2S3

# M.W. 337.45

Scheme 1. The chemical formula of famotidine

# 1. Introduction

Famotidine is a crystalline substance with a white to light yellow appearance. It is almost insoluble in ethanol and has a high solubility in glacial acetic acid, a mild solubility in methanol, and an extremely

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slight solubility in water. The active component in famotidine injection acts as a histamine H2 receptor antagonist and has the chemical name [1-Amino-3-[2-[(diaminomethylene)amino]-4-thiazolyl]-methyl] thio] propylidene] sulfamide [1]. Famotidine is commonly known by its generic name, which is "Famotidine" (pronounced fam OH ti deen), and it is available under the trade name "Pepcid." (Scheme1) [2,3].

Famotidine is used as a treatment for gastrointestinal ulcers and the regulation of acid secretion in the digestive system. Its therapeutic applications extend to addressing the Zollinger-Ellison syndrome, a condition characterized by excessive gastrin secretion, leading to heightened acid production and ulcer development. Functioning as a histamine-2 receptor inhibitor, famotidine effectively diminishes the concentration and volume of gastric acids [3-5]. This medication finds utility in various gastric hyperplasia cases, including Zollinger-Ellison syndrome, active gastroesophageal gastric ulcers. reflux. and esophagitis resulting from reflux. Additionally, famotidine is employed for the treatment of active duodenal ulcers, gastric ulcers, heartburn, acid indigestion, and sour stomach. Administration of the drug can be accomplished through oral ingestion or intravenous injection[6]. Chemiluminescence is the phenomenon of a substance emitting light spontaneously, without the involvement of heat; hence, it is often referred to as "cold light" [7]. This term encompasses all processes wherein materials can convert various forms of energy into visible light [8]. In simpler terms, it encompasses any process that releases energy in the form of visible light [9]. Chemiluminescence is the generation of light produced as a consequence of a chemical reaction. Occasionally referred to as "chemiluminescence," this phenomenon doesn't exclusively involve the emission of light; there may also be a concurrent release of heat, rendering the reaction exothermic. In the process, two chemicals undergo a reaction, forming an excited (high-energy) intermediate. This intermediate subsequently breaks down, releasing a portion of its energy in the form of light photons as it returns to its ground state [10]. When the emission of light arises from a chemical reaction occurring within a living organism, the term used is "bioluminescence." In this context, the light produced is a result of a chemical reaction taking place in vivo[11,12]. The accurate detection of ranitidine and famotidine hydrochloride in pharmaceutical formulations has been made possible by the development, validation, and application of a fast and extremely sensitive highperformance liquid chromatographic (HPLC) method [13]. Four distinct medication classes can be analyzed using a unique capillary gas chromatography (GC) method: metformin, ranitidine, cimetidine, and fumarate. Methylglyoxal (MGo) is a prerequisite for this analytical technique, which improves the precision and effectiveness of the inspection procedure [14]. A technique for identifying cimetidine, famotidine, and ranitidine hydrochloride was

developed that is distinguished by its selectivity, precision, and accuracy. This method, which combines scanning densitometry with high-performance thinlayer chromatography (HPTLC), can be used for both pure and dose forms [15, 16]. The detection of famotidine plasma concentrations has been made possible by the development of a fast and accurate high-performance liquid chromatography (HPLC) technology that uses a monolithic column. Additionally, famotidine and ibuprofen have been simultaneously determined in composition using a validated reversephase HPLC[17]. A comprehensive comparison analysis was carried out to assess the pharmacological determination of diclofenac sodium utilizing two distinct analytical methods: high-performance liquid chromatography (HPLC) flow-injection and chemiluminescence<sup>[18]</sup>. The Flow Injection Chemiluminescence (FI-CL) method was used to determine the presence of famotidine, Diclofenac sodium (Voltaren), and Ethambutol HCl as active components and in pharmaceutical formulations [19]. Three H2-receptor antagonists have been identified using a novel, very sensitive liquid-injection chemiluminescence technique that has been designed, tested, and used: cimetidine, ranitidine hydrochloride, and famotidine. This technique is dependent on light intensity. This technique exhibits encouraging promise for precise and trustworthy screening of these medicinal substances [20]. Three newly created UV spectrophotometric techniques that make it easier, more affordable, sensitive, and accurate to determine the presence of famotidine in tablet and bulk drug formulations are described. These techniques offer useful methodological benefits for the quantitative pharmacokinetic analysis of famotidine [21]. Alamgir introduced a novel high-performance liquid chromatography (HPLC) technique in 2017 [22]. This approach, which is intended for the quantification of metformin, famotidine, and ranitidine, involves the pre-derivatization of columns using benzoin. Additional research also concentrated on the pharmacological determination of ranitidine-HCl through the use of active chemiluminescence, flow injection, spectrophotometric, and kinetic techniques. These analytical methods add to the vast array of accurate and thorough instruments for chemical analysis that are now accessible<sup>[23]</sup>. Ranitidine and famotidine levels in serum, urine, and pharmaceutical formulations can be directly determined using the capillary electrophoresis CE technique [24]. For the measurement of captopril, a novel flow-injection Chemiluminescence (FI-CL) approach without the need for a Chemiluminescence reagent has been

developed. This technique takes advantage of the fact that captopril increases chemiluminescence, which is mostly obtained from the system diperiodatoargentate (III)-sulfuric acid. This creative method provides a special and practical option for the accurate measurement of captopril in analytical applications [25, 26]. Penicillin antibiotics in medications and human urine can now be found using a flow-injection chemiluminescence approach. By making use of the luminol-Ag (III) complex system, this technique offers a sensitive and effective way to analyze penicillin antibiotics in a variety of matrices [27]. A technique that combines flow-injection chemiluminescence (FI-CL) and microdialysis has been developed to ascertain the binding characteristics of a medication that interacts with a protein. This novel method offers insights into the kinetics of drug-protein interaction by using the binding of the antibiotic tetracycline hydrochloride to bovine serum albumin as a model system [28]. A chemiluminescence technique that is both quick and effective has been created to measure and examine the pharmacokinetics of paclitaxel in rat plasma. Paclitaxel levels may be quickly and efficiently analyzed with this technology, which advances our knowledge of the drug's pharmacokinetics in biological materials<sup>[29]</sup>. The development of quick and reliable techniques for wastewater analysis is becoming more and more important due to the toxicological effects of several pollutants that are frequently found in wastewater from a variety of sources, including industrial and urban sources. Because of its great sensitivity, the chemiluminescence technique is used in wastewater analysis to identify and measure various contaminants. This method supports efforts to analyze and protect the environment by enabling sensitive and effective monitoring of water quality[30, 31]. Several nanomaterial-assisted chemiluminescence systems have been developed to increase sensitivity and expand their analytical uses. An overview of recent developments in electrochemiluminescence that make use of nanotechnology is provided, with a focus on analytical applications. This is important because it growing demonstrates the contribution of nanotechnology to the advancement of analytical techniques and includes applications in immunoassays, DNA analysis, and other biological analyses [32]. The primary goals of the present study are to use the chemiluminescence approach to estimate tiny amounts of famotidine and to establish an analytical method that is sensitive, quick, and inexpensive for detecting famotidine in pharmaceutical preparations. This method aims to measure the energy emitted during the chemiluminescent reaction between famotidine and

Luminol, establish a calibration curve with a high correlation coefficient, determine the molecular absorption coefficient, assess Sandell's sensitivity, establish the detection limit, evaluate the quantumlimited sensitivity, and determine the recovery rate while maintaining precision with a relative standard deviation not exceeding 1.981%. Furthermore, this developed method is intended for successful application in pharmaceutical analysis.

# 2. Material and Methods

#### 2.1. Instruments

Chemiluminescence measurements are performed Lumat LB 9507. Tube Luminometer, Firmware Version 5.03 (Berthold Technologies GmbH & Co.KG, Germany), and using 5ml Glass test tube.

#### 2.2. Reagents

Sodium carbonate (Na<sub>2</sub>CO<sub>3</sub>), which has a CAS number of 497-19-8 and is sourced from the USA, was purchased from Honeywell Fluka to be used as reagents in this investigation. An essential part of our experimental setup, famotidine, was obtained from Sammarra Drugs in Iraq (SDI). The hydrogen peroxide (CAS number: 7722-84-1) is necessary for oxidative processes, was acquired from Sigma-Aldrich® and originated in Germany. Furthermore, Laminol, a crucial stabilizing agent, was purchased from BDH and acquired from the UK with a CAS number of 521-31-3. Each reagent underwent stringent quality control procedures to guarantee the precision and dependability of our experimental findings.

#### 2.3. Solutions

1.060 g of pure material (Sodium Carbonate1.0×10<sup>-</sup> <sup>1</sup>M) was dissolved in a particular amount of distilled water to create a solution. In a volumetric flask, the solution was then added to distilled water until it reached a final volume of 100 mL. To create this solution, 0.01 g of the medication famotidine (100 mg mL<sup>-1</sup>) was dissolved in distilled water, and the volume was then completed in a volumetric flask at 100 mL. The process involves dissolving 0.1771 g of Luminol  $1.0 \times 10^{-3}$  M) in a specific volume of a  $1.0 \times 10^{-1}$ molar sodium carbonate solution. The solution is then adjusted to a total volume of 100 mL in a volumetric flask until the pH reaches 10.5. The preparation of this solution involved diluting 4.572 ml of hydrogen peroxide (1.0 M) in a volume of distilled water and then filling a volumetric flask to the full 100 mL with distilled water(DW). To prepare the solution of the pharmaceutical composition at 100 mg mL<sup>-1</sup>

concentration, a tablet equivalent to one pill, weighing 0.1495 g was taken. It contained 20 mL of dissolved famotidine. This solution was filtered and then adjusted to a total volume of 100 mL in a volumetric flask, resulting in a concentration of 200 mg mL<sup>-1</sup>. Subsequently, 50 mL of the prepared solution was transferred to a 100 mL container and topped up to the mark with DW, yielding a concentration of 100 mg mL<sup>-1</sup>. Additional concentrations were created by further dilution.

#### 2.4. General Procedure

To develop a new and rapid analytical method for the first time to determine the famotidine drug using the chemiluminescence technique based on the chemical reaction between Luminol and the drug in the presence of the hydrogen peroxide solution as a catalyst to produce an intermediate state of electronic excitement Initial experiments were carried out for the reaction by taking 500  $\mu$ L of the drug and adding 50  $\mu$ L of Hydrogen Peroxide 1.0 M concentration and 50  $\mu$ L of reagent. The concentration of Luminol was  $1.0 \times 10^{-3}$  M. After mixing, the chemiluminescence intensity (Relative Light Units (RLU)) of the product was measured. It is necessary to determine the limits of quantification (LOQ) and the limits of detection (LOD; sometimes known as the detection limit, or DL) in the context of purity tests carried out during method validation. The calibration curve, also referred to as the calibration curve procedure in the literature, is frequently the basis for this decision. The formula for the computation is  $DL = 3.3x \sigma / S$ , where  $\sigma$  is the response standard deviation and S is the calibration curve's slope. Several techniques can be used to calculate the standard deviation.

## 3. Results and discussion

For the inaugural development of a novel and expeditious analytical method, the determination of the famotidine drug was undertaken using the chemiluminescence technique (Fig.1). This method relies on the chemical interaction between Luminol and the drug in the presence of hydrogen peroxide solution as a catalyst, resulting in the formation of an electronically excited intermediate state. Initial experiments involved taking 500  $\mu$ L of the drug and supplementing it with 50  $\mu$ L of a 1.0 M concentration of hydrogen peroxide, along with an additional 50  $\mu$ L of a specific reagent. The concentration of Luminol in the mixture was maintained at  $1.0 \times 10^{-3}$  M. Following thorough mixing, the chemiluminescence intensity of the resultant product was then measured.



Fig. 1. Schematic of the chemiluminescence unit

# 3.1. Optimization of the Experimental Conditions

A study was executed for various factors affecting the chemiluminescence intensity of the produced complex to reach the best conditions for the reaction between the drug and the luminol in an alkaline medium and hydrogen peroxide.

# 3.2. Delay last injection measure

An experiment was carried out to determine the optimal time for injecting hydrogen peroxide at a concentration of 1.0 M into the Luminol at a concentration of  $1.0 \times 10^{-3}$  M several times, between

1-35 seconds. The results showed that the optimal time at which the highest luminescence intensity happened was one second, as shown in Figure 2.

### 3.3. Effect of Hydrogen Peroxide Volume

A study was executed to fix the best volume of hydrogen peroxide at a concentration of 1.0 M by injecting increased volumes between 50 and 300  $\mu$ L and keeping other conditions constant. The results explained that the best volume is 250  $\mu$ L, which is dependent on the best volume for sodium hydroxide in subsequent experiments, as shown in Figure 3.



**Fig. 2.** Effect of injection time on the hydrogen peroxide intensity (RLU) of the chemiluminescence



**Fig. 3.** Effect of hydrogen peroxide on the intensity (RLU) of the chemiluminescence

#### 3.4. Effect of the Luminol volume

A study was conducted to know the optimal volume, which gives the highest chemiluminescence intensity, at a reaction of 500  $\mu$ L from the famotidine drug (8.0 mg mL<sup>-1</sup>) concentration and 250  $\mu$ L from 1.0 M hydrogen peroxide and increased volumes from the Luminol solution between 50-300  $\mu$ L 1×10<sup>-3</sup>M. After mixing, the chemical intensity of the solution was measured. The result was that the optimal volume for the reagent that gives the highest luminescence intensity as a reaction product is 100  $\mu$ L, as shown in Figure 4.

#### 3.5. Effect of measurement time

A study was conducted to determine the optimal time, which gives the highest intensity of luminescence when 500  $\mu$ L of Famotidine drug and 250  $\mu$ L of Hydrogen Peroxide 1.0 M, 100  $\mu$ L of Luminol  $1.0 \times 10^{-3}$  M and measuring the chemiluminescence intensity of the solution in different times ranged from 1-35 second. The results showed that the optimal time to give the highest intensity of the chemiluminescence of the reaction product is 20 seconds, as shown in Figure 5.







of the luminescence

#### 3.6. Sequence added

A study was carried out to determine the optimal sequence for the addition of the Luminol reagent and sodium hydroxide, which gives the highest intensity (RLU) of luminescence when 500  $\mu$ L of famotidine with a concentration of 8 mgml<sup>-1</sup>, 250  $\mu$ L of hydrogen peroxide 1.0 M, and 100  $\mu$ L of Luminol of  $1.0 \times 10^{-3}$  M. The results showed that the optimal sequence that gives the highest intensity of the luminescence of the reaction product is the addition of hydrogen peroxide to the luminol reagent. Results were obtained in Table 1.

# 3.7. Procedure and Construction of Calibration Curve

After adjusting the optimal conditions for the reaction by the single variable method, by changing one of the conditions and keeping the other condition constant, the calibration curve was prepared by taking a set of concentrations between 1 - 16 mg mL<sup>-1</sup>, by drawing increasing volumes of 0.1 -1.6 ml of the famotidine drug 100 mg mL<sup>-1</sup> concentration in a 10 ml volumetric flask and then completing the volume to the mark with distilled water. Then 500 µL was taken from each concentration and placed in a test tube and setting equipment to the best volume of the Luminol reagent is 100  $\mu$ L of 1.0  $\times$  10<sup>-3</sup> M and the best volume of 250 µL hydrogen peroxide 1.0 M. The intensity (RLU) of the chemiluminescence was measured at a rate of six readings per concentration. It was found that the concentrations that give Linear ranged 2 -12 mg mL<sup>-1</sup> of famotidine drug solution and that the correlation coefficient value was 0.9969, the straightline equation  $y = 77.7 \times + 998.27$ , the detection limit (LOD) was found to be 0.0314 mg mL<sup>-1</sup>and the limit of quantification (LOQ) was 0.0952 mg mL<sup>-1</sup>, the molecular absorption factor was 2621×104 1 /mol, and the sandal sensitivity was  $1.287 \times 10^{-5}$  mg cm<sup>-2</sup>, as shown in Figure 6.

#### Table 1. Effect of Sequence of Addition

Order of Addition	CL. intensity (RLU)
$H_2O_2 \rightarrow Luminol (1 \rightarrow 2) + Famotidine$	1567
Luminol $\rightarrow$ H <sub>2</sub> O <sub>2</sub> (2 $\rightarrow$ 1) + Famotidine	1215



Fig. 6. Calibration curve of Famotidine

#### 3.8. Accuracy and Precision

After studying and determining optimum conditions for the reaction of the luminescence, to estimate the famotidine drug, conduct six experiments for each measurement process carried out for all concentrations of the calibration curve for the drug and find the rate of readings. Used REC% and relative standard deviation RSD% to express the accuracy of the results, where the results showed that the method had high accuracy and compatibility, as shown in Table 2.

# 3.9. Application of the Method

The method was applied to pharmaceutical compositions, such as 20 mg of famotidine tablets produced by SDI Iraq, where the method was directly applied.

#### 3.10. Direct Method

The direct method was applied to the above pharmaceutical composition with three different concentrations (4, 8, and 10 mg mL<sup>-1</sup>) of the

pharmaceutical product and was treated with the same steps as in the preparation of the calibration curve. The intensity of the luminescence of the above concentrations was measured by six readings per concentration, RE%, and the relative standard deviation of RSD% was calculated as in Table 3.

The chemiluminescence method for famotidine analysis is compared with other analytical methods and estimates, as detailed in Table 4. The table provides insight into the efficiency and operating time of the chemiluminescence method compared to other methods. Also, the organic compounds may be determined by other techniques such as gas chromatography and UV-Vis spectroscopy in water samples[35-38].

### 4. Conclusion

In this work we successfully concentration of famotidine was measured by a novel, and quick, chemiluminescence-based analytical method. It does

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Sample (µg mL <sup>-1</sup> )	CL. intensity (RLU)	Found* (µg mL <sup>-1</sup> )	R(%)	RSD(%)
2.0	1190	$2.076{\pm}~0.08$	103.82	1.965
4.0	1256	$3.976{\pm}~0.17$	99.40	1.178
6.0	1430	$5.799{\pm}\ 0.24$	96.65	0.891
8.0	1560	$8.320{\pm}0.36$	104.00	1.935
10.0	1809	$10.386{\pm}0.44$	103.86	1.987
12.0	1964	$12.295{\pm}0.57$	102.45	1.211

 $<sup>^{*}</sup>$  Mean of three determinations  $\pm$  confidence interval (P = 0.95, n = 8)

R: Recovery

Table 3. Direct Method for determination of famotidine tablets(20 mg)

Drug	Taken (µg mL <sup>-1</sup> )	CL. intensity (RLU)	Found* (µg mL <sup>-1</sup> )	R%	Average of R%	RSD%
Famotidine Tablets	4	1284	$3.921{\pm}0.14$	98.025	98.913	0.098
	8	1605	$7.874{\pm}0.28$	98.425		1.781
	10	1780	$10.029{\pm}~0.44$	100.29		1.209

 $^{*}$  Mean of three determinations  $\pm$  confidence interval (P = 0.95, n = 8)

R: Recovery

Parameters	Electrogenerated chemiluminescence (ECL) [33]	Flow-injection chemiluminescence (FT-CL) [34]	Chemiluminescence (CL)
L aanar ranga	$1.0 \times 10^{-9} - 1.0 \times 10^{-6}$	5.0×10 <sup>-5</sup> - 7.0×10 <sup>-5</sup>	2.0 - 12
	g mL <sup>-1</sup>	g mL <sup>-1</sup>	mg mL <sup>-1</sup>
Electro-oxidation		Rhodanium-6G	Luminol
Slope	4.4	4.5	77.7
Detection limit LOD	$1.0 \times 10^{-10}$	2.035×10-9	0.0314
Detection limit LOD	g mL <sup>-1</sup>	g mL <sup>-1</sup>	mg mL <sup>-1</sup>
Detection quantum LOQ			0.0952
RSD%		2.87	1.981
Correlation coefficient (r)	0.9990	0.990	0.9993

<b>Fable 4.</b> Evaluation of Famotidine Com	paratively Using	Chemiluminescence and	Additional Analy	vtical Techniques.
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not require heating or extraction. The technique showed good, sample and sensitivity, The LOD was found to be  $0.0314 \text{ mg ml}^{-1}$ , and the LOQ was  $0.0952 \text{ mg ml}^{-1}$  with a correlation value of 0.9929. and it was found that the concentrations that give Linear ranged from 2 - 12 mg mL<sup>-1</sup>. The method has been successfully applied to estimate micro concentrations in pharmaceutical preparations containing famotidine.

# **5. References**

- M. S. Islam, M. M. Narurkar, Solubility, stability and ionization behavior of Famotidine, J. Pharm. Pharmacol. 45 (1993) 682-686. https://doi.org/10.1111/j.2042-7158.1993. tb07088.x
- [2] A. Adejare, Remington: The Science and Practice of Pharmacy, Twenty-third Edition, Elsevier Book, 2020. https://doi.org/10.1016/ C2018-0-04991-9
- [3] J.E.F. Reynolds, martindale, the extra pharmacopoeia, 32nd ed., the pharmaceutical press, Massachusetts 810-812, 1999. https:// www.pharmaceuticalpress.com/about/aboutpharmaceutical-press/
- [4] T. O. Al- Khesraji, A. Y. Al-Hayawi, Five new taxa records for the macromycota of Iraq from Suliamaniya and Tikrit provinces (Iraq), Plant Arch., 19 (2019) 4067-4072. https://www. plantarchives.org/
- [5] A. Y. Al-Hayawi, M. H. Sh. Al Jubori, The inhibitory effect of garlic and onion root exudates on escherichia coli from urinary tract infection and molecular detection of hlyA virulence gene, Int. J. Drug Deliv. Technol., 10 (2020) 131-135. https://doi.org/10.25258/ ijddt.10.1.18
- [6] P. Ravisankar, O. S. Sai Koushik, A. A Reddy, U. E Kumar, P. Sai Anvith, P. A. Pragna, Detailed

analysis on acidity and ulcers in esophagus, gastric and duodenal ulcers and management, IOSR J. Dental Med. Sci., 15 (2016) 94-114. https://doi.org/10.9790/0853-1511094114

- M. Vacher, F. I. Galván, B. W. Ding, S. Schramm,
   R. Berraud-Pache, P. Naumov, N. Ferré, Y. J.
   Liu, I. Navizet, D. Roca-Sanjuán, W. J. Baader,
   R. Lindh, Chemi- and bioluminescence of cyclic peroxides, Chem. Rev., 118 (2018) 6927–6974.
   https://doi.org/10.1021/acs.chemrev.7b00649
- [8] K. Gundermann, luminescence, Encyclopedia Britannica, June 29, 2023. https://www. britannica.com/science/luminescence.
- [9] M. A. Tzani, D. K. Gioffsidou, M. G. Kallitsakis, N. V. Pliatsios, N. P. Kalogiouri, P. A. Angaridis, I. N. Lykakis, M. A. Terzidis, Direct and indirect chemiluminescence: reactions, mechanisms and challenges, Molecules, 26 (2021) 7664. https://doi.org/10.3390/molecules26247664
- [10] E. G. Brandão, S. R. W. Perdigão, B. F. Reis, A new flow cell design for chemiluminescence detection using an improved signal transduction network. Determination of hydrogen peroxide in pharmaceuticals, Microchem. J., 171 (2021) 106789. https://doi.org/10.1016/j.microc.2021.106789
- [11] S. N. A. Shah, J-M. Lin, Recent advances in chemiluminescence based on carbonaceous dots, Adv. Colloid Interface Sci., 241 (2017) 24-36. https://doi.org/10.1016/j.cis.2017.01.003
- [12] A. M. Helmenstine, What Is Chemiluminescence? ThoughtCo, New York, NY 10281, 2019. thoughtco.com/ chemiluminescence-definition-4142622.
- [13] P. Gumułka, J. Żandarek, M. Dąbrowska, M. Starek, UPLC technique in pharmacy—An important tool of the modern analyst, Processes, 10 (2022) 2498. https://doi.org/10.3390/ pr10122498

- [14] R. A. Zounr, M. Y. Khuhawar, T. M. J. Khuhawar, M. F. Lanjwani, M. Y. Khuhawar, Analysis of metformin, ranitidine and famotidine from pharmaceuticals and human serum, J. Chromatogr. Sci., 6 (2023) bmad047. https://doi.org/10.1093/chromsci/bmad047
- [15] K. M. Kelani, A. M. Aziz, M. A. Hegazy, L. A. Fattah, Determination of cimetidine, famotidine, and ranitidine hydrochloride in the presence of their sulfoxide derivatives in pure and dosage forms by high-performance thin-layer chromatography and scanning densitometry, J. AOAC Int., 85 (2002) 1015-1020. https://doi.org/10.1093/jaoac/85.5.1015
- [16] G. Z. Tsogas, D. L. Giokas, A. G. Vlessidis, N. P. Evmiridis, The effects of solvent preoxidation on inhibited chemiluminescence of pyrogallol oxidation in flow injection analysis and liquid chromatography, Anal. Chim. Acta, 565 (2006) 56-62. https://doi.org/10.1016/j. aca.2006.02.018
- [17] S. Elbaramawi, M. El-Sadek, M. Baraka, L. Abdel-Aziz, M. Sebaiy, Instrumental analysis of some anti-ulcer drugs in different matrices, Chem. Reports, 2 (2020) 156-172. https://doi. org/10.25082/CR.2020.01.005
- [18] J. Shamar, S. Abbas, Z. Abbas, Analytical methods for determination of ketoprofen drug: A review, Ibn AL-Haitham J. Pure Appl. Sci., 35 (2022) 76–82. https://doi. org/10.30526/35.3.2842
- [19] E. F. Silva, P. R. B. Gomes, R. N. Fernandes, W. S. Lyra, Flow-injection spectrometric determination of sodium diclofenac in pharmaceutical formulations, J. Chil. Chem. Soc., 63 (2018) 3941-3946. http://dx.doi. org/10.4067/s0717-97072018000203941
- [20] R. P. Adhikari, S. N. Rahman, S. Lamichane, A. Bora, Overview of analytical methods for the determination of H2 receptor blockers: A review, J. Drug Deliv. Ther., 13 (2023) 137-144. http://dx.doi.org/10.22270/jddt.v13i10.6245
- [21] O. Z. Devi, K. Basavaiah, P. J. Ramesh, K. B. Vinay, Simple and sensitive UV spectrophotometric methods for determination of famotidine in tablet formulations, Farmacia, 59 (2011) 647-658. https://farmaciajournal. com/arhiva/20115/issue52011art05.html
- [22] M. Alamgir, M. Y. Khuhawar, S. Q. Memon, A. Hayat, R. A. Zounr, A. Chanar, HPLC determination of metformin, famotidine, and ranitidine by derivatization with benzoin from drugs and biological samples, Pharm.

Anal. Acta, 8 (2017) 1-7 http://dx.doi. org/10.4172/2153-2435.1000546

- [23] A. M. K. Ahmed, A. I. Khaleel, S. T. Amine, Determination of ranitidine - HCl in pharmaceutical formulations by kinetic spectrophotometric and flow injection activated chemiluminescence methods, Iraqi Nat. J. Chem., 24 (2006) 534-550. https:// injchemistry.uobabylon.edu.iq/index.php/ chem/article/view/633/580
- [24] T. Pérez-Ruiz, C. Martinez-Lozano, V. Tomás, E. Bravo, Direct determination of ranitidine and famotidine by CE in serum, urine, and pharmaceutical formulations, J. Pharm. Biomed. Anal., 30 (2002) 1055-1061. https:// doi.org/10.1016/S0731-7085(02)00444-2
- [25] Z. Fu, W. Huang, G. Li, Y. Hu, A chemiluminescence reagent-free method for the determination of captopril in medicine and urine samples by using trivalent silver, J. Pharm. Anal., 7 (2017) 252-257. https://doi. org/10.1016/j.jpha.2017.05.005
- [26] C. Lau, X. Qin, J. Liang, J. Lu, Determination of cysteine in a pharmaceutical formulation by flow injection analysis with a chemiluminescence detector, Anal. Chim. Acta, 514 (2004) 45-49. https://doi.org/10.1016/j.aca.2004.01.007
- [27] L. Ma, W. J. Kang, X. D. Xu, L. M. Niu, H. M. Shi, S. Li, Flow-injection chemiluminescence determination of penicillin antibiotics in drugs and human urine using luminol Ag(III) complex system, J. Anal. Chem., 67 (2012) 219-225. https://doi.org/10.1134/S1061934812030070
- [28] Y. Huang, Z. Zhang, Binding study of a drug with bovine serum album using a combined technique of microdialysis with flow-injection chemiluminescent detection, J. Pharm. Biomed. Anal., 35 (2004) 1293-1299. https:// doi.org/10.1016/j.jpba.2004.04.003
- [29] X. Xiong, X. Zhao, Y. Li, Z. Song, A fast and efficient chemiluminescence method for determination and pharmacokinetic study of paclitaxel in rat plasma, Curr. Pharm. Anal., 10 (2014) 246-254. https://doi.org/10.2174/15734 12910666140716164950
- [30] J. A. Ocaña-González, M. Ramos-Payán, R. Fernández-Torres, M. V. Navarro, M. Á. Bello-López, Application of chemiluminescence in the analysis of wastewaters—A review, Talanta, 122 (2014) 214–222. https://doi.org/10.1016/j. talanta.2014.01.028
- [31] B. Khadro, B. D. Leca-Bouvier, F. Lagarde, F. Barbier, L. J. Blum, C. Martelet, L.

Marcotte, M. Tabrizian, N. Jaffrezic-Renault, Chemiluminescence of luminol for the determination of cobalt(II) adsorbed on a chitosan membrane, Sensor Lett., 7 (2009) 833-838. https://doi.org/10.1166/sl.2009.1158

- [32] Y.Su,Y.N.Xie,X.D.Hou,Y.Lv,Recentadvances in analytical applications of nanomaterials in liquid-phase chemiluminescence, Appl. Spectrosc. Rev., 49 (2014) 201-232. https://doi. org/10.1080/05704928.2013.819514
- [33] Z. Chang, X. Zheng, Highly sensitive electrogenerated chemiluminescence (ECL) method for famotidine with preanodizing technique to improve ECL reaction microenvironment at graphite electrode surface, J. Electroanal. Chem., 587 (2006) 161–168. https://doi.org/10.1016/j.jelechem.2005.11.006
- [34] Y-H. Tang, N-N. Wang, X-Y. Xiong, F-M. Xiong, S-J. Sun, A new sensitive flowinjection chemiluminescence method for the determination of H<sub>2</sub>-receptor antagonists, Luminescence, 22 (2007) 343–348. https://doi. org/ 10.1002/bio.969
- [35] S. Teimoori, An immobilization of aminopropyl trimethoxysilane-phenanthrene carbaldehyde on graphene oxide for toluene extraction and separation in water samples, Chemosphere, 316 (2023) 137800. https://doi.org/10.1016/j. chemosphere.2023.137800
- [36] M Arjomandi, A review: analytical methods for heavy metals determination in environment and human samples, Anal. Methods Environ. Chem. J., 2 (2019) 97-126. https://doi.org/10.24200/ amecj.v2.i03.73
- [37] S. Teimoori, A.H. Hassani, M. Panahi, N. Mansouri, Rapid extraction of BTEX in water and milk samples based on functionalized multi-walled carbon nanotubes by dispersive homogenized-micro-solid phase extraction, Food Chem., 421 (2023) 136229. https://doi. org/10.1016/j.foodchem.2023.136229
- [38] S. Teimoori, A.H. Hassani, New extraction of toluene from water samples based on nanocarbon structure before determination by gas chromatography, Int. J. Environ. Sci. Technol., 20 (2023) 6589–6608. https://doi.org/10.1007/ s13762-023-04906-9



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Determination and evaluation of trace elements in the blood of radiography workers using graphite furnace atomic absorption spectrometry

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#### ABSTRACT

This study aimed to evaluate the potentially toxic effects of trace elements in the blood of Iraqi medical radiography workers by analyzing them through the graphite furnace-atomic absorption spectrometry method (GF-AAS). The study involved 50 male blood radiography workers from the medical imaging field at Al-Shatrah General Hospital in Thi-Qar City, Iraq. All workers were aged between 35-50 years and had worked for less than 10 years. The study compared these workers with 50 healthy men. The study found a significant increase in the levels of Cu, Pb, Cd, and Ca among radiography workers compared to the healthy control group, while Zn and Se levels decreased significantly. Moreover, Specificity and confidence interval (95%) were estimated via the receiver operating characteristic curve (ROC). The study provided conclusive evidence of disturbances in the levels of trace elements in the blood of radiographer workers, which makes them more susceptible to many diseases because of their radiation exposure. which portends the use of more preventive measures and commitment to the principles of radiation protective protocols to reduce the effects of radiation exposure and an increase in the occupational dose. The linear range of Cd, Cu, Zn, Se and Pb in human serum were obtained 0.2-6.0 µg dL<sup>-1</sup>, 6.0-200 μg dL<sup>-1</sup>, 8.0-200 μg dL<sup>-1</sup>, 10-250 μg dL<sup>-1</sup>, 4-120 μg dL<sup>-1</sup> by GF-AAS after dilution samples with DW up to 20 (n=10, RSD< 5%)

# 1. Introduction

The utilization of medical imaging procedures like X-rays, CT scans, and MRI, which have quickly advanced over the last three decades, has become standard practice in healthcare facilities. The main advantage of such technologies is the structural information they provide about the human organism, which helps with the identification of illnesses, internal body tissue investigation, evaluation, and medical treatment. As a result, these technologies are crucial to both the safety and quality of healthcare for patients

\*Corresponding Author: Ahmed Jaber Ibrahim Email: ahmed.jibrahim@alayen.edu.iq https://doi.org/10.24200/amecj.v6.i04.321 [1]. Radiographers' radiation exposure risk is a danger associated with all medical imaging modalities, but it varies greatly depending on the characteristics of the person who is the radiographer. Radiographers may be more susceptible to cancer or have higher quantities of trace elements in their blood, which could lead to damage to their DNA [2, 3]. There are many ways that DNA can change, but one of them involves reacting ROS that break DNA strands and crosslink DNA proteins. According to recent studies, radiation may indirectly alter DNA through the accumulation of trace elements and minerals [4, 5]. Throughout the human body, trace elements are present in minute amounts and support the functions of the body. Trace elements are categorized into essential and non-essential elements
based on the demands of the organism [6]. Essential elements, which make up 0.02% of body weight, are crucial in physiological functions like metabolic processes, blood cell production, reproduction, and immunity. The occurrence of trace elements in quantities that are either too low or too excessive can lead to metabolic problems and disruptions of cellular growth, including the occurrence of cancer and mutagenesis [7]. Because they can disrupt cellular processes and change the expression patterns of numerous genes, trace elements are hazardous because they increase the likelihood of developing disease [8]. Trace elements can interact with DNA in a variety of ways. Large numbers of strongly bonded metal ions can be found in nucleic acids. The incorporation of genetic mistakes may increase due to changes in DNA polymerase activity caused by the binding of particular metals. Trace elements interfere with the function of reparative proteins, altering the outcome of the repair of double-strand breaks (DSBs), and also changing the distribution of altered DNA, potentially increasing the risk of developing any type of cancer. ROS generation from exposures to trace elements can result in damage to DNA [9]. Human radiation exposure has an impact on trace element levels, as long-term exposure leads to changes in the physiological functions of the organism's body [10]. The normal range of elements such as Cd (  $>0.3 \ \mu g \ dL^{-1}$ ), Cu(130-160  $\ \mu g \ dL^{-1}$ ), Zn (120-170 µg dL<sup>-1</sup>), Se (15-25 µg dL<sup>-1</sup>), Pb(5-25 µg dL<sup>-1</sup>) <sup>1</sup>), and Ca (85 to 100 mg L<sup>-1</sup>) were reported in human serum samples [11]. Spectroscopy techniques have been developed in many areas of analytical chemistry, such as the environmental, medical, and industrial fields [12]. The purpose of this investigation was to evaluate the potentially toxic effects of radiation on the levels of trace elements in the blood of Iraqi medical radiography workers by estimating them through the atomic absorption spectrometry method

#### 2. Materials and methods

#### 2.1. Instruments

A centrifuge (Biofuge B, Heraeus–Christ, Germany) was used to separate blood samples, and an oil bath (F3– HAKE, Germany) to heat samples. Copper (Cu), lead (Pb), calcium (Ca), cadmium (Cd), zinc (Zn), and selenium (Se) were determined by Graphite furnace atomic absorption Spectrophotometer (280Z AA/Agilent, USA).

#### 2.2. Chemicals

Each chemical has been sourced to the highest possible level of purity and from discreet origins, including hydrochloric acid (HCl 37%, CAS Number:7647-01-0, Merck, Germany), nitric acid (HNO<sub>3</sub> 69%, CAS Number:7697-37-2, Scharlau-Espane), perchloric acid (HClO<sub>4</sub> 70%, CAS Number: 7601-90-3, HIMEDIA, India), and sulfuric acid (H<sub>2</sub>SO<sub>4</sub> 98%, CAS Number: 7664-93-9, HIMEDIA, India). Also, Standard solutions of copper, lead, cadmium, calcium, zinc, and selenium (1000  $\pm$  51 mg L<sup>-1</sup> in 2 % HNO<sub>3</sub>) were prepared from Merck (Germany).

#### 2.3. Subjects

The current study included 50 male radiography workers working in the field of medical imaging (MRI, CT-scan, and X-rays departments) at Al-Shatrah General Hospital in Thi-Qar City Iraq, with an age range of 35-50 years and 10 years of periods of working. The subjects compared with 50 males who did not work in the field of medical imaging, with the same age range of 35-50 years, and good health served as a control group. Participants suffering from chronic diseases such as heart disease, hypertension, thyroid disorders, and diabetes were excluded. Also, The Body Mass Index was determined via the Equation 1 [13]:

#### BMI (kg m<sup>-2</sup>) = Weight (kg) / [Height(m)]<sup>2</sup> (Eq.1)

#### 2.4. Blood Samples Collection

Blood samples were obtained from radiography workers and health control groups at AL-Shatrah General Hospital. Around 5.0 mL of blood was haggard from a vein and allowable to clot for about 15 min at  $\degree \circ \degree C$  and then centrifuged at  $\degree \cdot \cdot \cdot rpm$  for  $\degree \cdot$ min. The obtained serum was stored in a deep freeze at  $- \degree \wedge \degree \degree C$  and was ready for use in the measurement of trace elements.

Everyone who participated in the study gave their consent, and it was made public. The 1964 Declaration of Helsinki and its later amendments or comparable ethical norms were followed in conducting this study, which was approved by the Al-Ayen University Ethical Committee) (Date: 5 Jan 2024, AUEC, Number ECN:2024- 892)

#### **2.5.** Determination of Trace Elements

#### 2.5.1. Preparation of Standard Solutions

Standard solutions of copper, lead, cadmium, calcium, zinc, and selenium (1000  $\pm$  51 mg L<sup>-1</sup> in 2 % HNO) were obtained from Merck company. required working solutions were ready by diluting a specific stock solution with aquatic to obtain a standard calibration curve.

#### 2.5.2.Samples Preparation and procedure

Serum samples were digested by incorporating 2.0 mL of concentrated HNO<sub>2</sub> and 1.0 mL of concentrated HClO<sub>4</sub> to 0.5 mL of serum in a Pyrex tube heated in an oil bath for 1 hour at 165 °C. So that the digested samples were clear, the tubes were cooled at <sup>Yo</sup> °C, and the quantity was increased to  $\cdot$  mL by 0.3 M HCl. Graphite furnace-atomic absorption spectrometry (GF-AAS) was utilized to analyze of copper, lead, cadmium, calcium, zinc, and selenium in standards and sample solutions under the ideal conditions shown in Table 1. Also, the elements in serum samples of medical radiography workers was determined by the GF-AAS which was shown in Table 2. The proposed method was validated by certified reference material which was analyzed by ICP-MS in serum samples (Table 3).

intended by Equation 2. The limit of quantification (LOQ) for trace elements was calculated by Equation 3. Where is the standard deviation of blank solution for 10 reads and S is the slope of the standard curve.

$$LOD=3.3 SD/S$$
 (Eq.2)

LOQ = 10SD/S (Eq.3)

#### 2.6. Statistical Examining

The data were assessed using the statistical program SPSS, version 23.0. A Student's t-test was used to determine the significance of any differences that were found to exist between each group. The mean  $\pm$  standard deviation (SD) of the data was displayed. The operating characteristics (ROC) curve was used to calculate specificity, specificity, and the 95% confidence interval. All p-values were two-tailed and p<0.05 was deemed significant for statistical analysis.

The limit of detection (LOD) for trace elements was

 Table 1. Ideal circumstances for the experiment were employed

 to determine Cu, Pb, Cd, Ca, Zn, and Se by GF-AAS with Zeeman background correction.

	, , , , ,	5		8		
Parameters	Cu	Pb	Ca	Cd	Zn	Se
Lamp Current (mA)	5	10	10	4	5	5
Wavelength (nm)	234.8	283.3	422.4	228.8	217	196
Slit Width (nm)	1	1	1	1	1	1
Ac FR(dm <sup>3</sup> min <sup>-1</sup> )	0.8 -1	0.8-1	0.8-1	0.8-1	0.8-1	0.8-1
Read Time (s)	3	3	3	3	3	3

Ac FR: Acetylene flow rate

**Table 2.** Statistical results for determination elements in serum workers by GF-AAS ( $\mu g dL^{-1}$ )

GF-AAS	Cd	Cu	Zn	Se	Pb
LR	0.01-0.3	0.3-10.0	0.4-12.0	0.5-12.5	0.2-6.0
LOD	0.003	0.084	0.12	0.14	0.066
LOQ	0.011	0.302	0.40	0.50	0.201
*LR Method	0.2-6.0	6.0-200	8.0-200	10.0-250.0	4.0-120
LOD	0.06	2.02	2.60	3.30	1.33
LOQ	0.20	6.03	8.00	10.0	4.00
Mean RSD%	2.20	2.01	1.33	1.34	3.42

\*Procedure: Dilution Factor=20 (0.5 mL of Serum+10 mL HCL/DW)

LR: Linear Range, Concentration of  $\mu g dL^{-1} = 10 \times \mu g L^{-1}$ 

<b>Table 3.</b> Validation	procedure by certified	l reference material b	v ICP-MS in serum	samples ( $\mu g dL^{-1}$ )
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Elements	Certified Value (ICP-MS)	Found (GF-AAS)	Recovery (%)
Cu	83.28±1.52	81.28±3.39	97.6
Pb	20.96±0.77	19.84±0.98	94.7
Cd	0.902±0.02	$0.817{\pm}0.04$	95.1
Ca	9.13±0.35	8.98±1.44	98.4
Zn	79.26±2.22	76.44±3.75	96.4
Se	148.93±31.16	138.77±30.96	٩٣.1

#### 3. Results and Discussion

The descriptive data used in the current study indicates as in Table 4, that there were no significant variances in the mean of age and BMI between the study groups  $(37.93\pm4.44$  years, and  $33.87\pm1.55$ , respectively compared to  $38.01\pm4.68$  years, and  $34.22\pm2.76$ , respectively). and this is due to the selection of the ages and BMI of the control group close to the ages and BMI of the radiography workers being studied so that there was no discrepancy between the ages and BMI of the studied subjects. Therefore, nonsignificant variances (p>0.05) were in the age and BMI of the studied groups [14].

The results of this investigation displayed a high significant increase (p<0.01) in the levels of serum copper (Cu) in the blood of radiography workers ( $81.28\pm16.39 \ \mu g \ dL^{-1}$ ) compared to the control group ( $60.1\pm15.21 \ \mu g \ dL^{-1}$ ) as in Figure 1. It also showed the acquired AUC data that serum copper (Cu) might be a more precise predictive biomarkers in radiography workers (AUC = 0.819) as demonstrated in Figure 2.

Table 4. The general descriptive data for every individual taking part in this research

The traits	Radiography workers group	Control group	p-value
Number (N)	50	50	
Age± SD (year)	37.93±4.44	38.01±4.68	0.183
BMI± SD (kg m <sup>-2</sup> )	33.87±1.55	34.22±2.76	0.114

BMI: body mass index; SD: Standard devation; P>0.05: non-significant variance; P<0.05 significant variance; P<0.01: high significant variance



Fig.1. Serum levels of Copper (Cu) in the radiography workers group and the control group



Fig.2. The ROC curve for levels of serum Copper (Cu) in the radiography workers group and the control group

Regarding of serum lead (Pb), the results of this investigation displayed a high significant increase (P< 0.01) in the blood of radiography workers (19.84±6.8  $\mu$ g dL<sup>-1</sup>) compared to the control group (10.14±3.44  $\mu$ g dL<sup>-1</sup>) as in Figure 3. It also showed the acquired AUC data that serum copper (Cu) might be a more precise predictive biomarkers in radiography workers (AUC = 0. 891) as demonstrated in Figure 4.



Fig.3. Serum levels of Lead (Pb) in the radiography workers group and the control group



Fig.4. The ROC curve for levels of serum Lead (Pb) in

the radiography workers group and the control group Regarding serum cadmium (Cd), the results of this investigation displayed a highly significant increase (P<0.01) in the blood of radiography workers ( $0.81\pm0.18 \ \mu g \ dL^{-1}$ ) compared to the control group ( $0.42\pm0.07 \ \mu g \ dL^{-1}$ ) as in Figure 5. It also showed the acquired AUC data that serum copper (Cu) might be a more precise predictive biomarker in radiography workers (AUC= 0.990) as demonstrated in Figure 6.



**Fig.5.** Serum levels of Cadmium (Cd) in the radiography workers group and the control group



Fig.6. The ROC curve for levels of serum Cadmium (Cd) in the radiography workers group and the control group

Regarding of serum Calcium (Ca), the results of this investigation displayed a high significant increase (p < 0.01) in the blood of radiography workers ( $8.98\pm1.95 \mu g dL^{-1}$ ) compared to the control group ( $6.18\pm1.66 \mu g dL^{-1}$ ) as in Figure 7. It also showed the acquired AUC data that serum copper (Cu) might be a more precise predictive biomarkers in radiography workers (AUC = 0. 862) as demonstrated in Figure 8.



Fig.7. Serum levels of Calcium (Ca) in the radiography workers group and the control group



**Fig.8.** The ROC curve for levels of serum Calcium (Ca) in the radiography workers group and the control group

Regarding of serum Zinc (Zn), the results of this investigation displayed a high significant increase (p<0.01) in the blood of radiography workers

 $(76.44\pm10.22 \ \mu g \ dL^{-1})$  compared to the control group  $(96.31\pm12.88 \ \mu g \ dL^{-1})$  as in Figure 9. It also showed the acquired AUC data that serum copper (Cu) could not be a predictive biomarker in radiography workers (AUC = 0. 118) as demonstrated in Figure 10.



**Fig.9.** Serum levels of Zinc (Zn) in the radiography workers group and the control group



**Fig.10.** The ROC curve for levels of serum Zinc (Zn) in the radiography workers group and the control group

Finally, The results of this study showed a significant decrease (p < 0.01) in the levels of serum Selenium (Se) in the blood of radiography workers ( $131.77\pm30.96$  ng

dL<sup>-1</sup>) compared to the control group  $(154.37\pm$ °3.85 ng dL<sup>-1</sup>) as in Figure 11. It also showed the acquired AUC data that serum Selenium (Se) could not be a predictive biomarker in radiography workers (AUC= 0.322) as demonstrated in Figure 12.



**Fig.11.** Serum levels of Selenium (Se) in the radiography workers group and the control group



Fig.12. The ROC curve for levels of serum Selenium (Se) in the radiography workers group and the control group

Medical radiography workers run a greater risk of poisoning since they are frequently exposed to electromagnetic radiation. Iraqi medical radiography

workers exposure to radiation is evaluated for possible hazardous effects using several parameters including measurements of trace element levels. Trace elements are necessary for many different biological activities. On the other hand, these elements can cause metabolic problems and disturb cellular growth, leading to the emergence of genetic abnormalities and cancer, if their levels are too high or low [15, 16]. Many risk factors, including smoking and hepatic, renal, cardiovascular, and DM illnesses, can raise the body's levels of elements. A diagnostic for prolonged exposure, an increase in blood trace element values revealed the body's stress [17, 18]. The best way to determine the exposure to hazardous elements (like Lead) that cling tightly to intracellular proteins is to analyze serum [19, 20]. It has been shown that radiation exposure raises Cd and Pb levels, which in turn overproduces Reactive oxygen species (ROS) and damages DNA in male sperm cells [21]. Moreover, working-related Cd exposure impairs lung function or may result in COPD (chronic obstructive pulmonary disease) [22]. Additionally, studies show that high plasma Pb concentrations harmed the reproductive system of men, leading to sperm quality declines, morphological changes, and broken-down DNA [23, 24]. Furthermore, it has been proposed that elevated blood levels of copper and iron may elevate the likelihood of developing Alzheimer's illness [25, 26].

Radiation exposure may cause problems with lipid membranes, chromosomal integrity, enzyme activities, and cell-to-cell communication. These problems can then affect tissue respiration, proliferation, and hemopoiesis [27, 28]. Previous studies have also proven that exposure to radiation for a long period leads to the appearance of problems in the hair follicles due to damage to the keratin protein, which leads to the appearance of structural symptoms in the hair [29, 30]. Other layers of cells are then affected by the low molecular mass for degraded protein fragments that migrate outside the skin cells, leading to skin-related problems when exposed to low amounts of radiation [31, 32]. The results of this study offer compelling evidence that for a long time, radiation exposure puts radiologists at elevated risk for specific disorders, each of which has a unique effect on trace element quantities. To fully understand the relationship between long-time radiation exposure and the amounts of trace elements in human beings, more research is necessary [33]. Professional radiographers should implement safety precautions and preventive measures, such as wearing protective clothing and shielding, using beam-limiting devices, and using appropriate beam filtering, based on this study's results. In addition, basic guidelines regarding distance, duration, and shields must be followed to reduce radiation exposure in a work environment [34, 35]. Also, some methodology was used for the determination of trace elements by ET-AAS or AAS [36-39]. Additionally, radiographers who have been exposed to prolonged radiation need to have frequent blood element testing performed to avoid or diagnose illnesses [40].

#### 4. Conclusion

Spectroscopy techniques have been developed in many areas of analytical chemistry, such as the environmental, medical, and industrial fields. This study aimed to evaluate the potentially toxic effects of radiation on the levels of trace elements in the blood of Iraqi medical radiographer workers by estimating them through the atomic absorption spectroscopy method (RSD< 5%), which has proven its efficiency in the measurement and estimation process. LOD of Cd, Cu, Zn, Se, and Pb were obtained at 0.06, 2.02, 2.60, 3.30, and 1.33, respectively (n=10). The study provided conclusive evidence of disturbances in the levels of trace elements in the blood radiographer workers in the medical radiation field, which makes them more susceptible to many diseases because of their radiation exposure. which portends the use of more preventive measures and commitment to the principles of radiation protective protocols to reduce the effects of radiation exposure and an increase in the occupational dose.

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#### 6. References

- [1] E. Nabrawi, A.T. Alanazi, E. Al Alkhaibari, Imaging in healthcare: A glance at the present and a Glimpse into the future, Cureus, 15 (2023) e36111. https://doi.org/10.7759/cureus.36111
- [2] M.L. Kwan, D.L. Miglioretti, E.J. Bowles, S. Weinmann, R.T. Greenlee, N.K. Stout, A.K. Rahm, S.A. Alber, P. Pequeno, L.M. Moy, Quantifying cancer risk from exposures to medical imaging in the risk of pediatric and adolescent cancer associated with medical imaging (RIC) study: research methods and cohort profile, Canc. Causes Contr., 33 (2022) 711-726. https://doi.org/10.1007/s10552-022-01556-z
- [3] S.I.S. Al-Hawary, E.A.M. Saleh, N.A. Mamajanov, G.N. Sergeevna, H.O. Alsaab,

A. Alghamdi, S.A. Ansari, A.H.R. Alawady, A.H. Alsaalamy, A.J. Ibrahim, Breast cancer vaccines; A comprehensive and updated review, Pathol. Res. Pract., 249 (2023) 154735. https:// doi.org/10.1016/j.prp.2023.154735

- [4] H. Moeini, M. Mokari, DNA damage and microdosimetry for carbon ions: track structure simulations as the key to quantitative modelling of radiation-induced damage, Med. Phys., 49 (2022) 4823-4836. https://doi.org/10.1002/ mp.15711
- [5] A.K. Aranda-Rivera, A. Cruz-Gregorio, Y.L. Arancibia-Hernández, E.Y. Hernández-Cruz, J. Pedraza-Chaverri, RONS and oxidative stress: An overview of basic concepts, Oxygen, 2 (2022) 437-478. https://doi.org/10.3390/ oxygen2040030
- [6] N. Esmaeili, Rapid speciation of lead in human blood and urine samples based on mwcnts@ dmp by dispersive ionic liquid-suspensionmicro-solid phase extraction, Biol. Trace Elem. Res., 199 (2021) 2496-2507. https://doi. org/10.1007/s12011-020-02382-7
- [7] M.R. Islam, S. Akash, M.H. Jony, M.N. Alam, F.T. Nowrin, M.M. Rahman, A. Rauf, M. Thiruvengadam, Exploring the potential function of trace elements in human health: a therapeutic perspective, Mol. Cell Biochem., 478 (2023) 2141–2171. https://doi.org/10.1007/ s11010-022-04638-3
- [8] A. Monga, A.B. Fulke, D. Dasgupta, Recent developments in essentiality of trivalent chromium and toxicity of hexavalent chromium: implications on human health and remediation strategies, J. Haz. Mat. Adv., 7 (2022) 100113. https://doi.org/10.1016/j.hazadv.2022.100113
- [9] S. Maksoud, The DNA double-strand break repair in glioma: Molecular players and therapeutic strategies, Mol. Neurobiol., 59 (2022) 5326-5365. https://doi.org/10.1007/ s12035-022-02915-2
- [10] L. Bradney, H. Wijesekara, K.N. Palansooriya, N. Obadamudalige, N.S. Bolan, Y.S. Ok, J. Rinklebe, K.-H. Kim, M. Kirkham, Particulate plastics as a vector for toxic trace-element uptake by aquatic and terrestrial organisms and human health risk, Environ. Int., 131 (2019) 104937. https://doi.org/10.1016/j.envint.2019.104937
- [11] J.S. Tratnik, D. Mazej, M. Horvat, Analytical quality requirements in human biomonitoring programs: trace elements in human blood, Int. J. Environ. Res. Public Health, 16 (2019) 2287. https://doi.org/10.3390/ijerph16132287

- [12] A. J. Ibrahim, Adsorption behavior of crystal violet dye in aqueous solution using Co<sup>+2</sup> hectorite composite as adsorbent surface, Anal. Meth. Environ. Chem. J., 6 (2023) 5-16. https:// doi.org/10.24200/amecj.v6.i01.219.
- [13] A. j. Ibrahim, H.A.W. Dwesh, R.A. Shahid, Evaluation of serum leptin levels in hypertensive men in Thi Qar City-Iraq (a comparative study), J. Pop. Therapeut. Clin. Pharmacol., 30 (2023) 54-62. https://doi.org/10.47750/ jptcp.2023.30.03.007
- [14] A.j. Ibrahim, A.H. AL–Saeed, Evaluation of oxidative status, potassium, magnesium, and lipid profile in serum of patients with  $\beta$ -thalassemia major, Thi-Qar, Iraq, Maaen J. Med. Sci., 2 (2023) 108-115. https://doi. org/10.55810/2789-9128.1029
- [15] R.M. El-Gharabawy, M.A. Al-Dubayan, M.S. Alsharidah, A.A. Al-Hadyab, S.A. Alsuhaibani, Potential toxic effects triggered by radiation exposure among medical radiographers through an imbalance in trace elements and redox status, Poll Res., 39 (2020) 531-541. http://www. envirobiotechjournals.com/PR/vol39i32020/ Poll%20Res-4.pdf
- [16] A.G. Godswill, I.V. Somtochukwu, A.O. Ikechukwu, E.C. Kate, Health benefits of micronutrients (vitamins and minerals) and their associated deficiency diseases: A systematic review, Int. J. Food Sci., 3 (2020) 1-32. https:// doi.org/10.47604/ijf.1024
- [17] S. Saedi, S.E. Watson, J.L. Young, Y. Tan, K.A. Wintergerst, L. Cai, Does maternal low-dose cadmium exposure increase the risk of offspring to develop metabolic syndrome and/or type 2 diabetes, Life Sci., 315 (2023) 121385. https:// doi.org/10.1016/j.lfs.2023.121385
- [18] A. Mirończuk, K. Kapica-Topczewska, K. Socha, J. Soroczyńska, J. Jamiołkowski, M. Chorąży, A. Czarnowska, A. Mitrosz, A. Kułakowska, J. Kochanowicz, Disturbed ratios between essential and toxic trace elements as potential biomarkers of acute ischemic stroke, Nutrients, 15 (2023) 1434. https://doi. org/10.3390/nu15061434
- [19] S. Satarug, G.C. Gobe, D.A. Vesey, K.R. Phelps, Cadmium and lead exposure, nephrotoxicity, and mortality, Toxics, 8 (2020) 86. https://doi. org/10.3390/toxics8040086
- [20] S. Proença, B.I. Escher, F.C. Fischer, C. Fisher, S. Grégoire, N.J. Hewitt, B. Nicol, A. Paini, N.I. Kramer, Effective exposure of chemicals in in vitro cell systems: A review of

chemical distribution models, Toxicol. Vitro, 73 (2021) 105133. https://doi.org/10.1016/j. tiv.2021.105133

- [21] A.C. Nsonwu-Anyanwu, E.R. Ekong, S.J. Offor, O.F. Awusha, O.C. Orji, E.I. Umoh, J.A. Owhorji, F.R. Emetonjor, C.A.O. Usoro, Heavy metals, biomarkers of oxidative stress and changes in sperm function: A case-control study, Int. J. Reprod. Biomed., 17 (2019) 163-174. https://doi.org/10.18502%2Fijrm. v17i3.4515
- [22] D.L. Knoell, T.A. Wyatt, The adverse impact of cadmium on immune function and lung host defense, Semin. Cell Biol., 115 (2021) 70-76. https://doi.org/10.1016/j.semcdb.2020.10.007
- [23] I. Chabchoub, M.A. Nouioui, M. Araoud, M. Mabrouk, D. Amira, M.H. Ben Aribia, K. Mahmoud, F. Zhioua, G. Merdassi, A. Hedhili, Effects of lead, cadmium, copper and zinc levels on the male reproductive function, Andrologia, 53 (2021) e14181. https://doi.org/10.1111/ and.14181
- [24] A. Agarwal, S. Baskaran, M.K.P. Selvam, R. Finelli, C. Barbarosie, K.A. Robert, C. Iovine, K. Master, R. Henkel, Scientific landscape of oxidative stress in male reproductive research: A scientometric study, Free Radic. Biol. Med., 156 (2020) 36-44. https://doi.org/10.1016/j. freeradbiomed.2020.05.008
- [25] M.A. Sahraian, Blood concentrations of cadmium and lead in multiple sclerosis patients from Iran, Iran. J. pharm. Res.: 15 (2016) 825. https://www.ncbi.nlm.nih.gov/pmc/articles/ PMC5316261
- [26] R. Squitti, P. Faller, C. Hureau, A. Granzotto, A.R. White, K.P. Kepp, Copper imbalance in Alzheimer's disease and its link with the amyloid hypothesis: Towards a combined clinical, chemical, and genetic etiology, J. Alzheim. Dis., 83 (2021) 23-41. https://doi. org/10.3233/JAD-201556
- [27] M. Karabulutoglu, R. Finnon, T. Imaoka, A.A. Friedl, C. Badie, Influence of diet and metabolism on hematopoietic stem cells and leukemia development following ionizing radiation exposure, Int. J. Radiat. Biol., 95 (2019) 452-479. https://doi.org/10.1080/09553 002.2018.1490042
- [28] W. Meng, J.D. Palmer, M. Siedow, S.J. Haque, A. Chakravarti, Overcoming radiation resistance in gliomas by targeting metabolism and DNA repair pathways, Int. J. Mol. Sci., 23 (2022) 2246. https://doi.org/10.3390/

ijms23042246

- [29] C.F. Cruz, C. Costa, A.C. Gomes, T. Matamá, A. Cavaco-Paulo, Human hair and the impact of cosmetic procedures: A review on cleansing and shape-modulating cosmetics, Cosmetics, 3 (2016) 26. https://doi.org/10.3390/ cosmetics3030026
- [30] M. Zawrzykraj, M. Deptuła, K. Kondej, A. Tymińska, M. Pikuła, The effect of chemotherapy and radiotherapy on stem cells and wound healing. Current perspectives and challenges for cell-based therapies, Biomed. Pharmacother., 168 (2023) 115781. https://doi. org/10.1016/j.biopha.2023.115781
- [31] Z. Wang, L. Scheres, H. Xia, H. Zuilhof, Developments and challenges in self-healing antifouling materials, Adv. functional Mat., 30 (2020) 1908098. https://doi.org/10.1002/ adfm.201908098
- [32] I. Sadowska-Bartosz, G. Bartosz, Antioxidant defense of Deinococcus radiodurans: How does it contribute to extreme radiation resistance. Int. J. Radiat. Biol., 99 (2023) 1803-1829. https:// doi.org/10.1080/09553002.2023.2241895
- [33] J. Boice Jr, L.T. Dauer, K.R. Kase, F.A. Mettler Jr, R.J. Vetter, Evolution of radiation protection for medical workers, British J. Radio., 93 (2020) 20200282, https://doi.org/10.1259/ bjr.20200282
- [34] O. Lakhwani, V. Dalal, M. Jindal, A. Nagala, Radiation protection and standardization, J. Clin. Orthop. Trauma, 10 (2019) 738-743. https://doi.org/10.1016/j.jcot.2018.08.010.
- [35] S.M. Ridzwan, L. Fritschis, N. Bhoo-Pathyi, Radiation safety and radiation monitoring practices among medical radiation workers in Malaysia, Int. J. Radiat. Res., 21 (2023) 459-468. https://doi.org/10.52547/ijrr.21.3.15
- [36] M. Arjomandi, A review: analytical methods for heavy metals determination in environment and human samples, Anal. Methods Environ. Chem. J., 2 (2019) 97-126. https://doi.org/10.24200/ amecj.v2.i03.73
- [37] N. Esmaeili, J. Rakhtshah, Ultrasound assisteddispersive-modification solid-phase extraction using task-specific ionic liquid immobilized on multiwall carbon nanotubes for speciation and determination mercury in water samples, Microchem. J., 154 (2020) 104632. https://doi. org/10.1016/j.microc.2020.104632
- [38] M.K. Abbasabadi, Nanographene oxide modified phenyl methanethiol nanomagnetic composite for rapid separation of aluminum

in wastewaters, foods, and vegetable samples by microwave dispersive, Food Chem., 347 (2021) 129042. https://doi.org/10.1016/j. foodchem.2021.129042

- [39] M.K. Abbasabadi, Speciation of cadmium in human blood samples based on Fe<sub>3</sub>O<sub>4</sub>supported naphthalene-1-thiol- functionalized graphene oxide nanocomposite by ultrasoundassisted dispersive magnetic micro solid phase extraction, J. Pharm. Biomed. Anal., 189 (2020)113455. https://doi.org/10.1016/j. jpba.2020.113455
- [40] L. Nekoozad, M.S. Barough, B. Salmasian, The Effect of X-Ray on plasma and erythrocyte concentration of Zn and Cu in radiology staff of Tehran oil Hospital, Int. J. Biomed. Biolo. Eng., 13 (2019) 480-484. https://doi.org/10.5281/ zenodo.3593166



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# A review: Exploratory analysis of recent advancement in green analytical chemistry application

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#### A B S T R A C T

It is always a concern to ensure personnel and environment safety in the field of chemistry which has caused to development in green analytical chemistry methods. Green chemistry aims to create an eco-friendly environment in laboratories by using various analytical methods/strategies to reduce the use of toxic solvents which are harmful to humans and the environment. It is a way to protect the environment by using green solvents and methods. Green analytical chemistry is a rapid analytical technique that describes the separation, identification, and quantification of an analyte in drugs, environments, and humans. Various green methodologies such as automation, miniaturization, precipitations, and passivation are utilized in the recovery of solvents and reagents. Green analytical chemistry aims to create an eco-friendly environment in the laboratories by using various analytical methods/strategies to reduce the use of toxic solvents which are harmful to the environment/ humans as well as to decrease the amount of waste generated. In this review, we explore different green solvents that can replace other toxic solvents used during extraction processes. In this review, the various extraction methods and analytical techniques used to analyze different components have been discussed.

#### 1. Introduction

Green chemistry is an emerging field that is concerned with the concept of process design and yielding products that are sustainable and benign to the environment and humans, it was introduced by Anastas in the year 1999 and works on the 12 principles points [1]. The green chemistry fields provide continuously environment-friendly compounds or develop processes that avoid the use of hazardous organic chemicals (benzene, toluene, VOCs solvents). Optimizing the quality of the result

\*Corresponding Author: Yunes M. M. A. Alsayadi Email: yunes20171@gmail.com https://doi.org/10.24200/amecj.v7.i01.279 as well as enhancing environmental friendliness prove to be a major hurdle in the future of green analytical chemistry. The first action taken by chemists for the design of green pharmaceutical products and industrial-scale processes offers enhanced economic development [2]. The use of green raw materials, and avoiding toxic chemicals are needed to be considered by chemical industries and companies for the safety of workers and environments. Further, these steps account for better yields and lesser waste. In green analytical chemistry (GAC), the environmentalfriendly analyte in samples is of greatest demand. This is important to improve the quality of the sample. The main problems that come in an environment because of using traditional methods can be minimized by

the use of the *in-situ* process, automation process, taking a minimum number of samples, minimizing reagent waste production, determination of multianalyte, direct analysis method, use of safe reagent and miniaturized processes. GAC is environmentfriendly, and safer to the analyst achieved by the use of more suitable alternative solvents, avoiding too many steps that will result in a clean, healthy and safe environment [3]. In green analytical chemistry, the strategies are applied to utilizing green methodologies and safe solvents to minimize the hazardous waste [4]. Sometimes, it is very difficult to avoid the use of harmful chemicals, in such cases, a minimum volume of solvents should be used. There must be work for the recovery of solvents and reagents. The best method to reduce sample consumption is by automation, miniaturization, precipitations, and passivation [5]. Automation and miniaturization are considered green analytical methodologies that reduce the sample size and reagent consumption. In green analytical chemistry, the miniaturized techniques are solvents, more sustainable, and minimize the risk of exposure from hazardous solvents to analysts [6]. The National Environmental Methods Index (NEMI) was established to evaluate the greenness of designed analytical methods concerning four criteria i.e. use of PBT (persistent, bio-accumulative, and toxic), use of hazardous chemicals, corrosiveness based on pH time of the analysis and waste [7]. Pfizer scientists in the practical guide have mentioned a new green solvent for medicinal chemists [8] Titanocene dichloride (Cp,TiCl) that confirmed the principle of green chemistry with high catalytic activity, high selectivity, low toxicity and environmentally compatible industrial chemical process. The catalysis procedure minimizes waste generation and contributes to the development of a sustainable chemical process [9]. This contributes to developing a suitable process with a high step economy and with a low risk of human health [10]. The most widely used safe transition metal on the earth is titanium which can be converted into Cp<sub>2</sub>TiCl, which is eco-friendly and catalyses the haemolytic cleavage of C-O, C-halogen, O-O bonds, and carbonyl compounds [11]. The idea of Green analytical chemistry (GAC) emerged and focused mainly on environmentally friendly laboratory practices rather than industrial scale. Certain modifications in terms of extraction (solventless extraction), analysis, and miniaturization were required to make the laboratory practice more environment-friendly and increase the quality of the result (Fig. 1). This led to the design of guidelines in the form of 12 principles which are as follows [12]:

- Use of Direct Analytical technique to avoid sampling preparation.
- Keeping the size and quantity of the sample as small as possible.
- Performing in-situ analysis.
- Carrying out the integration of the analytical process and operations.
- Selection of automated and miniature methods.
- Minimizing or avoiding unnecessary derivatization.
- Avoiding the generation of a large amount of analytical waste.
- The method that analyses multiple analytes at a time is preferred.
- Minimizing the use of electrical energy.
- Selecting the reagents from a renewable source.
- Removal and replacement of toxic reagents.
- Increasing and ensuring the safety of operators.

## **2.** Green solvents and sample preparation in analytical chemistry

Sample preparation and analysis are the two major steps in the analytical techniques. In analytical chemistry, sample preparation is considered the most important and time-consuming step [13]. The solvents to be used in sample preparation for analysis should meet the criteria of being as non-toxicity, biodegradability, recyclability, and availability [14]. Various organic solvents (methanol, ethanol, acetonitrile, hexane, etc.) are widely used for sample preparation because of easy availability and economical. Some of these solvents show unwanted residue because of long persistence and may be toxic [15]. So, in recent years, a step to develop environmentally friendly solvents has been given much emphasis (such as ionic liquids). The various green solvents developed are discussed as below sections.



Fig. 1. Principles of using green analytical chemistry in different fields of science

#### 2.1. Conventional green solvents

The solvents are considered a major part of the chemical industry and play a determinant role that impacts the safety, cost, and concerning health issues. Ethanol is a green solvent, less toxic compared to methanol and acetonitrile. Ethanol is one of the most used solvents. In analytical chemistry, especially in HPLC an ethanol/water mixture is being widely used as a replacement for other solvents and is considered a greener alternative to methanol and acetonitrile. e.g. separation of sunscreen molecules and pesticides (triazines) achieved through chromatographic techniques [16]. In another example, ethanol suitably replaces acetonitrile and methanol in HPLC i.e. the study of two test mixtures of a series of alkylbenzenes and a mixture of compounds of different classes such as caffeine and *p*-hydroxybenzoic acid [17].

The use of butyl alcohol replacing methanol and acetonitrile was performed for the separation of vitamins. In this study vitamins A, E, K1, and D3 were separated using a modified C-18 column eluted using surfactant sodium dodecyl sulfate (SDS) [18]. Ethyl lactate is a common solvent used in product purification in pharmaceutical, and paint industries because it has low toxicity, low viscosity and is biodegradable [19]. Glycerol (studied as an effective solvent), a major byproduct of biodiesel production, is non-volatile, non-toxic, non-flammable, and biodegradable [20]. The solvent 2-methyl tetrahydrofuran (2-methyl THF) is a renewable source and can be used as a substitute for THF and other solvents [21]. It showed a quick and greener approach to LC-MS detection of multiple mycotoxins. The study includes replacement of the solvent acetonitrile with ethyl acetate for mycotoxin analysis and minimizing solvent consumption. Extraction was promoted with the help of sonication and the addition of sodium sulphate [22]. Shawky *et al* in the year 2018 changed the extraction solvent to a greener one and evaluated the cytotoxic activities of Crinum (Amaryllidaeae) alkaloid. The results showed that the use of natural deep eutectic mixtures (Choline chloride: Fructose, molar ratio 5:2) along with the non-ionic surfactants (Genapol X-80) enhance the biological activities of the extracted drug [23]. Bi *et al* in the year 2018 developed a novel green and stable dissolving system KOH/ urea to successfully dissolve chitosan chains. The KOH-urea bonded with chitosan showed good thermal stability. Thus the solvent mixtures were used for further modifications of the chitosan as carboxymethyl chitosan, *N*,*N*,*N*-trimethyl chitosan and hydroxyl butyl chitosan [24]. Table 1 and Table 2 shows solvent selection and solvent replacement adopted by several companies as a green chemistry approach [25].

No.	Green solvents	Acceptable solvents	Disagreeable solvents
1	Ethyl acetate	Isooctane	Dichloroethane
2	Ethanol	Toluene	Di-isopropyl ether
3	Methyl ethyl ketone	Tetrahydrofuran	Methylpyrrolidinone
4	Water	Cyclohexane	Pentane
5	Acetone	Heptane	Hexane
6	Isopropyl acetate	Acetonitrile	Chloroform
7	Methanol	2-methyltetrahydrofuran	Dimethylformamide
8	t-Butanol	Dimethyl sulfoxide	Dimethyl acetate
9	2-Propanol	Methylcyclohexane	Diethyl ether

 Table 1. Pfizer solvent selection, solvent solvents and comparing them

 Table 2. Replacement of unwanted solvents with alternative solvents with lower toxicity

No.	<b>Unwanted solvents</b>	Alternative
1.	Pentane	Heptane
2.	Hexane	Heptane
3.	Dioxane or Dimethoxyethane	2-MeTHF or tert-butyl methyl ether
4.	Dimethylformamide, Dimethyl acetamide	Acetonitrile
5.	N-methyl pyrrolidinone	Acetonitrile
6.	Dichloromethane (extractions)	EtOAc, MTBE, toluene, 2-MeTHF
7.	Benzene	Toluene
8.	Dichloromethane (chromatography)	EtOAc/heptanes

#### 2.2. Unconventional solvents 2.2.1.Ionic liquids (ILs) and deep eutectic solvents

Ionic liquids (ILs) are organic salts that are liquid at room temperature and have low melting points (<100°C). Ionic liquid has low vapour pressure, low combustibility, and higher thermal stability and, can easily dissolve various polar and nonpolar compounds [13]. The ionic liquids work as an extraction solvent in dispersive liquid-liquid micro-extraction (D-LLME). Magnetic ionic liquids as extraction solvents can be prepared by incorporating paramagnetic components into the IL structure. The solvent thus prepared can be used in various liquid-phase micro-extraction methods. The ionic liquids can also be employed in single-drop micro-extraction. Ionic liquidbased surfactants as solvents are used in magneticassisted extraction methods for the analysis of biological samples [26]. Despite the numerous applications of ILs in sample preparation methods, toxicity profile and poor biodegradability posed a serious problem. Also, the synthesis process of ILs is found to be expensive and time-consuming. So to overcome these problems many research works were carried out which gave rise to a new class of solvents called deep eutectic solvents (DESs) [14]. DESs are synthesized by mixing a hydrogen bond donor and a hydrogen bond acceptor-containing compound whose melting point is lower than the melting point of the individual components. Due to its low cost, low toxicity, and biodegradability nature, CHCl is the preferred choice of organic salt to produce the eutectic mixture with various hydrogen bond donors such as urea, thiourea, glycerol, ethylene glycol etc. The DES thus formed is biodegradable and non-flammable making it easy to store. The synthesis process of DES is easy, no purification is required and environment friendly which makes it a better choice of solvent [14, 15]. DES is safe with low toxicity as a green solvent compared with other solvents [27]. The acid-base produced from fatty gas-based deep eutectic solvents significantly improves extraction efficiency using

the micro-extraction technique. The phenolic compounds were extracted through the microextraction technique with the support of N<sub>2</sub>. The fatty acids used in the preparation of deep eutectic solvents have carboxylic acids with long alkyl chains C<sub>8</sub>, C<sub>9</sub>, C<sub>10</sub>, C<sub>11</sub>, and C<sub>12</sub>. Taking the molar concentration ratio and a class of fatty acids into consideration, the effects of DESs can be adjusted or improved. By applying the principle of green chemistry, the separation and extraction of phenolic compounds from a large volume of water samples can be increased. The use of NH<sub>2</sub>.H<sub>2</sub>O as an emulsifier after reaction with DES forms a cloudy solution and thus increases the efficiency of extraction. The maximum extraction efficiency was achieved > 91.0%. The DESs technique was eco-friendly during the air-assisted liquid-liquid micro-extraction with the good recoveries of phenolic compounds in water samples (87.4% and 106.6%) [28]. Deep eutectic solvents are also used in the desulfurization process *i.e.* for the removal of organic sulphur from liquid fuels. It involves the extraction methods for the removal of organosulfur from fuels based on differences in solubility. The examples of non-toxic deep eutectic solvents identified are acetone, methanol, sulfolane, and nitrogenous solvents. The ecofriendly deep eutectic solvents prepared by using CHCl: Ph (1:4 molar ratio) were selected for the desulfurization of model liquid fuel. It is a new perspective for industry and is beneficial because it is important to recycle the solvents rather than degenerate them. During the desulfurization, solvent utilization was reduced to around 50-60% and desulfurization of fuel was achieved up to 99.99%. Thus this method of desulfurization may be regarded as one of the potential applications for the green industry [29]. Deep eutectic solvents reportedly improve therapeutic efficiency in the evaluation of drug delivery systems [30], used to determine trace constituents of curcumin in nutrition and herbal tea samples [31] and also in the quantification of Aflatoxins (AFs) types in rice samples [32]. Deep eutectic solvents (DESs) are categorized as probable replacements

of volatile organic compounds. Li et al in the year 2020 synthesized three hydrophobic DESs by mixing trioctylmethyl ammonium chloride as a hydrogen bond acceptor and decanoic acid, ketoprofen and gemfibrozil as hydrogen bond donors. The mixtures were characterized by their melting points, viscosity, density etc. The solvents were used to extract the bisphenol in water contaminants using vortex-assisted liquidliquid micro extraction and were analyzed with the help of HPLC. The methods developed have a limit of detection (LODs) and (LOQs) in the range of 0.3-0.5µg  $L^{-1}$  and 0.06-0.08µg  $L^{-1}$  and were used successfully for the determination and extraction of four bisphenol in the food contacted plastic samples [33]. In another study micro-scale extraction of bioactive phenolics from Vitis vinifera leaves using microwave-assisted solid-liquid extraction with ionic liquids (ILs) based surfactant was used by Mastellone et al in the year 2020. The extracted phenolics were analyzed by HPLC and photodiode array detection [34]. Natural deep eutectic mixtures (NADES) are another class of green solvents. These are different from eutectic mixtures in that they contain natural compounds (sugars, amino acids and organic acids and bases). The solvents with glucose and lactic acid were used by Paradiso et al in the year 2019 for the liquid-liquid extraction of selected polyphenols e.g. hydroxytyrosol and its derivatives in olive oil [35]. Another scientist Fraige et al used the natural eutectic mixture fort the extraction of metabolites in Byrsonima intermedia leaves. The mixtures are easy to prepare, non-toxic, inexpensive and are alternative to organic solvents. The natural eutectic mixtures contain five choline chloride-based deep eutectic solvents. The choline chlorides/glycerol considered as best and is having similar efficiency as that of most common solvents. The mixtures was used to extract seven phenolics contents viz. digalloyl quinic acid, proanthocyanidin dimer, dimer, quercetin-Ogalloylproanthocyanidin hexoside, galloyl quercetin hexoside, quercetin-O-pentoside, and galloyl quercetin pentoside [36]. Zhang et al in the year 2019 also utilized natural deep eutectic mixtures as both extraction and dilution matrices for the analysis of volatile components in Ipomoea-carried sweet (ICS) leaves. He used choline chloride along with glucose in the ratio of 1:1 molar ratio for the extraction of volatile components (2,4-di-tert-butylphenol),  $\beta$ -caryophyllene,  $\beta$ -element and others. This makes natural eutectic mixtures work as both extraction and dilution matric for static headspace gas chromatography-mass spectrometry [37]. Liu et al utilize the high viscosity of green deep eutectic solvents (choline chloride) for the extraction of polyphenols in palm samples. Choline chlorideoxalic acid-water *i.e.* Deep eutectic mixtures: H<sub>2</sub>O in the ratio of 1:0.1 to 1:50 were used for the extraction of target compounds (polyphenols, protocatechuic acid, catechins, epicatechins and caffeic acid). Thus proper mixtures of deep eutectic mixture with water work as efficient extraction solvents for bioactive compounds [38]. Also, the green eutectic solvents, zinc chloride and acetamide in different molar ratios in non-ionic surfactant (Triton X-114) as extraction methods used for the determination of trace vanadium in water as vanadium with ammonium pyrrolidine in complex form [39]. Figure 2 shows conventional analytical chemistry vs green analytical chemistry approaches.

#### 2.3. Reuse of solvents

Solvents are not reused in pharmaceutical chemistry because of the concern for contaminants. However ionic liquids (ILs) are good alternatives to organic solvents because of their good solvent properties for both organic and inorganic materials and low volatility. They are a group of salts that are liquid at room temperature and thermally robust (stable up to 260°C), which makes them reusable and suitable to work in high temperatures. The physical-chemical properties of ILs can easily be tuned by design, as it is influenced by their cationic and anionic species for extraction and removal of pollutant application in air or water samples [40, 41]. Figure 3 shows the green analytical chemistry based on IL approaches.



Fig. 2. Green analytical chemistry approaches based on green solvents[39]



Fig. 3. Tree of green analytical chemistry approaches [40,41]

## **3. Methods: Different techniques in green analytical chemistry**

#### 3.1. Infra-red spectroscopy

The infrared range of its three regions; middle infrared region (MIR), near-infrared region (NIR) and Raman spectroscopy are the main vibrational techniques employed for the direct analysis of samples. IR spectroscopy detects vibrations when there is a change in electrical dipole moment in compounds, whereas Raman spectroscopy is a measurement of the electrical polarizability of excited molecules induced by the electric field. In biomedical research, infrared spectroscopy is a rapid, accurate, and sensitive technique mainly for the detection of illness. In research, IR can be used in cancer detection and provides a green analytical approach in clinical oncology and cancer research. Different sampling modes can be used in NIR for various applications. Different modes of measurement in the instruments are transmittance, diffuse transmittance, transflectance and diffuse reflectance. Among these diffuse reflectance and diffuse transmittance are the two most used measurement modes. Diffuse reflectance measurements are performed in the range from 1100 to 2500 nm with a sample thickness of 1 cm, whereas diffuse transmittance is performed in the range of 800-1100 nm using a sample thickness of 1-2 cm. Due to its reliability, accuracy, and nondestructive nature, the NIR technique is considered the most widely used on/in-line process for checking the physical and chemical parameters [42]. Filter instruments used in the NIR range are considered rapid techniques and fulfill the criteria of robustness, compactness and reduced cost. There are three types of NIR on-line analyzers: remote sensors, bypass analyzers, and fibre optic probes [42]. The first on-line NIR sensor was based on a sensing head remote approximately 200 mm at an angle of  $60^{\circ}$  from the horizontal of the flow sample. Their advantages rely on the low cost of instrumentation and easy installation. It is sensitive to atmospheric humidity, interference from ambient light variation and dust build-up on the optical surface. IR can be used in the food

industry for moisture determination on powders, bread [43], control of protein in flour [44] and meat composition analysis [45]. NIR by-pass samplers were first developed to measure the protein content of flour and are still the most popular application of the NIR feedback control system. In this method, the sample is placed against the optics window for each measurement cycle. The samples were taken directly from the flour stream either from the positive pressure blow line or gravity-fed spouting [46]. Fibre-optic probes have the widest range of applications as on-line analysis. Laboratory NIR analyzer platforms have been modified, enclosed in process-hardened housing, and used extensively for material identifications in the pharmaceutical [47] and nutraceutical industries [48]. Mazivila et al in the year 2019 developed FT-IR methods to study polymorph I and II of pharmaceuticals cocrystals of lamivudine and theophylline. With the combined effect of FT-IR and multivariate curve resolution with alternating least-squares (MCR-ALS), it was able to monitor in line synthesis of co-crystal. Also this method allows understanding the mechanism of synthesis through identification of intermediates. The spectral shows clear strong overlapping and quantification concludes for the existence of final products as co-crystals [49]. Weldegebreal et al demonstrate new fast and costeffective analytical methods for the determination of caffeine in green coffee beans using FT-IR and fluorescence spectrophotometry. Caffeine was also determined in the NIR range using dimethyl formamide (DMF) as the solution. The caffeine content in green coffee beans comes out to be  $1.50 \pm 0.14$  (% w/w) using FTIR-ATR in the NIR range and  $1.50 \pm 0.05$  (%w/w) using fluorescence spectroscopy [50].

#### 3.2. Raman spectroscopy

In Raman spectroscopy, a µm-size of samples can be analyzed by using a microscope, and no sample pre-treatment is required. This makes the process less tedious than other spectroscopy method [51]. Raman spectroscopy is a non-destructive technique that provides rapid analysis of untreated samples.

Raman spectroscopy is useful for acquiring inorganic and organic vibrational structure information. This technique is another alternative to analyzing the samples directly over the glass or plastic packages. Raman spectroscopy is efficiently used in the solid state to identify the graphene layers by the Raman spectroscopy. In the Forensic and pharmaceutical field, Raman spectroscopy is applied to analyze the entire sample. Cocaine, heroin, and various other phenethylamine moieties are quantified and identified accurately and rapidly by Raman spectroscopy. The other use of Raman spectroscopy is the identification and analysis of pigments used in antique paintings [52], and glass which may in the future help to restore ancient articrafts [53]. This technique was applied by Legner et al in the year 2019 for the real-time analysis of the alcohol fermentation. The conversion of sucrose was performed by yeast cells immobilized in alginate beads. This Raman technique provides faster and robust quantitative spectroscopy for ethanol, glycerol, and sugar content. The Raman spectroscopy is coupled with NMR for resolved signals and higher specificity. This provides the quantitative monitoring of the different reactants [54]. Molnar et al in the year 2019 used Raman food spectroscopy for authentication. The technique is green as it requires a minimum sample preparation step. The authors use this for the honey sample preparation (honey diluted to water 1:1 w/v) to get reproducible spectra and avoid the formation of crystallization and fluorescence [55].

Raman spectroscopy offers some advantages over NIR spectroscopy. The Raman spectroscopy gives a simple optical configuration that is efficiently interfaced for on-line quantification. Raman spectroscopies are used for quality control (QC) analysis for a wide range of liquid samples filled in glass (clear or amber) or plastic containers. Raman spectroscopy has been selected for the static analysis of ethanol content of spirits whiskey, vodka, and sugary alcoholic drinks in 200 ml (flat) and 700 ml (round) glass bottles. The technique is restricted to the analysis of clear glass bottles because coloured bottles exhibited strong fluorescence. The quantitative in situ analysis of povidone present in eyewash solutions in lowdensity polyethylene (LDPE) bottles was also made possible by Raman spectroscopy. For on-line and in-line applications of Raman spectroscopy, the spectrometer is incorporated into the sampling location using conventional optical fibre cables [56], enabling remote sampling at tens or even hundreds of meters from the spectrometer. The advantage of Raman spectroscopy is that it is used for both qualitative and quantitative analysis of materials kept inside containers of polymeric bottles, and blisters. Raman spectroscopy has been used to detect liquid explosives within bottles/ plastic packaging [57] and illegal drugs dissolved in beverages [58]. It can also be used to find out the active ingredients in pharmaceuticals inside plastic bottles [59], amber vials [60] or capsules [61]. The methods for the quantitative determination of ethanol contents in beverages inside glass bottles by Raman [62] are another application. This implies that bonds that connect two identical or practically identical parts of a molecule can be more active in Raman than in IR, thus providing complementary spectral information, *i.e.* O-H stretching vibration is very strong in IR but very weak in Raman.

Similarly, IR also holds certain advantages over Raman spectroscopy. The important one is cost-effectiveness. It is much cheaper than Raman as it uses high powered laser source to get sensitive results. The high power of the laser may cause heating and destroy the sample. Raman spectroscopy is fewer sensitive technique except surface-enhanced Raman spectroscopy than IR when used alone. NIR can sample large areas and has better penetrability than Raman spectroscopy. Both Raman and IR spectroscopy have advantages over each other but to get more sensitive results it is better to use both techniques together. In many studies, Raman was used for the characterization of nano adsorbents which were used for the extraction of pollutants from water samples. Faghihi et al showed that Mn ions were extracted in human blood/serum samples by AMTZ@MWCNTs adsorbent. They showed that Raman of AMTZ@

MWCNTs has two main peak shifts (D band: 1305 cm<sup>-1</sup>) and another peak at (G band: 1581 cm<sup>-1</sup>), which confirms the structure of MWCNTs. Raman peaks of MWCNTs with G and D bands confirm the carbon structures. Also, the low height of peaks at 243, 513, 705, and 2612 cm<sup>-1</sup> were observed by MWCNTs Raman. The ratio of the IG/ID bands showed the sample's graphitization and the quality of the tubes in MWCNTs. Also, Mohammadi et al used bismuth oxide/ titanium oxide nanoparticles functionalized nanographene oxide (Bi<sub>2</sub>O<sub>3</sub>/TiO<sub>2</sub>@ NGO) and IL-NGO for the removal of HCHO and toluene from the air, respectively. IR of Bi<sub>2</sub>O<sub>3</sub>/TiO<sub>2</sub>@NGO and IL-GO showed functionalized IL and Bi<sub>2</sub>O<sub>3</sub>/TiO<sub>2</sub> on NGO[63, 64].

#### 3.3. Chromatographic techniques

Capillary zone electrophoresis is an alternative to chromatographic techniques but cannot be used for non-charged molecules. Micro-emulsion electrokinetic chromatography (MEECK) is one of the techniques applied by Felici *et al* in the year 2016 for the analysis of non-charged antiparasitic drugs such as ivermectin (IVM) and moxidectin (MXD). The technique is environment-friendly, robust, specific, and sensitive techniques for analyzing these macrocyclic lactone drugs. The LOD value of the two drugs IVM and MXD were  $3x10^{-3} \mu g L^{-1}$  and  $3.6x10^{-3} \mu g L^{-1}$ [65].

#### 3.4. Sample Extraction

In the field of natural sources and agri-food industries supercritical fluid extraction (SFE) using CO<sub>2</sub> and pressurized liquid extraction (PLE) are the most widely green analytical techniques used apart from the conventional methods of solid-liquid extraction (SLE). Another scientist Abukhadra et al synthesized supported bentonite/ chitosan on green fabricated Co<sub>2</sub>O<sub>4</sub> and evaluated their adsorption properties for Congo red dyes and Cr (VI) ions. The chemisorption phenomenon exists for the removal of the dye and Cr (VI) and follows a pseudo-second-order model. The adsorption maximum was 303mg g<sup>-1</sup> and 250mg g<sup>-1</sup> for Congo red molecules and Cr (VI) metal ions. The bentonite/chitosan@Co3O4 was found to be highly useful in the purification of water and may act as promising adsorbents [66].

	<b>Table 3.</b> Various samples were analyzed using SPE based on different cartridges and solvent
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Sample	Cartridge	CS	Analyte	Solvent	Instrumental	Ref.
Wine	OASIS/ HBL	Methanol/DW	Azolic/fungicides	Methanol	MS-HPLC	[49]
Water	ENVI-18	Acetonitrile	PAHs	Hexane	GC-MS	[50]
Tomatoes	Silica(C <sub>18</sub> )	Methanol/water	P&H	Dichloromethane	HPLC	[51]
Plasma(H)	OASIS/ HBL	Methanol	PhCM	Methanol	UPLC/MS/MS	[52]
River	OASIS/ HBL	Acetonitrile/DW	**Antibiotic	***ACA	LC-MS	[53]

CS: Conditioning solvent

*P&H: Pesticides and Herbicides* 

*PhCM: Phenolic compounds and their metabolites* 

\*\*Antibiotic: Antibiotic Macrolides

\*\*\*ACA: Ammonium acetate/acetonitrile (50:50)

#### 3.4.1. Solid-phase extraction

Solid-phase extraction is the most widely used technique for extraction, and cleaning of analyte from environmental, food and drinks [67]. In SPE, the analyte solution to be extracted is loaded into a cartridge containing the sorbent. The impurities and undesired components are then washed away by using suitable solvents whereas the desired component is collected using a suitable solvent [67-71] (Table 3).

#### 3.4.2. Solid-phase micro-extraction (SPME)

The SPME method is identified as a green approach to reduce solvent consumption and waste during the preparation of the sample. SPME has a certain advantage such as it is solvent-free. A small volume of sample is required and is considered highly sensitive. SPME can analyze the non-polar compound in liquid, gas, and solid samples with various instruments with HPLC and GC, etc. In SPME, the coating of a fused silica fibre is done with a stationary phase. Equilibrium between the analyte in the sample and the fibre is attained by exposing the fibre to aqueous or gaseous samples. The analyte can be desorbed from the fibre thermally or by using solvents. The thermal desorption of the sample refrains the use of solvents. Similarly, chemical desorption uses a very low amount of sample (in  $\mu$ L). Thermal desorption and chemical desorption are two greener techniques. In a study conducted by Andrade M.A et al. to determine the Ochratoxin A in wine, they utilized the SPME technique for sample preparation as it is necessary for analyte purification and to achieve a lower detection limit of the sample or analyte [72]. Rivellino S. R. et al. utilized the SPME technique to detect the extraction artifacts in the analysis of honey volatiles. In this, the honey mixture was heated at 40°C for 30 seconds and was stirred before extraction. A specific amount of honey mixture was then added directly into a vial having a magnetic stirrer in a saturated aqueous solution of sodium chloride [73]. In another study performed by Merola G et al., they combined SPME and GC

for the measurement and determination of various recreational drugs such as ketamine, methadone, amphetamines, cocaine, coca ethylene, and  $\Delta^9$ -tetrahydrocannabinol (THC) in hair. The limit range for detection and quantification is from 0.01-0.12 ng mg<sup>-1</sup> and 0.02 to 0.37 ng mg<sup>-1</sup> [74]. Schacker R *et al.* carried out fermentation using *Monascus* fungus and checked the viability of the use of SPME in the extraction of various microbial volatile organic compounds such as ethanol, 2-methyl-propanol, 3-methyl-butanol, 2-methyl-butanol, and 2-phenyl-ethanol. During a fermentation process, the SPME provided greater accuracy when assessing the MVOCs (*in vivo*) [75].

#### 3.4.3. Liquid phase micro extraction

Liquid phase micro-extraction is a simple approach for sample pre-treatment and utilizes a very small amount of solvent. Less than 10 µL of extraction solvent is required to extract analyte from the sample. It can be combined with GC and CE for the detection of the extracted analyte [76]. Liquid phase micro-extraction finds its uses in clinical and forensic science as well as in the analysis of food and drinks. The proper selection of the different forms of liquid-liquid micro-extraction is very important. This was demonstrated by Saraji et al in the year 2018 using three solvents chloroform, 1-butyl-3-methylimidazolium tetrafluoroborate, and 1-hexyl-3-methylimidazolium hexafluorophosphate for phenolics, neutral phenolics and amino compounds. Analytes with polar groups give the best results with in-situ ionic liquid dispersiveliquid-liquid micro-extraction, whereas neutral hydrocarbon compounds with polar groups use chloroform solvent called dispersive liquid-liquid traditional microextraction techniques. Hydrophilic analytes were not suitable to be extracted by any of the liquidliquid micro-extraction [77]. Liquid phase microextraction is of three types.

- i. Hollow Fibre Liquid Phase Microextraction (HF-LPME)
- ii. Dispersive Liquid-Liquid Microextraction

### iii. Single Drop Microextraction (SDME)3.4.4. Hollow fibre liquid phase microextraction (HF-LPME)

In HF-LPME, the hydrophobic pores present in hollow fibre hold the extraction solvent during the extraction process. The hollow fibre is attached to the needle of a micro syringe. Once the extraction process is complete, the extraction solvent containing the analyte is collected and then analyzed using a suitable instrument [78]. In this process, less volume of solvent is consumed during extraction  $(2-25 \ \mu L)$ .

### 3.4.5. Dispersive liquid-liquid micro-extraction (DLLME)

In DLLME, the partitioning is between mixtures of extraction solvent-dispersive solvent and the aqueous phase in which the sample is dissolved. The mixture is then added to the sample solvent with the help of a micro syringe, which gives rise to an emulsion-like solution. The formation of cloudy microdroplets partitioned the sample between the extraction phase and the aqueous phase. The solution is centrifuged, breaks the emulsion into two phases and the sediment phase so formed is then collected and analyzed [79]. The technique uses minimum amounts of 1-100 µL of extraction solvents are used. In DLLME most common solvents used are acetonitrile, methanol, ethanol, and acetone whereas, other solvents such as carbon tetrachloride, toluene, hexane, chlorobenzene, 1,1,2,2-tetra chloroethane may also be used. The techniques were used for the determination of Caffeine, alkaloids, amino acids, cinnamic acids, coumarin, curcumin, essential oils, pesticides and many more [80]. Pacheco-Fernández et al used ILs-based surfactant watersoluble octyl guanidinium chloride (C8Gu-Cl) of low cytotoxicity to water-insoluble IL microdroplet by metathesis reaction and extracted personal care products. The methods were used in combination with HPLC and diode array detection. The metathesis used the addition of anion exchange reagent (bis[(trifluoromethyl) sulfonyl] imide-1:1

molar ratio). It gives low detection limits of 0.4µg L<sup>-1</sup> with the use of low volume of IL surfactants [81]. Sheikh et al developed novel methods for the determination of cadmium and lead in the ground water samples (tube well and hand pump) by liquid-based vortex-assisted dispersive liquid-liquid micro-extraction. The contents were determined ion the scalp of the children (1-3 years) exposed to contaminated water and domestically treated water. A green chelating agent ,-cysteine (2-amino-3-sulfhydrylpropanoic acid) was used to concentrate the Cd and Pb and ionic liquid 1-butyl-3-methylimidazolium hexafluorophosphate [BMIM] [PF<sub>6</sub>] used as micro extractant [82]. Fast deep eutectic mixtures were used for the first time for the extraction of phthalates from food beverages by Santana-Mayor et al. The technique used is dispersive liquid-liquid micro-extraction. Choline chloride: The phenol mixture shows good results for the extraction of different phthalates and was coupled with HPLC-diode array detection for separation and analysis [83].

#### 3.4.6. Ionic liquids application

Ionic liquids (ILs) as green solvents based on organic cation (R- imidazolium/pyridinium) and organic or inorganic anion (Halogen Cl, Tetrafluoroborate  $BF_4$  and Hexafluorophosphate  $PF_6$ ) have a critical role for extraction pollutants from different matrixes. ILs are three form, hydrophilic ([R-IM]  $[A^-]; A = NO_v, Cl, OH)$ , hydrophobic ( $[R-IM][A^-];$  $A = PF_{a}$  and intermediate ([R-IM][A<sup>-</sup>];  $A = BF_{a}$ ), so, they used in various matrixes. A special form of Task-Specific Ionic Liquids (TSILs) such as Trioctylmethylammonium thiosalicylate (TOMATS) was used for extraction of heavy metals, such as mercury in water samples (Fig. 4a). In TSIL, the thioether, urea, or thiourea functionalized on imidazolium cations act as metal ligating moieties, whereas the PF6<sup>-</sup> anions provide the desired water immiscibility. Due to the unique physicochemical properties of ionic liquids, they can replace traditional organic solvents in a very efficient manner. These properties involve their good solubility in organic solvents, low

volatility, and most importantly their higher thermostability when compared to the traditional solvents. Due to those highly appreciated properties, a novel method based on dispersive liquid-liquid microextraction was developed for the preconcentration of cadmium in human body fluids and determination by electro thermal atomic absorption spectrometry [84]. It also has been reported that speciation and determination of trace inorganic and organic mercury species in water and caprine blood samples were performed by the mean of Ultrasound assisted-dispersiveionic liquid-micro-solid phase extraction based on carboxyl-functionalized nanoporous graphene [85]. A method for removing hazardous toluene vapor from the air was developed based on an ionic liquidphase adsorbent. Due to its high toxic effects on the environment, toluene needs to be removed by an efficient and qualified method. Five ionic liquids were pasted on micro glassy balls and used for toluene removal from air by liquid-gas-phase extraction method. Based on the proposed procedure, toluene vapor was absorbed on ionic liquids (0.2 g, 25 °C) and desorbed from it at 110 °C before being determined by gas chromatography [86,87]. Mercury is another highly toxic element which can be found in the daily used water, so an Ultrasound assisted-dispersivemodification solid-phase extraction using taskspecific ionic liquid immobilized on multiwall carbon nanotubes was performed for speciation and determination of mercury in water samples. This method was based on 1-(3-aminopropyl)-3methylimidazolium hexafluorophosphate (TSIL) immobilized on multiwall carbon nanotubes (MWCNTs@ [Apmim][PF6]) was used for speciation of inorganic/organic mercury (Hg2+, R-Hg or O-Hg) by ultrasound assisted-dispersive solid-phase extraction modification (UAS-DMSPE) which was combined with cold vapour atomic absorption spectrometry (CV-AAS) [88]. The chemical and physical properties of ILs and their potential in many applications caused to use in different fields of science (Fig.4b).



Fig.4. TSIL of TOMATS (left, a), the application of ILs in different fields of science (Right, b)

#### 3.4.7. Single drop micro-extraction (SDME)

A micro drop of extraction solvent  $(1-10 \ \mu L)$  is used to extract the analyte from the sample. During extraction, the micro drop of the solvent is held over the sample with the help of a micro syringe. The extraction solvent used should have low volatility, and low water solubility, and must be immiscible in water [89]. SDME techniques were used for the determination of essential oils and volatile components [80]. SDME has 3 different modes such as:

#### 3.4.7.1. Direct SDME

To the bulk aqueous sample solution, a micro drop of water-immiscible extraction solvent is directly immersed and stirred during extraction. By this method, both volatile and non-volatile analytes can be extracted [89].

#### 3.4.7.2. Headspace SDME

In this technique, the extraction solvent is exposed at the headspace above the sample with the help of a syringe. The extraction of volatile and semivolatile analytes in the sample is carried out. As this extraction technique is time-dependent, an extraction solvent with low volatility is used to reduce any chance of drop evaporation during extraction [90].

#### 3.4.7.3. Liquid-liquid-liquid micro-extraction:

In this mode, a thin layer of organic extraction solvent having a low density is formed over the sample solution. To the layer of extraction solvent, a micro drop of an aqueous solution is immersed. For the extraction to occur by this method, the pH of the sample solution should be maintained in such a way that non-charged analytes are formed [91].

Various samples analyzed using different Liquidphase micro-extraction techniques are given in Table 4 [92-103].

#### 3.4.8. Supercritical fluid extraction

Several researchers have carried out an experimental and theoretical investigation on the SFE using CO<sub>2</sub>. Many articles have been published on the studies for the SFE. In comparison with different extraction methods available, SFE provides numerous choices of solvents to be used. Supercritical fluid extraction is used for the extraction of non-polar organic compounds but can be used for polar ones also with the aid of modifiers [98]. The technique is robust, fast and highly efficient and has the advantage that it is easily hyphenated with MS for the separation and identification of bioanalytics [104, 105]. Supercritical carbon dioxide is a fluid state of

carbon dioxide, green and eco-friendly technology with the capability to extract composition without thermal degradation and oxidation [106].

	*	•	• • •	· · /	
Techniques	Sample	Analyte	Solvent (volume)	Instruments	Ref.
HF-LPME	Cucumber	7 pesticides	Chloroform (5 mL)	UHPLCMS/MS	[64]
DLLME	Wine	Ochratoxin A	Chloroform (660 µL)	HPLC-LIFD	[65]
DLLME	Honey	5 Fungicides	Dibromoethane (130µL)	GC	[66]
HF-LPME	Milk	9 StHor	Methanol (400 µL)	GC-MS	[67]
SME	Urine	CCM	Chloroform (10 $\mu$ L)	GC-PDHID	[68]
HFMSME	Saliva	CCM	Chloroform (10 $\mu$ L)	GC-PDHID	[69]

**Table 4.** Various samples were analyzed using liquid-phase microextraction techniques (LPME)

CCM: Cocaine and cocaine metabolites,

StHor: Steroidal Hormones

The advancement of green analytical chemistry minimizes the risk in the first step during sample preparation. The corona-charged aerosol detector considered as universal detector used to analyze a wide range of analytes. The structures of related impurities in risperidone were analyzed by a suitable reverse phase HPLC method using suitable solvents ethanol, acetone, and another modifier. The steps were optimized by experimental designed methodology and better separation occurred by using the simple Derringer's desirability with 3D surface plots. In the case of ethanol, the selected condition of the mobile phase has a flow rate of 0.6 mol min<sup>-1</sup>, column temperature of 37.5°C and organic modifier content of 20% (v/v) whereas, another method utilizes acetone as the mobile phase with flow rate 0.8 mL min<sup>-1</sup>, organic modifier content 17% (v/v) and column temperature  $37.5^{\circ}$ C. Both methods lack waste management principles but are well-validated and selective. Green analytical procedure index pentagrams represent one of the reported methods and are assigned as eco-friendly analytical methods. Ethanol is unsafe or harmful as compared to acetone so acetone is mostly preferred [107]. Supercritical carbon dioxide has many excellent improvements as being nontoxic, harmless, high-quality extraction, and safe in use. The supercritical fluid has a low cost that overcomes the conventional method of extraction. Supercritical fluid extraction CO<sub>2</sub> is an alternative green chemistry solvent, considered eco-friendly with good performance for disposal of impurities in oil-based mud. Extraction is obtained with economic efficiency and is applicable to remove the hydrocarbon contamination from the wastage of oil mud. Supercritical fluids CO<sub>2</sub>, propane, and freon are used to treat the wastes in the process of drilling. The increased pressure of extraction reduces the diffusion coefficient and will increase the density of SC-CO<sub>2</sub>. The saturated vapour pressure leads to an increase in SC- $CO_2$  solubility with the increase in temperature. According to this research paper, the disposal of waste in OBM (oil-based mud) result was found

in good agreement with extraction performance of 98% utilizing the pressure 20mpa time for 60 min and temperature of 35°C. Among the many advantages of supercritical CO<sub>2</sub> in the industry for the preparation of polymer material, medicine, and chemical components is that it is recycled after use [108]. Carbon dioxide and modifiers or co-solvents increased the extraction yield of cinnamoyl pyrrolidine amides from dried leaves of Piper hispidum. Around 5% methanol was added and reported improvement in the extraction yield of amide components. Polar compounds are extracted with the low polarity of SC-CO<sub>2</sub>. The plants which have pharmacological activity like Panax ginseng, Taxusbacata, Piper nigrum, Acoruscalamus, Curcuma longa, and Ocimum gratissimum, are recognized by the eco-friendly supercritical fluid CO2. The supercritical fluid has a good resolution for bioactive compounds in plants [108]. Cádiz-Gurrea et al in the year 2019 used food-grade solvents in the extraction of bioactive from the barks of Sclerocarya birrea through green analytical techniques. The author utilizes supercritical fluid extraction (SFE) and pressurized liquid extraction (PLE) to extract flavonoid aglycones [109]. Supercritical fluid chromatography with reversed phase chromatography using modifiers such as MeOH, ACN and MeOH: ACN 50:50 was used for the separation of the chlorophyll pigments by Lefebvre *et al* in the year 2019 [110].

#### 3.4.9. Carbon dots extraction

Carbon dots nowadays are considered under green analytical techniques as C-dots also follow the basic principle of elimination of toxic, expensive, hazardous substances and minimization of time and energy consumption. C-dots are used in sample preparation, imaging, bio sensing, and drug delivery using photovoltaic. The application is because the C-dots are natural, biocompatible, low toxicity and show the phenomenon of photoluminescence. Yuvali *et al* in the year 2019 utilize a first-time magnetic carbon nanodot/ graphene oxide hybrid material (Fe<sub>3</sub>O<sub>4</sub>@C-

nanodot@GO) for solid phase extraction (MSPE) of the drug ibuprofen in human plasma and its determination by MSPE-HPLC-DAD. Graphene oxide (GO) nanodots, C-nanodots and magnetic properties of nano Fe<sub>2</sub>O<sub>4</sub> are combined altogether. C-nanodots are prepared from the pasteurized cow milk then a step hydrothermal method was applied for the preparation of Fe<sub>2</sub>O<sub>4</sub>@Cnanodot@GO hybrid material. The nanodots by were characterized Fourier transform infrared spectrometry (FT-IR), X-ray diffraction spectrometry (XRD), Raman spectrometry, scanning electron microscopy, vibrating sample magnetometry and energy dispersive X-rays. The limit of detection was found to be (LOD) 8.0 mg L<sup>-1</sup>[111].

## 4. Application of green analytical chemistry

In the past, many products such as medicines, dyes, cosmetics, paints, polymers, etc. were manufactured or synthesized using various chemical processes. Along with the beneficial products many undesired and harmful substances are also produced and the reduction or removal of these undesired substances has become a major issue. So the need to introduce a greener alternative has recently gained interest. Green chemistry contributes to the synthesis of products in such a way that it provides less threat to health and the environment.

#### 4.1. Pharmaceutical Field

The synthesis of aspirin was carried out by microwave irradiation and by using a catalyst such as  $H_2SO_4$ , or MgBr<sub>3</sub>.OEt<sub>2</sub>, AlCl<sub>3</sub>, and CaCO<sub>3</sub>. The synthesis process was solvent-free and the production of waste products was at a minimum. The process was thus more environments friendly and a greener approach. Celecoxib an anti-inflammatory agent was synthesized by using a green approach during the developmental stage and it was found that the yield increased from 63% to 84% whereas, the waste product was clean enough

and was purified by washing with more amounts of solvents. The synthetic methods avoided the use of undesirable solvents such as methylene chloride and hexane [112].

#### 4.2. Diagnosis of Cancer

IR spectroscopy has been recognized as a green tool in the diagnosis of Cancer. It provides a completely greener and environmentally friendly style of analyzing the blood sample as the analysis does not use any kind of chemicals during analysis. Mohammadi *et al* collected blood samples from two different groups and analyzed them by using IR Spectroscopy in the range of 1800 to 900cm<sup>-1</sup>. Spectra were observed for N-H stretching, amide I and II bands. They also confirmed the shift in the  $\alpha$ -helix and  $\beta$ -sheet amide I band in the case of malignant tumours. The results were compared with the current clinical methods and showed 97.6% accuracy [113].

## 4.3. Application in the detection of heavy metals

Several heavy metals such as arsenic, cadmium, lead, zinc, etc. can be detected in the soil by using portable X-ray fluorescence which provides a rapid, simple and accurate metal analysis. By this method, a large number of heavy metals can be detected with a very low generation of residues and high sensitivity [114].

## 4.4. Application in Detection of Organic Pollutants

Various organic pollutants such as sulphonamide can be detected in water by using a liquid-liquidliquid micro extraction technique combined with HPLC (265 nm) (Lin CY *et al.*)[115]. Similarly, acidic pharmaceuticals such as salicylic acid, ibuprofen, diclofenac, etc. can be detected in wastewater by using hollow fibre-based microextraction followed by HPLC/MS-MS [80]. Organophosphates and pyrethroid pesticides in water can be detected by SDME coupled with GC-FID [116].

#### 4.5. Application in Analysis of blood sample

Human blood is a very complex bio-fluid containing WBC, RBC, glucose, hormones, minerals, etc. Qualitative and quantitative analysis of blood samples through IR spectroscopy can be done for the diagnosis of various diseases or for determining various blood constituents such as albumin, glycoprotein, fibrinogen, etc. This estimation will help in understanding the disease patterns [117,118]. Figure 5 shows the application of green chemistry in the analysis of blood samples. Also, some methods with ionic liquids or solvent-free based on nanotechnology were used for extraction and determination heavy metals in human blood samples (Fig.5) [119-123].

#### 4.6. Analysis of Particulate Matters

Different microscopic solid or liquid matter suspended in the atmosphere (dust, metals, VOCs) may have some harmful effects on human health. The particles present can be analyzed based on their chemical composition (inorganic and organic). The inorganic particle can be analyzed by colorimetric methods, ion chromatography, and selective ion electrodes (requires sample solubilization) mass spectrometer, atomic absorption spectrometry (AAS), inductively coupled plasma mass spectrometry (ICP-MS), IR Spectroscopy, Raman spectroscopy (sample solubilization not required). Organic particulates can be analyzed by thermal, optical, and extraction techniques. The analysis of solvent extraction is coupled with GC-MS or HPLC. The solvent during extraction is replaced by supercritical fluid  $(CO_2)$  making it a green approach [124-128]. In addition, the pollutants such as metals and VOCs may be determined in water samples by GC-FID, GC-MS, AAS, and ICP-MS [129-132].

### 4.7. Patent application on green analytical techniques

After the scaling up of analytical techniques from the laboratory scale to the industrial scale, it was necessary to make sure that the technique being used was greener and had no effect on the environment or the analyst. In the recent era, various modifications have been carried out to the analytical technique which has led to the development of various modern analytical techniques which make the detection and analysis of samples easy, rapid, and safe. Many types of research have been carried out for the analysis and extraction of samples using these novel techniques and patents have been filed for it [133-135].

#### 5. Conclusion

Green analytical techniques have a goal to design processes and methods in a way that they produce products that are sustainable and benign to both humans and the environment. The



Fig. 5. Use of ILs as green solvents for analytical chemistry in blood samples

greener analytical laboratories can be made after meeting the guidelines or requirements of GAC. Significant efforts have been made in recent years to develop methods for product development using greener techniques. The methods include the use of greener solvents, analytical techniques and different extraction methods. The analytical methods can be described as green by substituting toxic reagents/solvents with green solvents, reducing the use of reagents. The use of greener solvents also increases the safety of the operator. The utilization of newer greener methods for analysis can have an impact on the workplace to perform work quickly. The emphasis of the review is to discuss some greener extraction techniques used by the researchers including micro-extraction techniques and supercritical fluid extraction. Selection of greener solvents and replacement are the two methods adopted to replace hazardous solvents. The development of greener analytical techniques results in a reduction of solvent and minimization of waste production.

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#### Abbreviation

ACN	Acetonitrile
AF	Aflatoxins
BMIM	1-Butyl
	-3-methylimidazolium
CE	Capillary Electrophoresis
ChCl	Choline chloride
DES	Deep eutectic solvents
DLLME	Dispersive liquid-liquid
	phase micro extraction

DMF	Dimethyl formamide
FI-IK-AIK	Fourier transform
	infrared – attenuated
	total reflection
GAC	Green analytical
	chemistry
GC	Gas chromatography
GC- FID	Gas chromatography-
	flame ionization detector
GO	Graphene oxide
HF-LPME	Hollow fibre liquid
	phase micro extraction
HPLC	High-performance liquid
	chromatography
ILs	Ionic liquid
IR	Infrared
IVM	Ivermectin
КОН	Potassium hydroxide
LOD	Limit of detection
MCR-ALS	Multivariate curve
WICK-ILD	
	resolution – alternating
MEEGW	least squares
MEECK	Microemulsion
	electrokinetic
	chromatography
MeOH	Methanol
MIR	Middle infrared
MSPE	Magnetic solid phase
	extraction
MVOC	Microbial volatile
	organic compounds
MXD	Moxidectin
NADES	Natural deep eutectic
	mixture
NIR	Near infrared
DIE	Pressurized liquid
ILL	
50.00	Extraction
$SC-CO_2$	Supercritical-carbon
~~~~	dioxide
SDME	Single drop micro
	extraction
SFE	Supercritical fluid
	extraction
SPE	Solid phase extraction
SPME	Solid phase micro
	extraction
XRD	X-ray diffraction
	spectrometry
	specificity

#### 7. References

- [1] P.T. Anastas, Green chemistry and the role of analytical methodology development, Crit. Rev. Anal. Chem., 29 (1999)167-175. https://doi. org/10.1080/10408349891199356
- [2] P.T. Anastas, J.C. Warner, Green Chemistry, Theory and Practice. Oxford University Press, 135 pages, 1998. https://global.oup. com/academic/product/green-chemistry-9780198506980?cc=ca&lang=en&
- [3] M.Tobiszewski, J. Namieśnik, Direct chromatographic methods in the context of green analytical chemistry, TrAC Trends Anal. Chem., 35 (2012) 67–73. https://doi. org/10.1016/j.trac.2012.02.006
- [4] L.H. Keith, L.U. Gron, J.L. Young, Green analytical methodologies, Chem. Rev., 107 (2007) 2695–2708. https://doi. org/10.1021/cr068359e
- [5] S. Garrigues, S. Armenta, M. Guardia, Green strategies for decontamination of analytical wastes, TrAC Trends Anal. Chem., 29 (2010) 592–601. https://doi. org/10.1016/j.trac.2010.03.009
- [6] M. Sajid, J. Płotka-Wasylka, Green analytical chemistry metrics: A review, Talanta, 238 (2022) 123046. https://doi. org/10.1016/j.talanta.2021.123046
- [7] D.L. Rocha, R.R.P. Rocha, An environmentally friendly flow-based procedure with photo-induced oxidation for the spectrophotometric determination of chloride in urine and waters, Microchem. J., 108 (2013)193–197. https://doi. org/10.1016/j.microc.2012.10.020
- [8] R.C. Gosselin, D.M. Adcock, S.M. Bates, J. Douxfils, E.J. Favaloro, I. Gouin-Thibault, C. Guillermo, International council for standardization in haematology (ICSH) recommendations for laboratory measurement of direct oral anticoagulants, J. Thromb. Haemost., 118 (2018) 437–450. https://doi.org/10.1055/s-0038-1627480

- M. Castro Rodríguez, I. Rodriguez Garcia, R.N. Rodríguez Maecker, L. Pozo Morales, J.E. Oltra, A. Rosales Martinez, Cp2TiCl: An ideal reagent for green chemistry, Org. process Res. Dev., 21 (2017) 911-923. https://doi.org/10.1021/acs.oprd.7b00098
- B.M. Trost, The atom economy-a search for synthetic efficiency, Sci., 254 (1991) 1471–1477. https://doi.org/10.1126/ science.1962206
- [11] A. Gansäuer, M. Pierobon, H. Bluhm, Catalytic, highly regio- and chemoselective generation of radicals from epoxides: titanocene dichloride as an electron transfer catalyst in transition metal catalyzed radical reactions, Angew. Chem. Int. Ed., 37 (1998) 101–103. https://doi.org/10.1002/ (SICI)1521-3773(19980202)37:1/2<101::AID-ANIE101>3.0.CO;2-W
- [12] A. Gałuszka, Z. Migaszewski, J. Namieśnik, The 12 principles of green analytical chemistry and the significance mnemonic of green analytical practices, TrAC Trends Anal. Chem., 50 (2013) 78–84. https://doi. org/10.1016/j.trac.2013.04.010
- [13] M. Vian, C. Breil, L. Vernes, E. Chaabani,
  F. Chemat, Green Solvents for sample preparation in analytical chemistry,
  Curr. Opin. Green Sustain. Chem., 5 (2017) 44–48. https://doi.org/10.1016/j.
  cogsc.2017.03.010
- Q. Zhang, K.D.O. De Oliveira Vigier, S. Royer, F. Jérôme, Deep eutectic solvents: syntheses, properties and applications, Chem. Soc. Rev., 41 (2012) 7108–7146. https://doi.org/10.1039/c2cs35178a
- [15] J. Chen, Y. Li, X. Wang, W. Liu, Application of deep eutectic solvents in food analysis: A review, Molecules, 24 (2019) 4594. https://doi.org/10.3390/ molecules24244594
- [16] E. Destandau, E. Lesellier, Chromatographic properties of ethanol/ water mobile phases on silica based monolithic C18, Chromatographia, 68

(2008) 985–990. https://doi.org/10.1365/ s10337-008-0819-8

- [17] C.J. Welch, T. Brkovic, W. Schafer, X. Gong, Performance to burn? re-evaluating the choice of acetonitrile as the platform solvent for analytical HPLC, Green Chem., 11 (2009) 1232. https://doi.org/10.1039/ b906215g
- [18] V. Kienen, W.F. Costa, J.V. Visentainer, N.E. Souza, C.C. Oliveira, Development of a green chromatographic method for determination of fat-Soluble vitamins in food and pharmaceutical supplement, Talanta, 75 (2008) 141–146. https://doi. org/10.1016/j.talanta.2007.10.043
- [19] A.I. López-Lorente, F. Pena-Pereira, S. Pedersen-Bjergaard, V.G. Zuin, S.A. Ozkan, E. Psillakis, The ten principles of green sample preparation, TrAC Trends Anal. Chem., 148 (2022) 116530. https:// doi.org/10.1016/j.trac.2022.116530
- [20] Y. Gu, F. Jérôme, Glycerol as a sustainable solvent for green chemistry, Green Chem., 12 (2010) 1127. https://doi.org/10.1039/ c001628d
- [21] D.F. Aycock, Solvent applications of 2-Methyltetrahydrofuran in organometallic and biphasic reactions, Org. Process Res. Dev., 11 (2007) 156–159. https://doi. org/10.1021/op060155c
- [22] A. Breidbach, A greener, quick and comprehensive extraction approach for LC-MS of multiple mycotoxins, Toxins, 9 (2017) 91. https://doi.org/10.3390/ toxins9030091
- [23] E. Shawky, S.S. Takla, H.M. Hammoda, F.A. Darwish, Evaluation of the influence of green extraction solvents on the cytotoxic activities of crinum (amaryllidaeae) alkaloid extracts Using In-Vitro-In-silico approach, J. Ethnopharmacol., 227 (2018) 139–149. https://doi.org/ 10.1016/j. jep.2018.08.040
- [24] S. Bi, S. Hu, Z. Zhou, M. Kong, Y. Liu, The green and stable dissolving system

based on KOH/Urea for homogeneous chemical modification of chitosan, Int. J. Biol. Macromol., 120A (2018) 1103–1110. https://doi.org/10.1016/j. ijbiomac.2018.08.150

- [25] D. Prat, A. Wells, J. Hayler, H. Sneddon,
   C.R. McElroy, S. Abou-Shehada, P.J.
   Dunn, Green Chem., 18 (2016) 288-296.
   https://doi.org/10.1039/C5GC01008J
- [26] I. Pacheco-Fernández, V. Pino, Green solvents in analytical chemistry, Curr. Opin. Green Sustain. Chem., 18 (2019) 42–50. https://doi.org/10.1016/j. cogsc.2018.12.010
- [27] A.P. Abbott, G. Capper, D.L. Davies, R.K. Rasheed, V. Tambyrajah, Novel solvent properties of choline chloride/urea mixtures, Chem. Commun., 2003, 70-71. https://doi.org/10.1039/B210714G
- [28] D. Yang, Y. Wang, J. Peng, C. Xun, Y. Yang, A green deep eutectic solvents microextraction coupled with acid-base induction for extraction of trace phenolic compounds in large volume water samples, Ecotoxicol. Environ. Saf., 178 (2019) 130–136. https://doi.org/10.1016/j. ecoenv.2019.04.021
- [29] P. Makoś, G. Boczkaj, Deep eutectic solvents based highly efficient extractive desulfurization of fuels-eco-friendly approach, J. Mol. Liq., 296 (2019) 111916. https://doi.org/10.1016/j. molliq.2019.111916
- [30] S.N. Pedro, M.G. Freire, C.S.R. Freire, Deep eutectic solvents comprising active pharmaceutical ingredients in the development of drug delivery systems, Expert Opin. Drug Deliv. 16 (2019) 497– 506. https://doi.org/10.1080/17425247.20 19.1604680
- [31] F. Aydin, E. Yilmaz, M. Soylak, Vortex assisted deep eutectic solvent (DES)-emulsification liquid-liquid microextraction of trace curcumin in food and herbal tea samples, Food Chem., 243

(2018) 442–447. https://doi.org/10.1016/j. foodchem.2017.09.154

- [32] S. Somsubsin, K. Seebunrueng, S. Boonchiangma, S.Srijaranai, A simple solvent based microextraction for high performance liquid chromatographic analysis of aflatoxins in rice samples, Talanta, 176 (2018) 172–177. https://doi. org/10.1016/j.talanta.2017.08.028
- T. Li, Y. Song, Z. Dong, Y. Shi, J. Fan, [33] Hydrophobic deep eutectic solvents as extractants for the determination of bisphenols from food-contacted plastics by high performance liquid chromatography with fluorescence detection, J. Chromatogr. 1621 (2020)461087. https://doi. A, org/10.1016/j.chroma.2020.461087
- [34] G. Mastellone, I. Pacheco-Fernández, P. Rubiolo, V. Pino, C. Cagliero, Sustainable micro-scale extraction of bioactive phenolic compounds from vitis vinifera leaves with ionic liquid-based surfactants, Molecules, 25 (2020) 3072. https://doi. org/10.3390/molecules25133072
- [35] V.M. Paradiso, G. Squeo, A. Pasqualone, F. Caponio, C. Summo, An easy and green tool for olive oils labelling according to the contents of hydroxytyrosol and tyrosol derivatives: extraction with a natural deep eutectic solvent and direct spectrophotometric analysis, Food Chem., 291 (2019) 1–6. https://doi.org/10.1016/j. foodchem.2019.03.139
- [36] K. Fraige, R.D. Arrua, A.T. Sutton, C.S. Funari, Using natural deep eutectic solvents for the extraction of metabolites in byrsonima Intermedia leaves, J. Sep. Sci., 42 (2019) 591–597. https://doi.org/ 10.1002/jssc.201800905
- [37] W. Zhang, X. Liang, Headspace gas chromatography-mass spectrometry for volatile components analysis in ipomoea cairica (L.) sweet leaves: Natural deep eutectic solvents as green extraction and dilution matrix, Foods, 8 (2019) 205.

https://doi.org/10.3390/foods8060205

- [38] X. Liu, N. Fu, Q. Zhang, S. Cai, Q. Wang, D. Han, B. Tang, Green tailoring with water of choline chloride deep eutectic solvents for the extraction of polyphenols from palm samples, J. Chromatogr. Sci., 57 (2019) 272–278. https://doi.org/10.1093/ chromsci/bmy099
- [39] J. Ali, M. Tuzen, T.G. Kazi, Green and innovative technique develop for the determination of vanadium in different types of water and food samples by eutectic solvent extraction method, Food Chem., 306 (2020) 125638. https://doi. org/10.1016/j.foodchem.2019.125638
- [40] R. Ashouri, Dynamic and static removal of benzene from air based on taskspecific ionic liquid coated on MWCNTs by sorbent tube-headspace solid-phase extraction procedure, Int. J. Environ. Sci. Technol., 18 (2021) 2377-2390. https:// doi.org/10.1007/s13762-020-02995-4
- [41] N. Esmaeili, J. Rakhtshah, Ultrasound assisted-dispersive-modification solidphase extraction using task-specific ionic liquid immobilized on multiwall carbon nanotubes for speciation and determination mercury in water samples, Microchem. J., 154 (2020) 104632. https:// doi.org/10.1016/j.microc.2020.104632
- [42] D.A. Burns, E.W. Ciurczak, Handbook of Near-Infrared Analysis; 3rd edition, CRC Press, 834 pages, 2007. https://doi. org/10.1201/9781420007374
- [43] I.J.Wesley, N. Larsen, B.G. Osborne, J.H. Skerritt, Non-invasive monitoring of dough mixing by near infrared spectroscopy, J. Cereal Sci., 27 (1998) 61–69. https://doi. org/10.1006/jcrs.1997.0151
- [44] M. De la Guardia, S. Garrigues, Handbook of Green Analytical Chemistry; John Wiley & Sons, 2012. https://doi. org/10.1002/9781119940722
- [45] K.I. Hildrum, B.N. Nilsen, F. Westad, N.M. Wahlgren, In-line analysis of ground

beef using a diode array near infrared instrument on a conveyor belt, J. Near Infrared Spectrosc., 12 (2004) 367–376. https://doi.org/10.1255/jnirs.445

- [46] T. Fearn, P.I. Maris, An application of box– Jenkins methodology to the control of gluten addition in a flour mill, Applied statistics, J. R. Stat. Soc.: Series C, 40 (1991) 477–484. https://academic.oup.com/jrsssc/issue/40/3
- [47] M. Blanco, M.A. Romero, Near-infrared libraries in the pharmaceutical industry: A solution for identity confirmation, Analyst, 126 (2001) 2212–2217. https:// doi.org/10.1039/b105012p
- [48] Y.A. Woo, H.J. Kim, J. Cho, Identification of herbal medicines using pattern recognition techniques with near-infrared reflectance spectra, Microchem. J., 63 (1999) 61–70. https://doi.org/10.1006/ mchj.1999.1768
- [49] S.J. Mazivila, R.A.E. Castro, At-line green synthesis monitoring of new pharmaceutical Co-crystals lamivudine: theophylline polymorph I and II, quantification of polymorph I among Its APIs using FT-IR spectroscopy and MCR-ALS, J. Pharm. Biomed. Anal., 169 (2019) 235–244. https://doi.org/ 10.1016/j.jpba.2019.03.014
- [50] B. Weldegebreal, M. Redi-Abshiro, B.S. Chandravanshi, Development of new analytical methods for the determination of caffeine content in aqueous solution of green coffee beans, Chem. Cent. J., 11 (2017) 126. https://doi.org/10.1186/s13065-017-0356-3
- [51] R.L. McCreery, Raman spectroscopy for chemical analysis; John Wiley & Sons, 448 pages, 2005. https://www.wiley.com
- [52] A. Hernanz, J.F. Ruiz-López, J.M. Gavira-Vallejo, S. Martin, E. Gavrilenko, Raman microscopy of prehistoric rock paintings from the Hoz de vicente, minglanilla, cuenca, Spain, J. Raman Spectrosc., 41 (2010) 1394–1399. https://doi. org/10.1002/jrs.2582

- [53] N. Welter, U. Schüssler, W. Kiefer, J. Raman Spect.: An international journal for original work in all aspects of Raman spectroscopy, Wiley, 2007. https:// publications.rwth-aachen.de/record/35314
- [54] R. Legner, A. Wirtz, T. Koza, T. Tetzlaff, A. Nickisch-Hartfiel, M. Jaeger, Application of green analytical chemistry to a green chemistry process: magnetic resonance and Raman spectroscopic process monitoring of continuous ethanolic fermentation, Biotechnol. BioEng., 116 (2019) 2874– 2883. https://doi.org/10.1002/bit.27112
- [55] C.M. Molnar, C. Berghian-Grosan, D.A. Magdas, An optimized green preparation method for the successful application of Raman spectroscopy in honey studies, Talanta, 208 (2020) 120432. https://doi. org/10.1016/j.talanta.2019.120432
- [56] P.M. Nowak, R. Wietecha-Posłuszny, J. Pawliszyn, White analytical chemistry: An approach to reconcile the principles of green analytical chemistry and functionality, TrAC Trends Anal. Chem., 138 (2021) 116223. https://doi. org/10.1016/j.trac.2021.116223
- [57] C. Eliasson, N.A. Macleod, P. Matousek, Noninvasive detection of concealed liquid explosives using Raman spectroscopy, Anal. Chem., 79 (2007) 8185–8189. https://doi.org/10.1021/ac071383n
- [58] C. Eliasson, N.A. Macleod, P. Matousek, Non-Invasive detection of cocaine dissolved in beverages using displaced Raman spectroscopy, Anal. Chim. Acta, 607 (2008) 50–53. https://doi. org/10.1016/j.aca.2007.11.023
- [59] M. Kim, H. Chung, Y. Woo, M.S.A. Kemper, New non-invasive, quantitative Raman technique for the determination of an active ingredient in pharmaceutical liquids by direct measurement through a plastic bottle, Anal. Chim. Acta, 587 (2007) 200–207. https://doi.org/10.1016/j. aca.2007.01.062

- [60] A. Ncube, S. Mtetwa, M. Bukhari, G. Fiorentino, R. Passaro, Circular economy and green chemistry: The need for radical innovative approaches in the design for new products, Energies J., 16 (2023) 1752. https://doi.org/10.3390/en16041752
- [61] C. Eliasson, N.A. Macleod, L.C. Jayes, Non-Invasive quantitative assessment of the content of pharmaceutical capsules using transmission Raman spectroscopy, J. Pharm. Biomed. Anal., 47 (2008) 221–229. https://doi.org/10.1016/j.jpba.2008.01.013
- [62] A. Nordon, A. Mills, R.T. Burn, F.M. Cusick, D. Littlejohn, Comparison of non-invasive NIR and Raman spectrometries for determination of alcohol content of spirits, Anal. Chim. Acta, 548 (2005) 148–158. https://doi.org/10.1016/j. aca.2005.05.067
- [63] A.F. Zarandi, An immobilization of 2-(Aminomethyl) thiazole on multiwalled carbon nanotubes used for rapid extraction of manganese ions in hepatic patients, J. Pharm. Biomed. Anal.,240 (2024) 115941. https://doi.org/10.1016/j. jpba.2023.115941
- [64] M.M. Asl, Functionalized graphene oxide with bismuth and titanium oxide nanoparticles for efficiently removing formaldehyde from the air by photocatalytic degradation-adsorption process, J. Anal. Test., 7 (2023) 444-458. https://doi. org/10.1007/s41664-023-00272-0
- [65] E. Felici, C. Casado, C.C. Wang, J. Raba, M.R. Gomez, A green alternative method for analysis of ivermectin and moxidectin in environmental water samples using automatized preconcentration previous MEEKC, Electrophoresis, 37 (2016) 2977–2985. https://doi.org/10.1002/ elps.201600303
- [66] M.R. Abukhadra, A. Adlii, B.M. Bakry, Green fabrication of bentonite/chitosan@ cobalt oxide composite (BE/CH@Co) of enhanced adsorption and advanced

oxidation removal of congo red dye and Cr (VI) from water, Int. J. Biol. Macromol., 126 (2019) 402–413. https:// doi.org/10.1016/j.ijbiomac.2018.12.225

- [67] M.J. Nozal, J.L. Bernal, J.J. Jiménez, M.T. Martín, J. Bernal, Determination of azolic fungicides in wine by solid-phase extraction and high-performance liquid chromatography-atmospheric pressure chemical ionization-mass spectrometry, J. Chromatogr. A, 1076 (2005) 90–96. https:// doi.org/10.1016/j.chroma.2005.04.044
- [68] M. Yuan, S. Tong, S. Zhao, C.Q. Jia, Adsorption of polycyclic aromatic hydrocarbons from water using petroleum coke-derived porous carbon, J. Hazard. Mater, 181 (2010) 1115–1120. https://doi. org/ 10.1016/j.jhazmat.2010.05.130
- [69] L.F. Melo, C.H. Collins, I.C. Jardim, High-performance liquid chromatographic determination of pesticides in tomatoes using laboratory-made NH<sub>2</sub> and C<sub>18</sub> solidphase extraction materials, J. Chromatogr. A, 1073 (2005) 75–81. https://doi. org/10.1016/j.chroma.2004.09.043
- [70] M. Suárez, M.P. Romero, A. Macià, R.M. Valls, S. Fernández, Improved method for identifying and quantifying phenolic compounds olive oil and their metabolites in human plasma by microelution solid-phase extraction plate and liquid chromatography-tandem mass spectrometry, J. Chromatogr. B, Analyt. Technol. Biomed. Life Sci., 877 (2009) 4097-4106. https://doi.org/10.1016/j. jchromb.2009.10.025
- [71] S. Abuin, R. Codony, R. Compañó, M. Granados, M.D. Prat, Analysis of macrolide antibiotics in river water by solid-phase extraction and liquid chromatographymass spectrometry, J. Chromatogr. A, 1114 (2006) 73–81. https://doi.org/10.1016/j. chroma.2006.02.032
- [72] M.A. Andrade, F.M. Lanças, Determination of ochratoxin A in wine by packed in-tube

solid phase microextraction followed by high performance liquid chromatography coupled to tandem mass spectrometry, J. Chromatogr. A, 1493 (2017) 41–48. https:// doi.org/10.1016/j.chroma.2017.02.053

- [73] Y. M. M. A. Alsayadi, S. Arora, A review: Total vaporization solid-phase microextraction procedure in different matrixes, Anal. Methods Environ. Chem. J., 5 (2022) 80–102. https://doi. org/10.24200/amecj.v5.i03.190
- [74] G. Merola, S. Gentili, F. Tagliaro, T. Macchia, Determination of different recreational drugs in hair by HS-SPME and GC/MS, Anal. Bioanal. Chem., 397 (2010) 2987–2995. https://doi.org/10.1007/s00216-010-3882-6
- [75] R.L. Schacker, D.F. Moritz, M.S. Caro, L.A. Madureira, A.N. Dias, Study of viability of solid-phase microextraction, in vivo, in the extraction of microbial volatile organic compounds associated to the pigment production process by the Monascus fungus, in submerged fermentation, J. Braz. Chem. Soc., 28 (2017) 1113-1122. https://dx.doi. org/10.21577/0103-5053.20160269
- [76] F. Pena-Pereira, I. Lavilla, C. Bendicho, Miniaturized preconcentration methods based on liquid–liquid extraction and their application in inorganic ultratrace analysis and speciation: A review, Spectrochim. Acta B, 64 (2009) 1–15. https://doi. org/10.1016/j.sab.2008.10.042
- [77] M. Saraji, H. Ghambari, Comparison of three different dispersive liquid-liquid microextraction modes performed on their most usual configurations for the extraction of phenolic, neutral aromatic, and amino compounds from waters, J. Sep. Sci., 41 (2018) 3275–3284. https:// doi.org/10.1002/jssc.201800133
- [78] K.E. Rasmussen, S. Pedersen-Bjergaard,M. Krogh, H.G. Ugland, T. Grønhaug,Development of a simple In-vial

liquid-phase microextraction device for drug analysis compatible with capillary gas chromatography, capillary electrophoresis and high-performance liquid chromatography, J. Chromatogr. A, 873 (2000) 3–11. https://doi.org/10.1016/ s0021-9673(99)01163-2

- [79] M.Rezaee, Y. Assadi, M.R. Milani Hosseini, E. Aghaee, F. Ahmadi, S. Berijani, Determination of organic compounds in water using dispersive liquid-liquid microextraction, J. Chromatogr. A, 1116 (2006) 1–9. https://doi.org/10.1016/j. chroma.2006.03.007
- [80] M. Ramos Payán, M.A. Bello López, R. Fernández-Torres, Application of hollow fiber-based liquid-phase microextraction (HF-LPME) for the determination of acidic pharmaceuticals in wastewaters, Talanta, 82 (2010) 854–858. https://doi. org/10.1016/j.talanta.2010.05.022
- [81] I. Pacheco-Fernández, V. Pino, J.H. Ayala, A.M. Afonso, Guanidinium ionic liquid-based surfactants as low cytotoxic extractants: analytical performance in an in-situ dispersive liquid-liquid microextraction method for determining personal care products, J. Chromatogr. A, 1559 (2018) 102–111. https://doi. org/10.1016/j.chroma.2017.04.061
- [82] R. Shaikh, T.G. Kazi, H.I. Afridi, A. Akhtar, J.A. Baig, An environmental friendly enrichment method for microextraction of cadmium and lead in groundwater samples: impact on biological sample of children, Chemosphere, 237 (2019) 124444. https://doi.org/ 10.1016/j. chemosphere.2019.124444
- [83] A.Santana-Mayor, B. Socas-Rodríguez, R. Rodríguez-Ramos, M.A. Rodríguez-Delgado, A green and simple procedure based on deep eutectic solvents for the extraction of phthalates from beverages, Food Chem., 312 (2020) 125798. https:// doi.org/10.1016/j.foodchem.2019.125798

- [84] H. Shirkhanloo, A. Khaligh, H.Z. Mousavi, A. Rashidi, Ultrasound assisted-dispersiveionic liquid-micro-solid phase extraction based on carboxyl-functionalized nanoporous graphene for speciation and determination of trace inorganic and organic mercury species in water and caprine blood samples, Microchem. J. 130 (2017) 245–254. https://doi.org/10.1016/j. microc.2016.09.012
- [85] H.Shirkhanloo, M.Ghazaghi, H.Z. Mousavi, Cadmium determination in human biological samples based on trioctylmethyl ammonium thiosalicylate as a task-specific ionic liquid by dispersive liquid–liquid microextraction method, J. Mol. Liq., 218 (2016) 478–483. https:// doi.org/ 10.1016/j.molliq.2016.02.035
- [86] A. Faghihi-Zarandi, H. Shirkhanloo, C.A. Jamshidzadeh, New method for removal of hazardous toluene vapor from air based on ionic liquid-phase adsorbent, Int. J. Environ. Sci. Technol., 16 (2019) 2797–2808. https://doi.org/10.1007/s13762-018-1975-5
- [87] S. Teimoori, H. Shirkhanloo, A.H. Hassani, M. Panahi, N. Mansouri, An immobilization of aminopropyl trimethoxysilanephenanthrene carbaldehyde on graphene oxide for toluene extraction and separation in water samples, Chemosphere, 316 (2023) 137800. https://doi.org/10.1016/j. chemosphere.2023.137800
- [88] N. Esmaeili, J. Rakhtshah, E. Kolvari, H. Shirkhanloo, Ultrasound assisteddispersive-modification solid-phase extraction using task-specific ionic liquid immobilized on multiwall carbon nanotubes for speciation and determination mercury in water samples, Microchem. J., 154 (2020) 104632. https://doi. org/10.1016/j.microc.2020.104632
- [89] K. Shrivas, H.F. Wu, A rapid, sensitive and effective quantitative method for simultaneous determination of cationic

surfactantmixtures from river and municipal wastewater by direct combination of single-drop microextraction with AP-MALDI mass spectrometry, J. Mass Spectrom., 42 (2007) 1637–1644. https://doi.org/10.1002/jms.1266

- [90] A.L. Theis, A.J. Waldack, S.M. Hansen, M.A. Jeannot, Headspace solvent microextraction, Anal. Chem., 73 (2001) 5651–5654. https://doi.org/10.1021/ ac015569c
- [91] M. Ma, F.F. Cantwell, Solvent microextraction with simultaneous back-extraction for sample cleanup and preconcentration: preconcentration into a single microdrop, Anal. Chem., 71 (1999) 388–393. https://doi.org/10.1021/ ac9805899
- [92] J. Wang, Z. Du, W. Yu, S. Qu, Detection of seven pesticides in cucumbers using hollow fibre-based liquid-phase microextraction and ultra-high pressure liquid chromatography coupled to tandem mass spectrometry, J. Chromatogr. A, 1247 (2012) 10–17. https://doi.org/10.1016/j. chroma.2012.05.040
- [93] N. Arroyo-Manzanares, L. Gámiz-Gracia, A.M. García-Campaña, Determination of ochratoxin A in wines by capillary liquid chromatography with laser induced fluorescence detection using dispersive liquid-liquid microextraction, Food Chem., 135 (2012) 368–372. https://doi. org/ 10.1016/j.foodchem.2012.05.009
- [94] M.A. Farajzadeh, M.R.A. Mogaddam, H. Ghorbanpour, Development of a new microextraction method based on elevated temperature dispersive liquid–liquid microextraction for determination of triazole pesticides residues in honey by gas chromatography-nitrogen phosphorus detection. J. Chromatogr. A, 1347 (2014) 8–16. https://doi.org/10.1016/j. chroma.2014.04.067
- [95] X. Xu, F. Liang, J. Shi, X. Zhao, Z. Liu,

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L. Wu, Y. Song, H. Zhang, Z. Wang, Determination of hormones in milk by hollow fiber-Based stirring extraction bar liquid-liquid microextraction gas chromatography mass spectrometry, Anal. Chim. Acta, 790 (2013) 39–46. https://doi. org/10.1016/j.aca.2013.06.035

- [96] L.S. De Jager, A.R. Andrews, Development of a screening method for cocaine and cocaine metabolites in urine using solvent microextraction in conjunction with gas chromatography, J. Chromatogr. A, 911 (2001) 97–105. https://doi.org/10.1016/ s0021-9673(00)01256-5
- [97] L. De Jager, A.R.J. Andrews, Development of a screening method for cocaine and cocaine metabolites in saliva using hollow fiber membrane solvent microextraction, Anal. Chim. Acta, 458 (2002) 311– 320. https://doi.org/10.1016/S0003-2670(02)00063-6
- [98] B. Mostafavi, A. Feizbakhsh, E. Konoz, H. Faraji, Hydrophobic deep eutectic solvent based on centrifugation-free dispersive liquid–liquid microextraction for speciation of selenium in aqueous samples: one step closer to green analytical chemistry, Microchem. J., 148 (2019) 582–590. https://doi.org/10.1016/j. microc.2019.05.021
- [99] J Rakhtshah, Simultaneously speciation and determination of manganese (II) and (VII) ions in water, food, and vegetable samples based on immobilization of N-acetylcysteine on multi-walled carbon nanotubes, Food Chem., 389 (2022) 133124. https://doi.org/10.1016/j. foodchem.2022.133124
- [100] J. Ali, M. Tuzen, D. Citak, O.D. Uluozlu, Separation and preconcentration of trivalent chromium in environmental waters by using deep eutectic solvent with ultrasound-assisted based dispersive liquid-liquid microextraction method, J. Mol. Liq., 291 (2019) 111299. https://doi.

#### org/10.1016/j.molliq.2019.111299

- [101] A. Safavi, R. Ahmadi, A.M. Ramezani, Vortex-assistedliquid-liquidmicroextraction based on hydrophobic deep eutectic solvent for determination of malondialdehyde and formaldehyde by HPLC-UV approach, Microchem. J., 143 (2018) 166–174. https:// doi.org/10.1016/j.microc.2018.07.036
- [102] R. Malaei, A.M. Ramezani, G. Absalan, Analysis of malondialdehyde in human plasma samples through derivatization with 2,4-Dinitrophenylhydrazine by ultrasound-assisted dispersive liquid–liquid microextraction-GC-FID approach, J. Chromatogr. B, 1089 (2018) 60–69. https://doi.org/ 10.1016/j. jchromb.2018.05.001
- [103] A.D. Sánchez-Camargo, On-Line coupling of supercritical fluid extraction and chromatographic techniques, J. Sep. Sci., 40 (2017) 213–227. https://doi.org/ 10.1002/ jssc.201601040
- [104] A. Dispas, H. Jambo, S. André, E. Tyteca, P. Hubert, Supercritical fluid chromatography: A promising alternative to current bioanalytical techniques, Bioanalysis, 10 (2018) 107–124. https:// doi.org/10.4155/bio-2017-0211
- [105] M. Antunes-Ricardo, T. García-Cayuela, J.A. Mendiola, E. Ibañez, J.A. Gutiérrez-Uribe, M.P. Cano, D. Guajardo-Flores, Supercritical CO2 enzyme hydrolysis as a pretreatment for the release of isorhamnetin conjugates from opuntia Ficus-Indica (L.) mill, J. Supercrit. Fluids, 141 (2018) 21–28. https://doi. org/10.1016/j.supflu.2017.11.030
- [106] N. Maljurić, B.Otašević, J. Golubović, A new strategy for development of eco-friendly RP-HPLC method using corona charged aerosol detector and its application for simultaneous analysis of risperidone and its related impurities, Microchem. J., 153 (2020) 104394. https:// doi.org/10.1016/j.microc.2019.104394

- [107] B. Ma, R. Wang, H. Ni, K. Wang, Experimental study on harmless disposal of waste oil based mud using supercritical carbon dioxide extraction, Fuel, 252 (2019) 722–729. https:// doi.org/10.1016/j.fuel.2019.04.111
- [108] R.N. Lima, A.D.C. Santos, A.S. Ribeiro, L. Cardozo-Filho, Selective amides extraction and biological activity from piper hispidum leaves using the supercritical extraction, J. Supercrit. Fluids, 157 (2020) 104712. https://doi. org/10.1016/j.supflu.2019.104712
- [109] M. L. Cádiz-Gurrea, J. Lozano-Sánchez, A. Fernández-Ochoa, A. Segura-Carretero, Enhancing the yield of bioactive compounds from sclerocarya birrea bark by green extraction approaches, Molecules, 24 (2019) 966. https://doi.org/10.3390/ molecules24050966
- [110] T. Lefebvre, A. Talbi, S. Atwi-Ghaddar, E. Destandau, E. Lesellier, Development of an analytical method for chlorophyll pigments separation by reversed-phase supercritical fluid chromatography, J. Chromatogr. A, 1612 (2020) 460643. https://doi.org/10.1016/j.chroma.2019.460643
- [111] D. Yuvali, I. Narin, M. Soylak, E. Yilmaz, Green synthesis of magnetic carbon nanodot/graphene oxide hybrid material (Fe3O4@C-Nanodot@GO) for magnetic solid phase extraction of ibuprofen in human blood samples prior to HPLC-DAD determination, J. Pharm. Biomed. Anal., 179 (2020) 113001. https://doi. org/10.1016/j.jpba.2019.113001
- [112] B.W. Cue, J. Zhang, Green process chemistry in the pharmaceutical industry, Green Chem. Lett. Rev., 2 (2009) 193–211. https:// doi.org/10.1080/17518250903258150
- [113] M. Khanmohammadi, R. Nasiri, K. Ghasemi, S. Samani, A.B. Bagheri Garmarudi, Diagnosis of basal cell carcinoma by infrared spectroscopy of whole blood samples applying soft independent modeling class analogy, J. Cancer Res. Clin. Oncol., 133

(2007) 1001–1010. https://doi.org/10.1007/ s00432-007-0286-x

- [114] M. Jang, Application of portable X-Ray fluorescence (pXRF) for heavy metal analysis of soils in crop fields near Abandoned mine sites, Environ. Geochem. Health, 32 (2010) 207–216. https://doi. org/10.1007/s10653-009-9276-z
- [115] R.V. Bordiwala, Green synthesis and applications of metal nanoparticles, A review, Results Chem., 5 (2023)100832. https://doi.org/10.1016/j. rechem.2023.100832
- [116] A. Pinheiro, J.B. de Andrade, Development, validation and application of a SDME/ GC-FID methodology for the multiresidue determination of organophosphate and pyrethroid pesticides in water, Talanta, 79 (2009) 1354–1359. https://doi.org/10.1016/j. talanta.2009.06.002
- [117] S. Winters, R.M. Gendreau, Fourier transform infrared spectroscopy of protein adsorption from whole blood: II. Ex Vivo sheep studies, Appl.Spectrosc., 36(1982) 404–409. https://doi.org/10.1366/0003702824639592
- [118] R.M. Gendreau, S. Winters, R.L. Leininger, Fourier transform infrared spectroscopy of protein adsorption from whole blood: Ex vivo dog studies, Appl. Spectrosc., 35 (1981) 353–357. https:// doi.org/10.1366/0003702814732562c
- [119] M.M. Eskandari, B. Kalantari, Dispersive liquid-liquid microextraction based on task-specific ionic liquids for determination and speciation of chromium in human blood, J. Anal. Chem., 70 (2015) 1448-1455. https://doi.org/10.1134/ S1061934815120072
- [120] H.Z. Mousavi, Chromium speciation in human blood samples based on acetyl cysteine by dispersive liquid–liquid biomicroextraction and in-vitro evaluation of acetyl cysteine/cysteine for decreasing of hexavalent chromium concentration, J. Pharm. Biomed. Anal., 118 (2016) 1-8.
https://doi.org/10.1016/j.jpba.2015.10.018

- [121] M.K. Abbasabadi, Speciation of cadmium in human blood samples based on Fe3O4supported naphthalene-1-thiol-functionalized graphene oxide nanocomposite by ultrasound-assisted dispersive magnetic micro solid phase extraction, J. Pharm. Biomed. Anal., 189 (2020) 113455. https:// doi.org/10.1016/j.jpba.2020.113455
- [122] Z Karamzadeh, A novel biostructure sorbent based on CysSB/MetSB@ MWCNTs for separation of nickel and cobalt in biological samples by ultrasound assisted-dispersive ionic liquid-suspension solid phase micro extraction, J. Pharm. Biomed. Anal., 172 (2019) 285-294. https://doi.org/10.1016/j.jpba.2019.05.003
- [123] S. Davari, A lead analysis based on amine functionalized bimodal mesoporous silica nanoparticles in human biological samples by ultrasound assisted-ionic liquid trapmicro solid phase extraction, J. Pharm. Biomed. Anal., 157 (2018) 1-9. https:// doi.org/10.1016/j.jpba.2018.05.004
- [124] J. Sunarso, S. Ismadji, Decontamination of hazardous substances from solid matrices and liquids using supercritical fluids extraction: A review, J. Hazard. Mater., 161 (2009) 1–20. https://doi.org/10.1016/j. jhazmat.2008.03.069
- [125] C. Walgraeve, K. Demeestere, J. Dewulf, R. Zimmermann, H. Van Langenhove, OxygenatedPolycyclicaromatichydrocarbons in atmospheric particulate matter: molecular characterization andoccurrence, Atmos. Environ., 44(2010) 1831–1846. https://doi. org/10.1016/j.atmosenv.2009.12.004
- [126] M. B. H. Abadi Air pollution control: The evaluation of TerphApm@ MWCNTs as a novel heterogeneous sorbent for benzene removal from air by solid phase gas extraction, Arab. J. Chem., 13 (2020) 1741-1751. https:// doi.org/10.1016/j.arabjc.2018.01.011
- [127] M. Osanloo, M. Ghazaghi, H. Hassani Validation of a new and cost-effective

method for mercury vapor removal based on silver nanoparticles coating on micro glassy balls, Atm. Pollut. Res., 8 (2017) 359-365. https://doi.org/10.1016/j. apr.2016.10.004

- [128] M. Osanloo, Nobel method for toluene removal from air based on ionic liquid modified nano-graphen, Int. J. Occup. Hyg., 6 (2014) 1-5. https://ijoh.tums.ac.ir/ index.php/ijoh/article/view/89
- [129] H. Zavvar Mousavi, Determination of Hg in water and wastewater samples by CV-AAS following on-line preconcentration with silver trap, J. Anal. Chem., 65 (2010) 935-939. https://doi.org/10.1134/ S106193481009008X
- [130] A. Khaligh, F. Golbabaei, Z. Sadeghi, A. Vahid, A. Rashidi, On-line micro column preconcentration system based on amino bimodal mesoporous silica nanoparticles as a novel adsorbent for removal and speciation of chromium (III, VI) in environmental samples, J. Environ. Health Sci. Eng., 13 (2015) 1-12. https:// doi.org/10.1186/s40201-015-0205-z
- [131] Hosseini, A.F. Zarandi Nanographene oxide modified phenyl methanethiol nanomagnetic composite for rapid separation of aluminum in wastewaters, foods. and vegetable samples bv microwave dispersive magnetic micro solid-phase extraction, Food Chem., 347 (2021)129042. https://doi.org/10.1016/j. foodchem.2021.129042
- [132] J. Rakhtshah, A rapid extraction of toxic styrene from water and wastewater samples based on hydroxyethyl methylimidazolium tetrafluoroborate immobilized on MWCNTs by ultra-assisted dispersive cyclic conjugation-micro-solid phase extraction, Microchem. J., 170 (2021) 106759. https:// doi.org/10.1016/j.microc.2021.106759
- [133] H.H.LuoJun,X.Yang,J.Hu,Y.Liao, Method for detecting perfluorinated compounds by using solid phase microextraction with

liquid phase chromatography-tandem mass spectrum, Patent CN105974041A,2016. https://patents.google.com/patent/ CN105974041A/en?oq=CN105974041A

- [134] T.D.LiYanfei,L.Hong,L.Genrong, Method for detecting hogwash oil by combination of headspace solid-phase microextraction and gas chromatography mass spectrometry, CN Patents CN101852783A,2010. https://patents.google.com/patent/ CN101852783A/en?oq=+CN101852783A
- [135] X. Shaoping, C. Xiaoling. Method for analyzing volatile components of coffee beans using automated solid-phase microextraction technology, CN Patent CN108693290A, 2018. https://patents. google.com/patent/CN108693290A/ en?oq=CN108693290A