

## Cactus and Holm Oak Acorn for Efficient Textile Wastewater Treatment by Coagulation-Flocculation Process Optimization Using Box-Benhken Design

Abderrazzak Adachi<sup>1\*</sup>, Radouane Soujoud<sup>2</sup>, Faiçal El Ouadrhiri<sup>1</sup>, Moubchir Tarik<sup>3</sup>, Anouar Hmamou<sup>1</sup>, Nouredine Eloutassi<sup>1</sup>, Amal Lahkimi<sup>1</sup>

<sup>1</sup> Laboratory of Engineering, Electrochemistry, Modelling and Environment, Faculty of Sciences Dhar El Mehraz, Sidi Mohamed Ben Abdellah University, Fez, Morocco

<sup>2</sup> Data Science for Sustainable Earth Laboratory (Data4Earth), Faculty of Sciences and Technics, Sultan Moulay Slimane University, 23000 Beni Mellal, Morocco

<sup>3</sup> Polyvalent Team in Research and Development, Department of Biology, Poly Disciplinary Faculty, Sultan Moulay Slimane University, Beni-Mellal 23000, Morocco

\* Corresponding author's e-mail: [abderrazzak.adachi@usmba.ac.ma](mailto:abderrazzak.adachi@usmba.ac.ma)

### ABSTRACT

In this study, the effectiveness of using natural bio-coagulants and bio-flocculants to treat textile wastewater through the coagulation-flocculation method was examined. These bio-based agents have several advantages over chemical agents, including biodegradability, natural abundance, low toxicity, and low cost. A bio-coagulant (holm oak acorn (HOA)) and a bio-flocculant (cactus juice) were used to investigate the capacity for turbidity removal and decolorization of textile wastewater. The UV spectrophotometer was used to characterize the discharges before and after treatment, and the chemical oxygen demand (COD) and biological oxygen demand (BOD<sub>5</sub>) levels were calculated. Box-Behnken design (BBD) coupled with response surface methodology (RSM) were utilized to optimize the process and reduce turbidity and decolorization in textile wastewater. The obtained results show that under the optimal conditions (0.5 g·L<sup>-1</sup> of HOA, 15 mL·L<sup>-1</sup> of cactus juice, and a pH of 7), decolorization and turbidity removal were achieved at 69% and 90%, respectively. This study demonstrates the potential of using bio-coagulants and bio-flocculants in the treatment of textile wastewater.

**Keywords:** coagulation-flocculation, textile wastewater, Response surface Methodology, decolorization, cactus, holm oak acorn.

### INTRODUCTION

Textile wastewater generated by various phases of textile processing contains the contaminants that can be harmful to the natural environment if not properly treated [Pandey et al., 2007; Rezouki et al., 2021; Faouzi et al., 2023]. The negative impacts of such discharges include eutrophication, the death of aquatic life, including fish, plants, and animals, and groundwater contamination from contaminants leaching through the soil [Georgiou et al., 2003; Merzouk et al., 2010; Sanae et al., 2021]. One commonly used method for reducing the turbidity and color of liquid effluents

is the use of chemical coagulants and flocculants [Teh et al., 2016], However, these chemicals can leave residues in treated water that can affect human health [Saratale et al., 2011]. For instance, the coagulation-flocculation process, which uses materials mostly made of aluminum and produces much hazardous sludge, might lead to the issues with secondary environmental pollution [Flaten, 2001; Divakaran and Pillai, 2002]. Research has shown that residual ferric chloride and aluminum sulfate can also contribute to the development of Alzheimer's disease [Campbell, 2002]. Using the naturally occurring coagulants and flocculants from plants and trees may provide a

solution to these issues, as the concept of using specific plants for water treatment is not new. In recent years, numerous studies have been published on various plant materials that can serve as natural coagulants, such as *Moringa oleifera* [Liew et al., 2006], common bean (*Phaseolus vulgaris*) [Antov et al., 2010], and Nirmali seeds (*strychnos-potatorum*) [Pramod Kumar Raghuvanshi, Monikamadloi, Arvind J. Sharma, Hanumat S. Malviya, 2002]. Other natural coagulants and flocculants, known as organic coagulants and flocculants, can be derived from animals (such as *chitosan* and *crustaceans*) [Bakshi et al., 2020; Mohd Yunus et al., 2017], and microorganisms (such as *fungi*, *algae*, and *bacteria*) [Vijayaraghavan and Shanthakumar, 2016; Abu Hasan et al., 2021; Rebah et al., 2018]. The use of bio-coagulants could enable the development of a sustainable, eco-friendly water treatment method using locally available, inexpensive, and renewable plant resources [Choy et al., 2014]. Also, they can be produced, collected and processed locally, natural coagulants generate slurries that are not harmful to the environment or human health [Sharma et al., 2006], and can be treated biologically or used as a soil stabilizer due to their non-toxicity [Narasiah et al., 2002].

This study aimed to remove pollutants from textile wastewater using natural bio-coagulants and bio-flocculants. The study's objectives were: i) to evaluate the effectiveness of the coagulation-flocculation process in treating textile wastewater; ii) study the impact of cactus dose, holm oak acorn dose, cactus juice volume, and pH on the treatment of textile wastewater; iii) to optimize the removal of turbidity and decolorization using the Box-Behnken design in combination with response surface methodology (RSM). The findings of this research offer practical suggestion for enhancing the effectiveness of the coagulation-flocculation process in eliminating turbidity and decolorization from textile wastewater.

## MATERIALS AND METHODS

### Wastewater from the textile industry

In this study, the samples of textile wastewater were kindly provided by the MULTIWACH factory in Fez, located in the Sidi Ibrahim district. The samples were collected on March 28<sup>th</sup>, 2022 at the entrance of the factory's treatment plant,

which receives wastewater from a mixture of different stages and finishing units. Field measurements of some parameters were made (pH, conductivity, dissolved oxygen, and temperature), while others were measured in the laboratory using standard methods (Baird et al., n.d.). The parameters of the raw textile wastewater are listed in Table 1. The samples were collected in polypropylene bottles, transported chilled, and stored at 4 °C until analysis [Torres et al., 2019].

### Coagulation-flocculation experiment

In this work, only analytical reagent-quality chemicals were used. Model jar test equipment (Phipps-Bird, Richmond, USA) that supports 6 units of beakers with a total volume of 1000 ml was used for the coagulation-flocculation study. The beakers consisted of 250 ml of textile wastewater. A turbidimeter (AL450T) was used to quantify the turbidity, and the results were represented in nephelometric turbidity units (NTU). The pH of the textile wastewater was determined with a pH meter (HANNA-instruments-HI98128). In order to determine the absorbance, a UV-visible spectrophotometer was used (UV-205, HJD501). To destabilize the colloidal particles and break up the bio-coagulant, the process was initiated by mixing (520 rpm) for 1min. Then, 15 minutes of slow (30 rpm) mixing to contact the unsteady particles and make substantial flocs [Amran et al., 2021]. The supernatant was harvested 3 cm under the sample surface after 30 minutes of floc setting and filtered through Whatman filter paper to assess turbidity reduction and decolorization before further analyses. Room temperature ( $25 \pm 1$  °C) was used for all experiments. Coagulation-flocculation efficiency was evaluated using the following formula [Choudhary et al., 2019; Bhatia et al., 2007]:

$$\begin{aligned} \text{Turbidity removal (\%)} &= \\ &= \frac{T_0 - T_f}{T_0} \times 100 \end{aligned} \quad (1)$$

where:  $T_0$  – represents the starting turbidity, and  $T_f$  – represents the finished Turbidity.

The reduction in absorbance (A) at the wavelength of 662 nm, shown in Figure 4, was used to measure the decolorization of the textile wastewater. The following expression determined the percentage of decolorization [Hameed et al., 2016; Moghazy, 2017]:



Figure 1. Jar test equipment for Coagulation-Flocculation experiment

$$\begin{aligned} \text{Decolorization removal (\%)} &= \\ &= \frac{A_0 - A_f}{A_0} \times 100 \end{aligned} \quad (2)$$

The solution's initial and final absorbances are  $A_0$  and  $A_f$ , respectively. The results of each experiment were repeated in duplicate (precision is set at +5%), and the mean values are shown.

### Preparation of natural bio coagulants

#### *Preparation of bio coagulant and bio flocculant of cactus*

The cactus leaves were collected in the Ain Cheggag region of Fes city. The cactus cladodes were carefully washed with water to remove dirt. The *Opuntia* dry powder was prepared by cutting the cactus leaves into 1 cm wide strips and drying them at 60 °C for 24 hours [Barka et al., 2013]. The cactus was crushed in a granulator and sieved to 0.2 mm particle size. The cactus powder was stored in a container. The liquid

bio-flocculant used in the experiments is extracted from the cactus [Taa et al., 2016]. This cactus, which is widely distributed throughout Morocco 45 000 area/hectare [Nharingo and Moyo, 2016]. With care, the paddle is cleansed using tap water, then purified water, and finally it is gently chopped into little pieces using a kitchen knife. Before dissecting the pads, the peel has been removed [Adjeroud et al., 2015]. The cactus parts were ground using a household blender, 20 g of ground cactus parts were added to 200 ml of purified water, then stirred to be homogenized. Finally, the viscous aqueous extract was passed through a 1 mm screen to eliminate large particles. The end result is viscous, Water-miscible liquid that is polymeric and ionic in nature and green in color [Ennawaoui et al., 2022].

#### *Preparation of acorn coagulant from holm oak acorn*

The holm oak acorns used in all experiments were sourced from the Atlas Mountains (Imouzz-er Kandar, Morocco). The walnut pericarp was

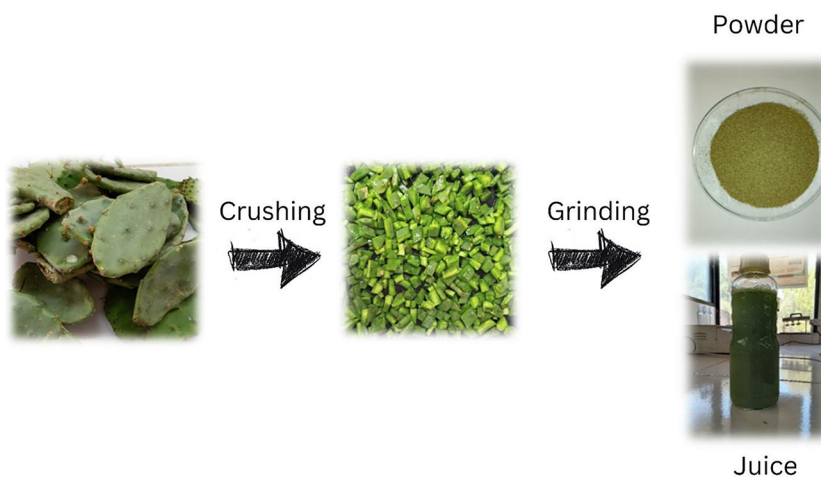


Figure 2. Powder and Juice cactus preparation



**Figure 3.** The holm oak acorn powder preparation

**Table 1.** List of textile wastewater properties for this study

Parameters	Raw wastewater mean ± S.D.	12h settled wastewater mean ± S.D.	Permissive levels (Morocco standard)
pH	5.6 ± 0.04	5.42 ± 0.38	9
Turbidity (NTU)	541 ± 50	473 ± 25	-
Absorbance	2.62 ± 0.1	2.42 ± 0.1	-
BOD <sub>5</sub> (mg·l <sup>-1</sup> )	515 ± 14,5	471 ± 10	500
COD (mg·l <sup>-1</sup> )	938 ± 40	837 ± 34	100
O <sub>2</sub> dissolves (mg·l <sup>-1</sup> )	4.69	4.01	-
Color	dark blue	dark blue	-
Conductivity (ms·cm <sup>-1</sup> )	2.15 ± 0.1	2.02 ± 0.02	2.7

manually scraped from the bark, cleaned with three washes of distilled water, and dried in a furnace for 24 hours at 60 °C. The dried material was ground using a grinder (SEB style, model M 8115) and filtered to remove any particles smaller than 0.2 mm. The resultant powder was oven-dried for an entire night before being kept in a closed container. Table 1 provides the analysis performed to identify the initial properties of the textile wastewater.

### Design of experiment

Design of experiments strategy is an effective method for understanding and optimizing turbidity removal and decolorization experimental parameters. It allows for a better understanding of their impact on the selected response, while significantly reducing the number of experiments and providing a clear understanding of the

**Table 2.** Model of the experimental design and response

Run	Factors			Responses			
	X <sub>1</sub> – biocoagulant (g·l <sup>-1</sup> )	X <sub>2</sub> – biofloculant (ml·l <sup>-1</sup> )	X <sub>3</sub> – pH	R <sub>1</sub> – decolorization removal (%)		R <sub>2</sub> – turbidity removal (%)	
				Experimental	Predicted	Experimental	Predicted
1	1.0	22.5	7	65.97	65.91	89.70	89.24
2	1.0	15.0	10	73.08	74.47	96.42	95.22
3	1.5	22.5	4	59.02	60.92	94.68	93.47
4	1.5	22.5	10	49.16	48.41	87.15	87.45
5	0.5	30.0	7	72.62	73.14	90.45	90.68
6	1.0	30.0	10	76.74	78.00	94.78	95.22
7	0.5	15.0	7	68.97	69.60	90.32	90.68
8	0.5	22.5	4	84.96	85.71	98.26	97.21
9	1.0	30.0	4	88.20	87.00	96.75	97.59
10	1.0	22.5	7	65.93	65.91	89.79	89.24
11	0.5	22.5	10	82.12	80.22	98.03	98.49
12	1.0	15.0	4	84.92	83.47	96.17	97.59
13	1.5	15.0	7	41.88	41.31	81.00	83.29
14	1.5	30.0	7	45.42	44.84	84.69	83.29
15	1.0	22.5	7	65.84	65.91	89.73	89.24

mechanism of the process [Huzir et al., 2019; Adachi et al., 2022]. In this study, a quadratic polynomial model was used to characterize the impact of three variables: bio- coagulant dosage ( $X_1$ ), bio-flocculant dose ( $X_2$ ), and textile wastewater pH ( $X_3$ ) on linear, interactive, and quadratic effects for three unrelated elements ( $q=3$ ):

$$Y_i = \beta_0 + \sum_{i=1}^n (\beta_i X_i) + \sum_{i=1}^n (\beta_{ii} X_i^2) + \sum_{i=1}^{n-1} \sum_{j=1}^n (\beta_{ij} X_i X_j) + \varepsilon \quad (3)$$

The Box-Behnken conceptual model was used to predict the response of an experiment, represented by  $Y_i$ , to certain predictor variables ( $X_i$  and  $X_j$ ). The model also considers the error, represented by  $\varepsilon$ , between the predicted and observed

responses. The model was created using Design-Expert software, and 15 experiments were conducted in a random order to account for potential hidden effects. The limitations of the factors used in the model are shown in Table 3. After the model was developed, optimization was performed to determine the optimal rate of turbidity and decolorization removal based on the standards and limitations outlined in Table 4. The optimization process employed the use of a desirability function (D) and a numerical optimization approach as described by [El Ouadrhiri et al., 2021].

## RESULT AND DISCUSSION

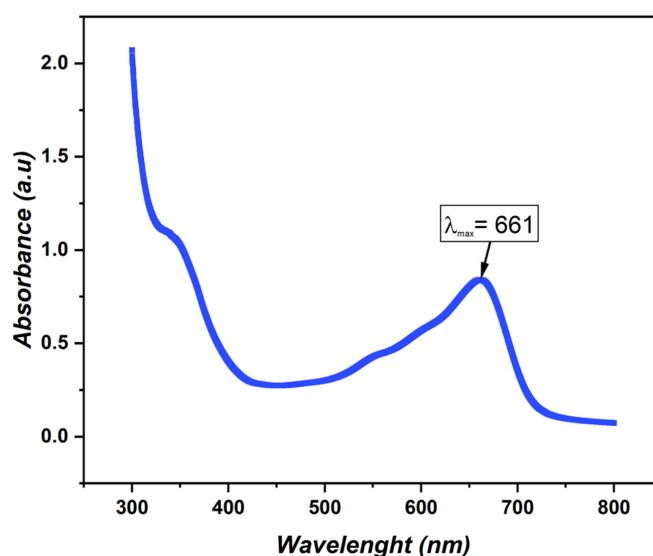
The UV-vis absorption spectra of the textile wastewater studied showed an absorption peak at 661 nm, as illustrated in Figure 4. The peak

**Table 3.** Factors limitations of the BBD plan

Factor	Name	Units	Minimum	Maximum	Coded low	Coded high
$X_1$	Biocoagulant	(g·l <sup>-1</sup> )	0.5000	1.50	-1 ↔ 0.50	+1 ↔ 1.50
$X_2$	Bioflocculant	(ml·l <sup>-1</sup> )	15.00	30.00	-1 ↔ 15.00	+1 ↔ 30.00
$X_3$	pH		4.00	10.00	-1 ↔ 4.00	+1 ↔ 10.00

**Table 4.** using the desirability function to enforce optimization restrictions

Name	Goal	Lower limit	Upper limit	Importance
$X_1$ – Biocoagulant ( g·l <sup>-1</sup> )	minimize	0.5	1.5	5
$X_2$ – Bioflocculant ( ml·l <sup>-1</sup> )	minimize	15	30	5
$X_3$ – pH	is target = 7	4	10	5
Turbidity removal (%)	maximize	80.9963	98.26	1
Decolorization removal (%)	maximize	41.8824	88.2039	5



**Figure 4.** The UV-vis absorption spectrum of textile effluents

was apparent (661 nm), showing the presence of the chromophore that gave the textile effluents their color.

### Cactus juice effect

Removing turbidity and decolorization are the most critical factors in improving the efficiency of the coagulation-flocculation process [Dkhissi et al., 2018]. Different quantities of cactus juice, ranging from 12 ml·l<sup>-1</sup> to 30 ml·l<sup>-1</sup>, were examined to determine the impact of cactus juice on the effectiveness of turbidity removal and decolorization of textile wastewater. The results are presented in Figure 5. When the volume of juice was increased from 12 ml·l<sup>-1</sup> to 24 ml·l<sup>-1</sup>, the removal efficiency of turbidity and decolorization improved until reaching 56.93% and 41.9%,

respectively. Increasing the volume of cactus juice from 24 ml·l<sup>-1</sup> to 30 ml·l<sup>-1</sup> results in a decrease in the efficiency of turbidity removal and decolorization from 21.8% to 17.93%, respectively. The higher the volume of cactus juice, the more efficient the aggregation and decantation of large polluting particles are. However, too much bio-flocculant, higher than the optimal value in the wastewater, would result in the dispersion of agglomerated particles and perturbed particle decantation [Mishra and Bajpai, 2005].

### Effect of pH on cactus juice

The effectiveness of natural coagulation as a treatment method is significantly affected by the hydrogen ion concentration, as noted in previous studies [Sethu et al., 2019; Zhao et al., 2021].

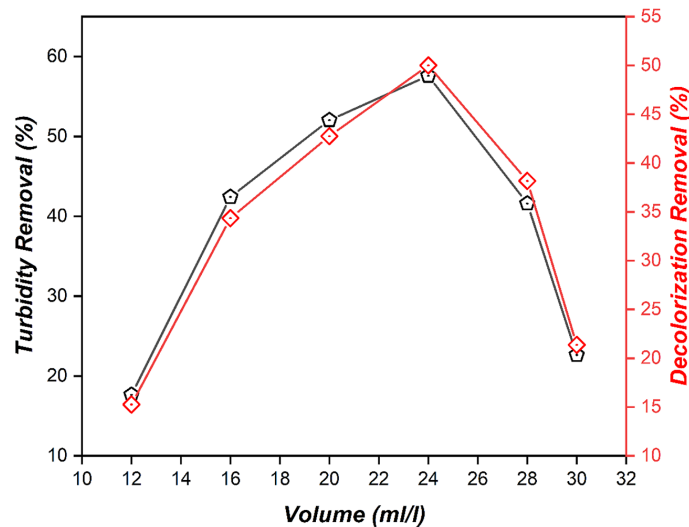


Figure 5. Effect of initial volume of cactus juice on the turbidity and decolorization removal

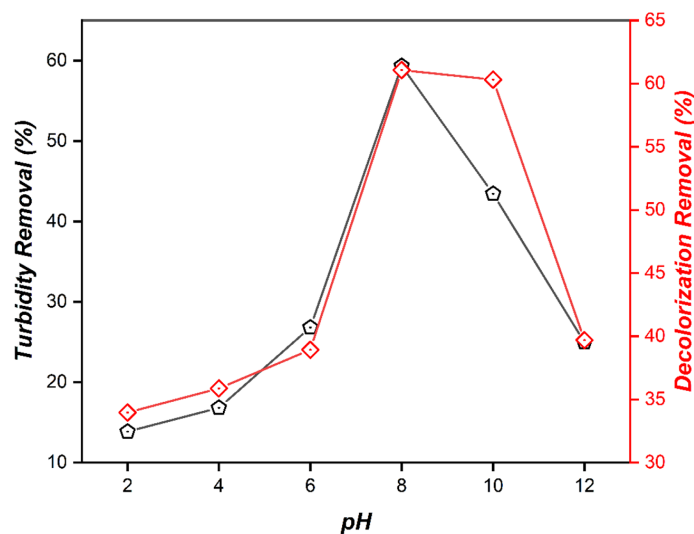


Figure 6. The pH effect of cactus juice on turbidity removal efficiency and decolorization

In this study, the bio-flocculant was tested under six different pH conditions (2, 4, 6, 8, 10, 12) [Daverey et al., 2019]. The pH was adjusted by adding drops of 0.1M NaOH and H<sub>2</sub>SO<sub>4</sub> solutions and was measured and verified using a pH meter. The optimal conditions for the cactus juice bio-flocculant are shown in Figure 6. At a pH of 8, the coagulation-flocculation process was found to be most successful, removing turbidity at a maximum rate of 56.74% and decolorization at a maximum rate of 57.25%. However, increasing the pH from 8 to 12 resulted in a decrease in both the turbidity removal rate and decolorization rate of 24.58% and 35.87%, respectively. These results align with previous research indicating that the optimal pH for bio-flocculant treatment is around 8 [ELsayed et al., 2020].

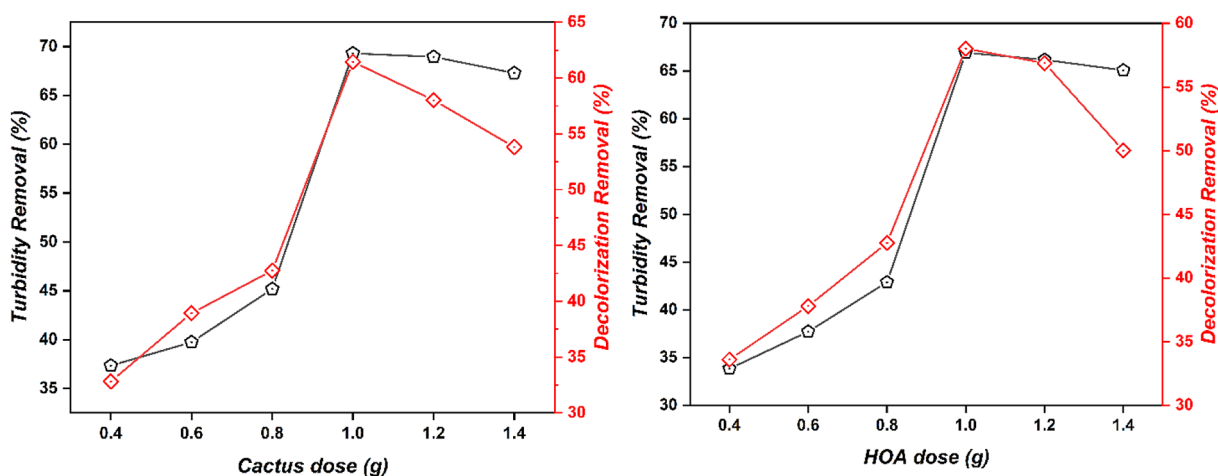
### Effect of the dose of the two bio coagulants

Figure 7 presents the results of measuring turbidity and absorbance at various bio-coagulant dosages, ranging from 0.4 g·L<sup>-1</sup> to 1.4 g·L<sup>-1</sup>, in textile wastewater with a pH of 5.6, to investigate the coagulation processes using holm oak acorn and cactus. Figure 7a illustrates that as the dose increases from 0.4 g·L<sup>-1</sup> to 1 g·L<sup>-1</sup>, the decolorization removal rate improves from 28.74% to 57.63%, and the turbidity increases from 36.59% to 68.57%. Conversely, when the dose is increased from 1 g·L<sup>-1</sup> to 1.4 g·L<sup>-1</sup>, the decolorization removal rate decreases from 57.63% to 50%, and the turbidity decreases from 68.57% to 66.54%. Figure 7b demonstrates similar trends, with an increase in the decolorization removal rate and turbidity from 29.77% to 54.19% and

33.08% to 66.17%, respectively, when the dose is increased from 0.4 g·L<sup>-1</sup> to 1 g·L<sup>-1</sup> and a decrease in the decolorization removal rate and turbidity from 54.19% to 46.18% and 66.17% to 64.32%, respectively, when the dose is increased from 1 g·L<sup>-1</sup> to 1.4 g·L<sup>-1</sup>.

### FTIR analysis

Figure 8a illustrates the infrared spectra of cactus powder (400–4000 cm<sup>-1</sup>). Chemical analysis of the powder reveals broad bands in the range of 3600–3000 cm<sup>-1</sup>, attributed to the presence of elongated O-H bands. The vibration of the asymmetric stretching group of CH<sub>2</sub> in aliphatic acids is what causes the band to be close to 2930 cm<sup>-1</sup> [Abdallah et al., 2016]. The band observed at 1621 cm<sup>-1</sup> is attributed to the stretching vibration of carboxylic groups. The stretching vibration of O-H groups in phenolic compounds is what causes the band at 1318. The peak at 1384 cm<sup>-1</sup> corresponds to the symmetric or non-symmetric valence vibration of pectin carboxyl radicals [Farinella et al., 2007]. Nitrogenous bioligands are responsible for absorption peaks in the spectral region below 800 cm<sup>-1</sup> [Barka et al., 2013]. These findings suggest that the cactus contains diverse organic functional groups. Figure 8b shows the infrared spectra of holm oak acorn powder, and chemical analysis reveals a broad and strong band in the range of 2500–3700 cm<sup>-1</sup>, indicative of O-H bonding groups. The bands at 2910 cm<sup>-1</sup> are likely attributed to aliphatic C-H groups. The band observed at approximately 1600 cm<sup>-1</sup> indicates the stretching of C-O bonds [Ghaedi et al., 2011].



**Figure 7.** Effect of bio-coagulant doses (a) cactus, (b) holm oak acorn on the efficiency of turbidity removal and decolorization, operating condition: pH = 6.5; V<sub>s</sub> = 250 ml

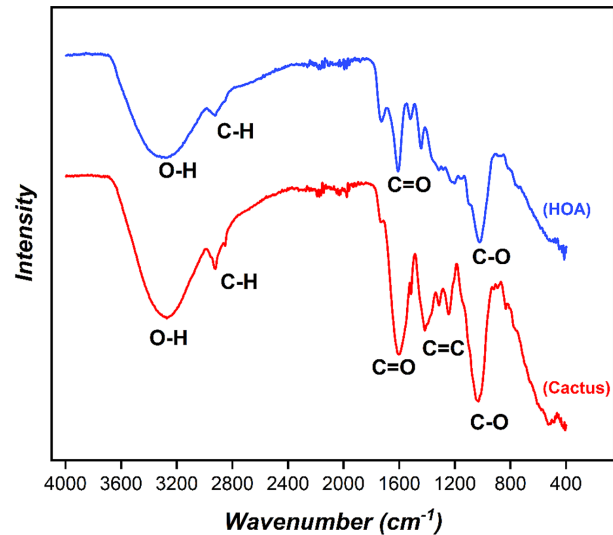


Figure 8. FTIR Spectrums of (a) the cactus powder and (b) holm oak acorn powder

### Scanning electron microscopy (SEM)

Scanning electron microscopy (SEM) was used to investigate the surface characteristics of cactus and holm oak acorn powder before and after treatment to understand the mechanism of coagulation-flocculation. Figure 9 presents an overview of the SEM microphotographs. As shown in Figure 9, the cactus and holm oak acorn powder

before treatment have a rough and porous structure (Figure 9a, 9b) [Subramonian et al., 2014]. The structured network has an irregular shape with a rough surface and irregular void spaces between them. As there are more adsorption sites with a higher density and larger surface area, this could improve the bridging process [Shak et Wu, 2014]. Following cactus and holm oak acorn treatment, the flocs formed showed close contact

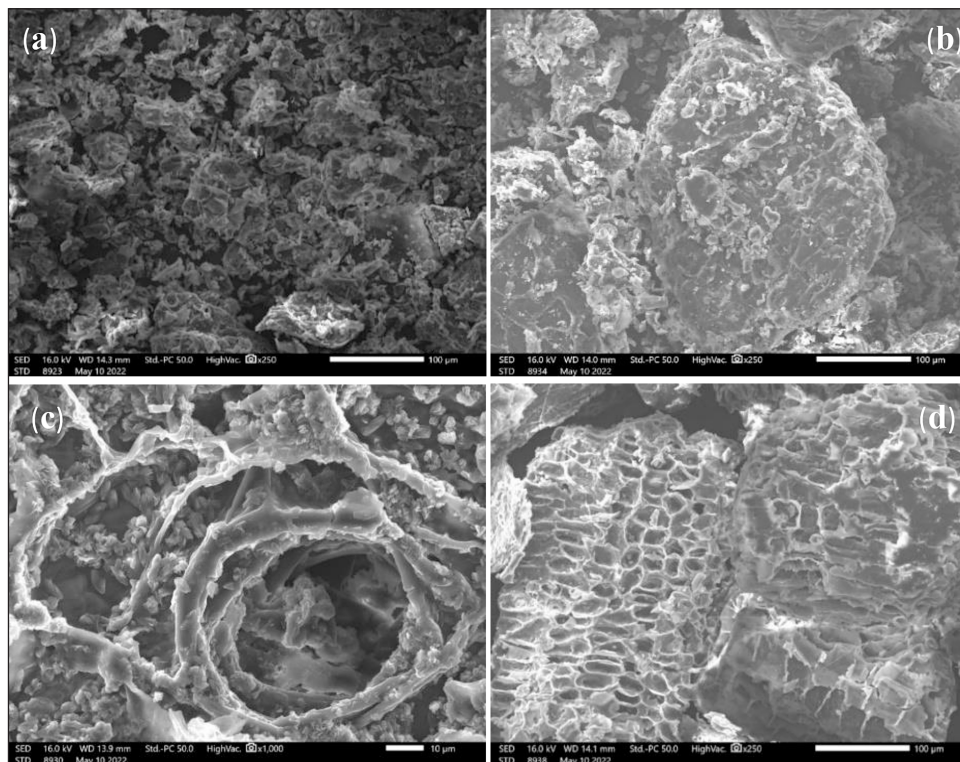


Figure 9. SEM images, (a) image of the cactus powder before treatment, (b) image of the holm oak acorn powder before treatment, (c) and (d) image of the cactus and holm oak acorn powder after the coagulation-flocculation process



between textile rejects and bio-coagulant (Figure 9c, 9d), indicating that the flocs are well-formed and compact. The principle of coagulation, particularly charge neutralization, involves the adsorption of coagulation-active components from cactus and holm oak acorn powder onto suspended particles of textile rejects [Muyibi et Evison, 1995]. The bio-coagulant surface, surface shape, pore size range, polarization, and functional groups associated with the bio-coagulant surface all play a role in the effectiveness of the coagulation process [Ali et al., 2016].

### Statistical analysis of the response

The Box-Behnken design (BBD) was employed in this study to investigate the association between turbidity and decolorization removal rate as well as the factors indicated in Table 2. To evaluate the model's accuracy and determine if it adequately represented the experimental results, lack of fit and analysis of variance were used [Mäkelä, 2017]. In addition, the multilinear regression coefficient  $R^2$ , the predicted coefficient ( $R^2_{\text{predicted}}$ ), and the adjusted coefficient ( $R^2_{\text{adjusted}}$ ) were employed to evaluate the predictive power of the models. Furthermore, the plot of predicted values vs actual values was employed to evaluate the distribution and normalization of the residuals. Upon analysis, it was found that the quadratic model fits the responses more closely than the linear model. On the one hand, the variance analysis results in Figures 5 and 6 show that the regression of turbidity and decolorization removal rate is highly significant with F values of 41.28 and 174.77 respectively, and P values of 0.0001. On the other hand, the lack of fit (F values of 1021.71

and 825.79 for turbidity and decolorization, respectively) indicates that they are significant. There is only a 0.01% chance that a lack of fit values as significant could occur due to noise. Additionally, the  $R^2_{\text{predicted}}$  of the model is 0.9582 and 0.99, indicating a strong correlation and good fit between the calculated and laboratory data. This is further supported by the distinction between  $R^2_{\text{adjusted}}$  (0.9350 and 0.9886) and  $R^2_{\text{predicted}}$  (0.8457 and 0.9432), which is less than 0.2, indicating a reasonable value.

The analytical results for the coded factors indicate that the dose of bio-coagulant ( $X_1$ ) and the pH ( $X_3$ ) of the solution has an important effect on the turbidity removal rate, as their P-value is less than 0.05. However, the P-value of the second term is higher than 0.1 indicating that it is not statistically significant. Therefore, the effect of the common points was considered in the following equation based on the coded factors to calculate the turbidity removal rate. Equation 1 presents the equation in terms of the actual factors:

$$R_1(\%) = 120.88 + 19.15X_1 - 10.32X_3 - 1.22(X_1 \times X_3) - 9.01X_1^2 + 0.79X_3^2 \quad (4)$$

The normal plot of residuals in Figure 10a shows that the distribution of residuals follows a linear line, supporting the assumption of uniform dispersion. In fact, the  $X_3$  term has a negative impact on the turbidity removal rate, with a value of -10.32. Plotting the predicted vs actual values further supports the validity of the experimental values. This leads to the conclusion that the Box-Behnken method is the most effective approach for the considered response. The ANOVA analysis of coded terms shows that the doses of

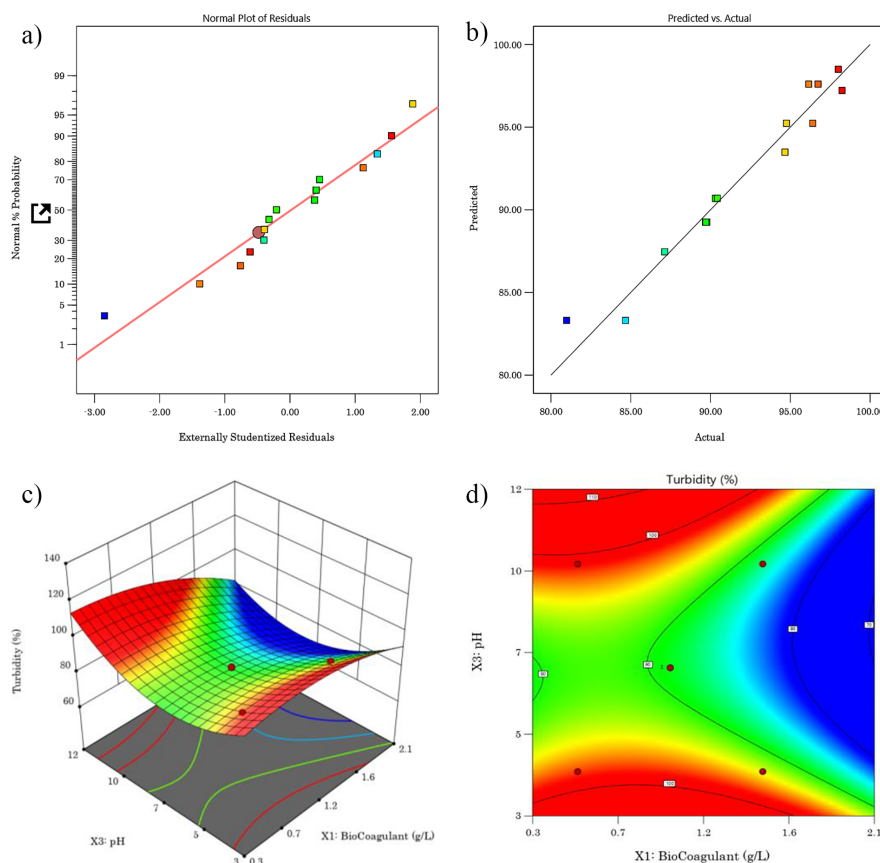
**Table 5.** ANOVA results of the turbidity removal

Source	Sum of squares	df	Mean square	F-value	p-value	
Model	352.96	5	70.59	41.28	< 0.0001	significant
$X_1$ – bio coagulant	109.16	1	109.16	63.83	< 0.0001	
$X_3$ – pH	11.25	1	11.25	6.58	0.0305	
$X_1X_3$	13.34	1	13.34	7.80	0.0210	
$X_1^2$	18.83	1	18.83	11.01	0.0090	
$X_3^2$	190.71	1	190.71	111.51	< 0.0001	
Residual	15.39	9	1.71			
Lack of fit	15.39	7	2.20	1021.71	0.0010	significant
Pure error	0.0043	2	0.0022			
Std. Dev.		1.31		$R^2$		0.9582
Mean		91.86		Adjusted $R^2$		0.9350
C.V. %		1.42		Predicted $R^2$		0.8457

bio coagulant ( $X_1$ ) and bio flocculant ( $X_2$ ) and the pH ( $X_3$ ) of the solution have a significant impact on the decolorization removal rate. Equation 5 shows the impact of the significant terms:

$$R_2 (\%) = 136.42 + 62.34X_1 - 1.06X_2 - 20.87X_3 - 1.17(X_1 \times X_3) - 41.22X_1^2 + 0.03X_2^2 + 1.47X_3^2 \quad (5)$$

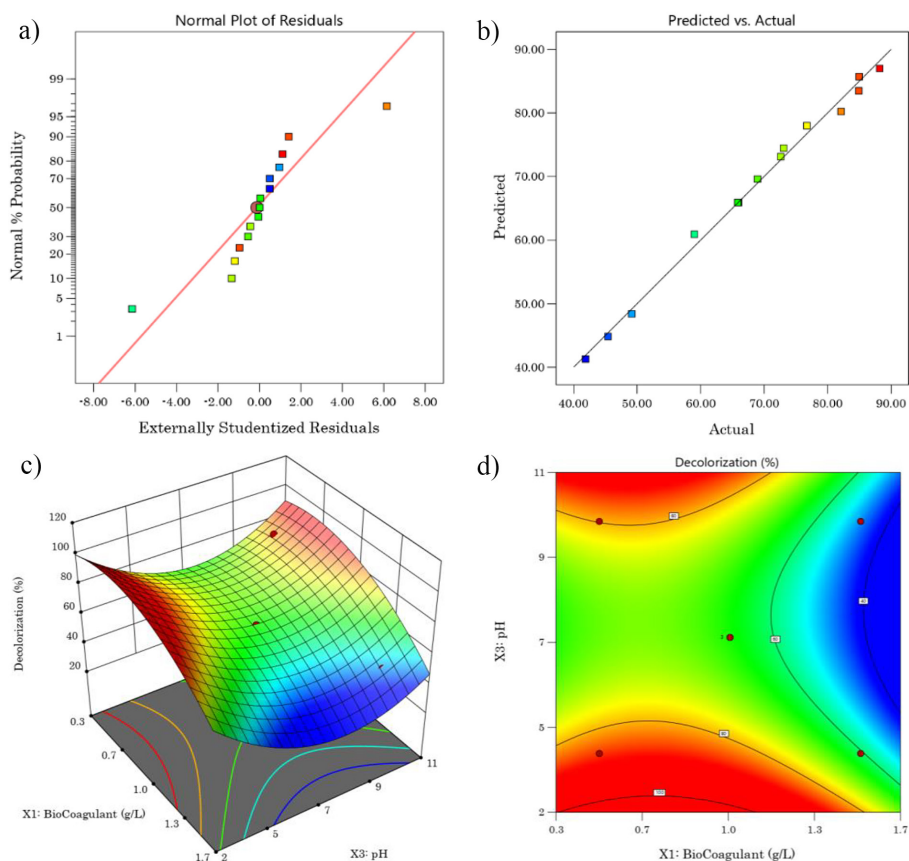
Figure 11a illustrates how the results are closely clustered around the diagonal line, indicating the validity and accuracy of the model's predictions. The plot of the 3D response surface generated from Equation 2 is shown in Figure 11d, demonstrating that the decolorization percentage increases when the bio-coagulant dosage is approximately  $1.3 \text{ g}\cdot\text{L}^{-1}$  and the pH is around 6.



**Figure 10.** (a) A normal probability plot (b) comparison of the predicted and real plot (c) contour plot and (d) 3D response surface of the turbidity response

**Table 6.** The ANOVA results of decolorization removal response

Source	Sum of squares	df	Mean square		p-value	
Model	2928.28	7	418.33	174.77	< 0.0001	significant
$X_1$ – Bio coagulant	1601.53	1	1601.53	669.09	< 0.0001	
$X_2$ – Bio flocculant	24.96	1	24.96	10.43	0.0145	
$X_3$ – pH	162.08	1	162.08	67.71	< 0.0001	
$X_1X_3$	12.34	1	12.34	5.15	0.0575	
$X_1^2$	392.11	1	392.11	163.81	< 0.0001	
$X_2^2$	9.62	1	9.62	4.02	0.0851	
$X_3^2$	643.94	1	643.94	269.03	< 0.0001	
Residual	16.76	7	2.39			
Lack of fit	16.75	5	3.35	825.79	0.0012	significant
Pure error	0.0081	2	0.0041			
Std. Dev.		1.55		R <sup>2</sup>		0.9943
Mean		68.32		Adjusted R <sup>2</sup>		0.9886
C.V. %		2.26		Predicted R <sup>2</sup>		0.9432



**Figure 11.** (a) A Normal probability plot, (b) comparison of the predicted and real plot, (c) contour plot and (d) 3D response surface plot of the decolorization response

### Numerical optimization of the models

One of the objectives of this study was to achieve maximum turbidity removal and decolorization according to the criteria listed in Table 4. To do this, a desirability function was used to determine a range of possible solutions, and selected the optimal solution from among these based on the highest D value that met the proportions listed in Table 4.

The Design-expert software was used to optimize process factors using the point prediction formula, and the optimized factors are presented in Table 7. To validate the RSM model based on the Box-Behnken approach under the chosen optimization conditions, the experiments were conducted to remove turbidity and decolorize real textile discharges. The experimental results showed that the percentage of turbidity removal

was 90.68%, and the decolorization percentage was 69.46%. The predicted turbidity removal and decolorization percentages were 90.34% and 69.46%, respectively, with errors of 0.14% for turbidity and 0.34% for decolorization. These results confirm that both equations 1 and 2 can accurately predict the removal rates of turbidity and decolorization from textile discharges.

### CONCLUSIONS

In conclusion, this study demonstrated the potential of coagulation-flocculation using holm oak acorn and cactus juice as bio-coagulant and bio-flocculant, respectively, for the treatment of effluent from textile wastewater. A Box-Behnken design and the response surface approach

**Table 7.** Verification tests in optimal conditions

Optimal conditions : $X_1=0.5$ (g.l <sup>-1</sup> ), $X_2=15$ (ml.l <sup>-1</sup> ), $X_3=7$ avec D=1			
Description	Predicted	Experimental	Error
Turbidity removal ( $R_1\%$ )	90.682	90.344	0.34
Decolorization removal ( $R_2\%$ )	69.605	69.461	0.14

were used to explore how bio-coagulant and bio-flocculant dosage, as well as textile wastewater pH, affected turbidity and decolorization. The results showed that the removal of turbidity and decolorization was influenced by the experimental factors studied, and the presence of oxygenated functional groups, as verified by chemical FTIR analysis, contributed to the coagulation-flocculation process. The optimal conditions for maximum turbidity and decolorization removal were found to be a bio-coagulant dosage of  $0.5 \text{ g}\cdot\text{L}^{-1}$ , a bio-flocculant dosage of  $15 \text{ mL}\cdot\text{L}^{-1}$ , and a textile wastewater pH of 7. The coagulation-flocculation process was also found to be improved by the BBD in combination with the RSM. On the basis of these findings, holm oak acorn and cactus can be considered promising bio-coagulants and bio-flocculants for the treatment of textile wastewater.

## REFERENCES

1. Abdallah, M., Hijazi, A., Hamieh, M., Alameh, M., Toufaily, J., Rammal, H. 2016. Étude de l'adsorption du Bleu de Méthylène sur un biomatériau à base de Cactus (Adsorption study of Methylene Blue on biomaterial using cactus). *Journal of Materials and Environmental Science*, 7, 4036–4048.
2. Abu Hasan, H., Alias, J., Arbain, F.N., Sheikh Abdullah, S.R., Kasan, N.A., Muhamad, M.H. 2021. Reusing sago mill effluent as a substrate for bio-based polymeric flocculant fermentation: Optimisation of operational conditions. *Environmental Technology and Innovation*, 23, 101704.
3. Adachi, A., El Ouadrhiri, F., Kara, M., El Manssouri, I., Assouguem, A., Almutairi, M.H., Bayram, R., Mohamed, H.R.H., Peluso, I., Eloutassi, N., Lakhimi, A. 2022. Decolorization and Degradation of Methyl Orange Azo Dye in Aqueous Solution by the Electro Fenton Process: Application of Optimization. *Catalysts*, 12, 1–12.
4. Adjeroud, N., Dahmoune, F., Merzouk, B., Leclerc, J.P., Madani, K. 2015. Improvement of electrocoagulation-electroflotation treatment of effluent by addition of *Opuntia ficus indica* pad juice. *Separation and Purification Technology*, 144, 168–176.
5. Ali, R.M., Hamad, H.A., Hussein, M.M., Malash, G.F. 2016. Potential of using green adsorbent of heavy metal removal from aqueous solutions: Adsorption kinetics, isotherm, thermodynamic, mechanism and economic analysis. *Ecological Engineering*, 91, 317–332.
6. Amran, A.H., Zaidi, N.S., Syafuddin, A., Zhan, L.Z., Bahrodin, M.B., Mehmood, M.A., Boopathy, R. 2021. Potential of carica papaya seed-derived bio-coagulant to remove turbidity from polluted water assessed through experimental and modeling-based study. *Applied Sciences*, 11(12), 5715.
7. Antov, M.G., Šćiban, M.B., Petrović, N.J. 2010. Proteins from common bean (*Phaseolus vulgaris*) seed as a natural coagulant for potential application in water turbidity removal. *Bioresource Technology*, 101, 2167–2172.
8. Baird, R.B., Eaton, A.D., Federation, W.E., n.d. *Standard Methods for the Examination of Water and Wastewater*, 23rd Edition Item Details.
9. Bakshi, P.S., Selvakumar, D., Kadirvelu, K., Kumar, N.S. 2020. Chitosan as an environment friendly biomaterial – a review on recent modifications and applications. *International Journal of Biological Macromolecules*, 150, 1072–1083.
10. Barka, N., Ouzaouit, K., Abdennouri, M., Makhfouk, M. El 2013a. Dried prickly pear cactus (*Opuntia ficus indica*) cladodes as a low-cost and eco-friendly biosorbent for dyes removal from aqueous solutions. *Journal of the Taiwan Institute of Chemical Engineers*, 44, 52–60.
11. Barka, N., Ouzaouit, K., Abdennouri, M., Makhfouk, M. El 2013b. Dried prickly pear cactus (*Opuntia ficus indica*) cladodes as a low-cost and eco-friendly biosorbent for dyes removal from aqueous solutions. *Journal of the Taiwan Institute of Chemical Engineers*, 44, 52–60.
12. Bhatia, S., Othman, Z., Ahmad, A.L. 2007. Pretreatment of palm oil mill effluent (POME) using *Moringa oleifera* seeds as natural coagulant. *Journal of Hazardous Materials*, 145, 120–126.
13. Campbell, A. 2002. The potential role of aluminium in Alzheimer's disease. *Nephrology Dialysis Transplantation*, 17, 17–20.
14. Choudhary, M., Ray, M.B., Neogi, S. 2019. Evaluation of the potential application of cactus (*Opuntia ficus-indica*) as a bio-coagulant for pre-treatment of oil sands process-affected water. *Separation and Purification Technology*, 209, 714–724.
15. Choy, S.Y., Prasad, K.M.N., Wu, T.Y., Raghunandan, M.E., Ramanan, R.N. 2014. Utilization of plant-based natural coagulants as future alternatives towards sustainable water clarification. *Journal of Environmental Sciences (China)*, 26, 2178–2189.
16. Daverey, A., Tiwari, N., Dutta, K. 2019. Utilization of extracts of *Musa paradisiaca* (banana) peels and *Dolichos lablab* (Indian bean) seeds as low-cost natural coagulants for turbidity removal from water. *Environmental Science and Pollution Research*, 26, 34177–34183.
17. Divakaran, R., Pillai, V.N.S. 2002. Flocculation of algae using chitosan. *Journal of Applied Phycology*, 14, 419–422.

18. Dkhissi, O., El Hakmaoui, A., Souabi, S., Chatoui, M., Jada, A., Akssira, M. 2018. Treatment of vegetable oil refinery wastewater by coagulation-flocculation process using the cactus as a bio-flocculant. *Journal of Materials and Environmental Science*, 9, 18–25.
19. El Ouadrhiri, F., Elyemni, M., Lahkimi, A., Lhasani, A., Chaouch, M., Taleb, M. 2021. Mesoporous Carbon from Optimized Date Stone Hydrochar by Catalytic Hydrothermal Carbonization Using Response Surface Methodology: Application to Dyes Adsorption. *International Journal of Chemical Engineering*, 2021.
20. ELSayed, E.M., Nour El-Den, A.A., Elkady, M.F., Zaatout, A.A. 2020. Comparison of coagulation performance using natural coagulants against traditional ones. *Separation Science and Technology (Philadelphia)*, 1–9.
21. Ennawaoui, A., Lalam, K., Harmen, Y., Morabit, A. El, Chhiti, Y., Chebak, A., Benssitel, M. 2022. Cactus Cladode Juice as Bioflocculant in the Flocculation-Thickening Process for Phosphate Washing Plant: A Comparative Study with Anionic Polyacrylamide. *Sustainability*, 14(13), 8054.
22. Farinella, N.V., Matos, G.D., Arruda, M.A.Z. 2007. Grape bagasse as a potential biosorbent of metals in effluent treatments. *Bioresource Technology*, 98, 1940–1946.
23. Faouzi, J., Rezouki, S., Bourhia, M., Moubchir, T., Abbou, M. B., Baammi, S., Lakimi, A. (2023). Assessment of impacts of industrial effluents on physico-chemical and microbiological qualities of irrigation water of the Fez River, Morocco. *Environmental Geochemistry and Health*, 1–14.
24. Flaten, T.P. 2001. Aluminium as a risk factor in Alzheimer's disease, with emphasis on drinking water. *Brain Research Bulletin*, 55, 187–196.
25. Georgiou, D., Aivazidis, A., Hatiras, J., Gimouhopoulos, K. 2003. Treatment of cotton textile wastewater using lime and ferrous sulfate. *Water Research*, 37, 2248–2250.
26. Ghaedi, M., Hossainian, H., Montazerzohori, M., Shokrollahi, A., Shojaipour, F., Soylak, M., Purkait, M.K. 2011. A novel acorn-based adsorbent for the removal of brilliant green. *Desalination*, 281, 226–233.
27. Hameed, Y.T., Idris, A., Hussain, S.A., Abdullah, N. 2016. A tannin-based agent for coagulation and flocculation of municipal wastewater: Chemical composition, performance assessment compared to Polyaluminum chloride, and application in a pilot plant. *Journal of Environmental Management*, 184, 494–503.
28. Huzir, N.M., Aziz, M.M.A., Ismail, S.B., Mahmood, N.A.N., Umor, N.A., Faua'ad Syed Muhammad, S.A. 2019. Optimization of coagulation-flocculation process for the palm oil mill effluent treatment by using rice husk ash. *Industrial Crops and Products*, 139, 111482.
29. Liew, A.G., Noor, M.J.M.M., Muyibi, S.A., Fugara, A.M.S., Muhammed, T.A., Iyuke, S.E. 2006. Surface water clarification using *M. oleifera* seeds. *International Journal of Environmental Studies*, 63, 211–219.
30. Mäkelä, M. 2017. Experimental design and response surface methodology in energy applications: A tutorial review. *Energy Conversion and Management*, 151, 630–640.
31. Merzouk, B., Madani, K., Sekki, A. 2010. Using electrocoagulation-electroflotation technology to treat synthetic solution and textile wastewater, two case studies. *Desalination*, 250, 573–577.
32. Mishra, A., Bajpai, M. 2005. Flocculation behaviour of model textile wastewater treated with a food grade polysaccharide. *Journal of Hazardous Materials*, 118, 213–217.
33. Moghazy, R.M. 2017. Preliminary study on the dye biosorption efficacy of raw and activated microalgal biomass, 7.
34. Mohd Yunus, F.H., Nasir, N.M., Wan Jusoh, H.H., Khatoon, H., Lam, S.S., Jusoh, A., 2017. Harvesting of microalgae (*Chlorella* sp.) from aquaculture bioflocs using an environmental-friendly chitosan-based bio-coagulant. *International Biodeterioration and Biodegradation*, 124, 243–249.
35. Muyibi, S.A., Evison, L.M. 1995. Optimizing physical parameters affecting coagulation of turbid water with *Moringa oleifera* seeds. *Water Research*, 29, 2689–2695.
36. Narasiah, K.S., Vogel, A., Kramadhati, N.N. 2002. Coagulation of turbid waters using *Moringa oleifera* seeds from two distinct sources. *Water Science and Technology: Water Supply*, 2, 83–88.
37. Nharingo, T., Moyo, M. 2016. Application of *Opuntia ficus-indica* in bioremediation of wastewaters. A critical review. *Journal of Environmental Management*, 166, 55–72.
38. Pandey, A., Singh, P., Iyengar, L. 2007. Bacterial decolorization and degradation of azo dyes. *International Biodeterioration and Biodegradation*, 59, 73–84.
39. Raghuvanshi, P.K., Madloi, M., Sharma, A.J., Malviya, H.S., Chaudhari, S. 2002. Agrobased Materials as Coagulant Aid. *Water Quality Research*, 37, 745–756.
40. Rebah, F.B., Mnif, W., Siddeeg, S.M. 2018. Microbial flocculants as an alternative to synthetic polymers for wastewater treatment: A review. *Symmetry*, 10, 1–19.
41. Rezouki, S., Allali, A., Touati, N., Mansouri, D., Eloutassi, N., Fadli, M. 2021. Spatio-temporal evolution of the physico-chemical parameters of the Inaouen wadi and its tributaries. *Moroccan Journal of Chemistry*, 9(3), 9–3.

42. Sanae, R., Aimad, A., Karim, B., Jamaa, H., Nouredine, E., Mohamed, F. 2021. The Impact of Physicochemical Parameters and Heavy Metals on the Biodiversity of Benthic Macrofauna in the Inaouene Wadi (Taza, North East Morocco). *Journal of Ecological Engineering*, 22(7), 231–241.
43. Saratale, R.G., Saratale, G.D., Chang, J.S., Govindwar, S.P. 2011. Bacterial decolorization and degradation of azo dyes: A review. *Journal of the Taiwan Institute of Chemical Engineers*, 42, 138–157.
44. Sethu, V., Selvarajoo, A., Chee Wei, L., Ganesan, P., See Lim, G., Xin Yuan, M. 2019. Progress in Energy and Environment *Opuntia cactus* as a novel bio-coagulant for the treatment of Palm Oil Mill Effluent (POME). *Progress in Energy and Environment*, 9, 11–26.
45. Shak, K.P.Y., Wu, T.Y. 2014. Coagulation-flocculation treatment of high-strength agro-industrial wastewater using natural *Cassia obtusifolia* seed gum: Treatment efficiencies and flocs characterization. *Chemical Engineering Journal*, 256, 293–305.
46. Sharma, B.R., Dhuldhoya, N.C., Merchant, U.C. 2006. Flocculants - An ecofriendly approach. *Journal of Polymers and the Environment*, 14, 195–202.
47. Subramonian, W., Wu, T.Y., Chai, S.P. 2014. A comprehensive study on coagulant performance and floc characterization of natural *Cassia obtusifolia* seed gum in treatment of raw pulp and paper mill effluent. *Industrial Crops and Products*, 61, 317–324.
48. Taa, N., Benyahya, M., Chaouch, M. 2016. Using a bio-flocculent in the process of coagulation flocculation for optimizing the chromium removal from the polluted water. *Journal of Materials and Environmental Science*, 7, 1581–1588.
49. Teh, C.Y., Budiman, P.M., Shak, K.P.Y., Wu, T.Y. 2016. Recent Advancement of Coagulation-Flocculation and Its Application in Wastewater Treatment. *Industrial and Engineering Chemistry Research*, 55, 4363–4389.
50. Torres, N.H., Souza, B.S., Ferreira, L.F.R., Lima, Á.S., dos Santos, G.N., Cavalcanti, E.B. 2019. Real textile effluents treatment using coagulation/flocculation followed by electrochemical oxidation process and ecotoxicological assessment. *Chemosphere*, 236.
51. Vijayaraghavan, G., Shanthakumar, S. 2016. Performance study on algal alginate as natural coagulant for the removal of Congo red dye. *Desalination and Water Treatment*, 57, 6384–6392.
52. Zhao, C., Zhou, J., Yan, Y., Yang, L., Xing, G., Li, H., Wu, P., Wang, M., Zheng, H. 2021. Application of coagulation/flocculation in oily wastewater treatment: A review. *Science of the Total Environment*, 765, 142795.