



## New Vegetal Biopolymeric Flocculant: A Degradation and Flocculation Study

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**Abstract:** Polyacrylamide (PAM) is known to pose a hazard to human health. A newly explored plant-based flocculant, malva nut gum (MNG) is hypothesized to treat water and as a green material. This study investigates the biodegradation, thermal degradation and photodegradation of MNG and its flocculation capability. The results show MNG has a high biodegradation rate and low oxygen consumption rate. MNG biodegraded at Day-17, whilst, PAM did not show completion of biodegradation by the end of the experiment. Thermal degradation analysis shows MNG has a few decomposition stages and ash content. The photodegradation analysis shows photochemical changes in the structure of MNG and PAM. Factorial experiments show pH, concentration of cation and the concentration of MNG were the influential factors in coagulation-flocculation process. Optimization study observed 97% turbidity reduction at pH 5.77, the concentration of cation at 0.05 mM and the concentration of MNG at 0.42 mg/L. Conclusively, MNG operates effectively at near neutral pH and at low concentration. Such results strongly suggest that MNG is a suitable candidate, which could be used to replace PAM, as flocculant in water treatment.

**Abbreviation:** Malva nut gum (MNG) • Polyacrylamide (PAM) • Response surface methodology (RSM)

**Key words:** Biopolymeric flocculant • Malva nut gum • Degradation • Response surface methodology • Turbid water treatment

### INTRODUCTION

Previous studies revealed that there are some environmental and health concerns of using alum salts and polyacrylamide in water and wastewater treatment, i.e. Alzheimer's disease, the residual and the derivatives of PAM i.e. its monomers, other reactants and reaction by-products [1] are neurotoxic and carcinogenic. Therefore, natural polysaccharide extracted from plants or animals are workable alternatives to synthetic polyelectrolytes like PAM. This is due to degradable properties, no risk to human health and has a wide effective dosage range for flocculation of various colloidal suspensions. Moreover, the need for readily degradable and environmental-friendly materials in water treatment process is essential. Owing to this fact, plant-based coagulant and flocculant have received increasing attention from worldwide researchers recently. The aims in this study include (i) prepare a new plant-based flocculant, (ii) study the degradation properties by

biodegradation, thermal degradation and photo degradation, (iii) investigate the significant factors i.e. pH, concentration of cation, concentration of polymer and its interaction in screening process and (iv) optimize the maximum turbidity reduction using response surface methodology (RSM). The findings bring significant impact on the use of green material in water treatment plants which may increase the economic contribution to the native grown country.

The selected potential plant-based flocculant is extracted from malva nut. Malva nut is the seed of the *Scaphium scaphigerum* tree. The properties of malva nut gum (MNG) contains the key properties as a flocculant where it has high molecular weight [2] and contains properties of uronic acid [3]. These properties influence bridging or adsorption process during flocculation. It is noteworthy that the molecular weight and intrinsic viscosity of MNG are much higher than most polysaccharide gums that are currently available on the market, i.e. guar gum, locust bean gum and pectin [4].

Malva nuts are cheap, easily available on the market and their interesting properties could establish it to become a new renewable, sustainable and degradable flocculant.

Biodegradation study is based upon the degradation of polymers in environment in presence of microorganism. The rate of biodegradation depends on many factors such as the environment (including surrounding temperature), population of microorganism and etc. [5]. However, biodegradation requires a long period to determine the degradability, thus, thermal degradation becomes an important method to investigate [6]. It is a fast, sensitive, uniform and reproducible technique. Another useful photodegradation is a degradation method is oxidative degradation where depolymerization occurs in presence of ultraviolet (UV). The action of UV radiation reduces the molar mass and changes the structure of polymer. This method is a convenient, time saving and cost efficient. It may replace the expensive and time consuming enzymatic or chemical methods that are being currently used [7].

## MATERIALS AND METHODS

**Preparation of MNG:** Simplified extraction process was performed in accordance to Somboonpanyakul *et al.* [4]. The dried malva nut was treated with ethanol at 85°C for 1 hour. Subsequently, the pH of the slurry was adjusted to pH 4.5 and filtered through a silk-screen filter cloth prior to precipitation with 95% ethanol for 1 hour. The gummy portion was washed with the ethanol twice. Lastly, the gum was air dried.

**Biodegradation Study:** Biodegradation study was carried out in accordance to Norli *et al.* [8]. Microcrystalline cellulose was used as a reference test. Inoculum solution was obtained by vermin-compost waste extraction. Often, before day 5, it is the adaptation of the inoculum to the test substance. Typically, degradation occurs between day 5-12 and ends at day 15 in an activated sludge system in a water treatment plant [9]. In this study, the experiment has a duration of 18 days with an extension of 3 days for further observation.

**Thermal Degradation Study:** The decomposition of MNG was performed by using thermogravimetric analysis (TGA). This study was carried out using Mettler Toledo, TGA/SDTA851e OMNIC. The weight of the samples used ranged from 10–12 mg. The heating rate was 20°C/min from 30 to 800°C under oxygen atmosphere at 30 mL/min. The decomposition temperature of polymer and ash volume produced were attained at the end of the analysis.

**Photodegradation Study:** Photodegradation was carried out by exposing the sample to natural environment and natural solar UV for 30 days in the month of August (2012). The weather was sunny, average temperature at around 23 to 32°C and accompanied with isolated rain for about 3 days. The degradation was examined using FTIR. This method was modified from dos Santos *et al.* [10] where the sample was exposed to natural solar light instead of artificial UV light. FTIR spectra were obtained using FTIR in mid infrared region (4000–400 cm<sup>-1</sup>) to determine the functional group of the flocculants by conventional method using potassium bromide (KBr) pallet [11]. The sample pallet was analyzed using Perkin Elmer, System 2000 FTIR.

**Turbid Water Treatment:** Jar test is used for the coagulation-flocculation process. Kaolin suspension is prepared to simulate turbid wastewater. The initial turbidity is around 400 NTU. In this study, ferric chloride was chosen as cation that acts as a coagulant. From here onwards, it is noted as Fe. The mixing procedures were performed with rapid mixing at 150 rpm for 3 minutes and subsequently, slow mixing at 30 rpm for 20 minutes. Then, it was left standing still for 5 minutes. A 10 mL of supernatant was withdrawn and continued with a turbidity examination using HACH Turbidimeter 2100Q.

**Assay of Turbidity Reduction:** The capability of flocculant is measured in the ability to reduce turbidity in synthetic wastewater. The experimental procedure was carried out according to Standard Methods, Method 2130 B [12]. Percentage of turbidity reduction was calculated as in Eq. (1):

$$\text{Turbidity reduction, \%} = \frac{A - A_0}{A} \quad (1)$$

where,

- $A_0$  was turbidity of the control sample before treatment, NTU
- $A$  was turbidity of the sample after treatment, NTU

**Design of Experiment:** The screening of factors determines influential factors to the process within the design boundary. Therefore, a screening experiment was carried out to select significant factors. Two-level full factorial designs were used with three variables or factors (2<sup>3</sup> factorial design), namely, pH, concentration of cation and concentration of flocculant which are denoted as factors A, B, C, respectively. The design boundary for pH is between 3 and 9, concentration of cation is between

0.01 mM to 0.1 mM and lastly, concentration of MNG is between 0.05 to 0.5 mg/L. Optimization experiment was carried out using face-centred design and turbidity reduction was the response output. Design Expert statistical software was used for the analysis.

## RESULTS AND DISCUSSION

**Biodegradation Study:** The biodegradation study was carried out to analyze the biodegradability of MNG and PAM. Fig. 1 shows the degradation of reference test (using microcrystalline), MNG and PAM.

Previous study on pectin (another emerging green flocculant) was degraded at day 14 [8]. As illustrated in Fig. 1, reference sample completed degradation process at day 10. MNG requires less oxygen throughout biodegradation process and completed degradation process at day 17. However, PAM showed drastic consumption of oxygen and did not exhibit any behaviour for the completion of biodegradation at the end of experiment. Furthermore, the amount of oxygen required for PAM degradation is much higher than microcrystalline cellulose and MNG. During degradation process, microorganisms use the substances as food and to grow and at the same time consume oxygen available. Conclusively, natural polysaccharide has better biodegradation capability than PAM.

**Thermal-degradation Study:** Decomposition study investigates the fate of polymer and emissions to air during degradation in the environment thermally.

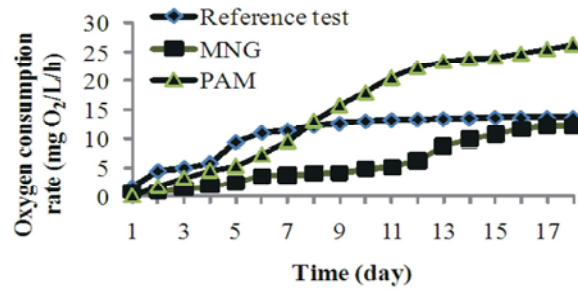


Fig. 1: Oxygen consumption by microorganisms to degrade MNG, PAM and reference test (microcrystalline cellulose)

During thermal degradation, the weight loss is mainly due to the oxidation process whereby the molecular bonds of a polymer are attacked by oxygen molecules and broken down [13]. The TGA curve of MNG illustrated in Fig 2.

The TGA result is in accordance with DSC result where there is a degradation step at an onset temperature of 33°C. Nonetheless, the analysis is set to end at 300°C which is the limit of the instrument. Therefore, TGA is carried out to determine further thermal degradation behaviour.

The curve consists of four sections: (1) a weight loss of 18.5% from 29.63°C to 213.38°C, (2) tremendous weight loss of 41.12% from 213.38°C to 337.48°C, (3) weight loss of 10.42% from 337.48°C to 531.89°C, (4) weight loss 21.43% from 531.89°C to 799.12°C. The decomposition of MNG is completed at temperature 800°C where only 8.53% residue was left.

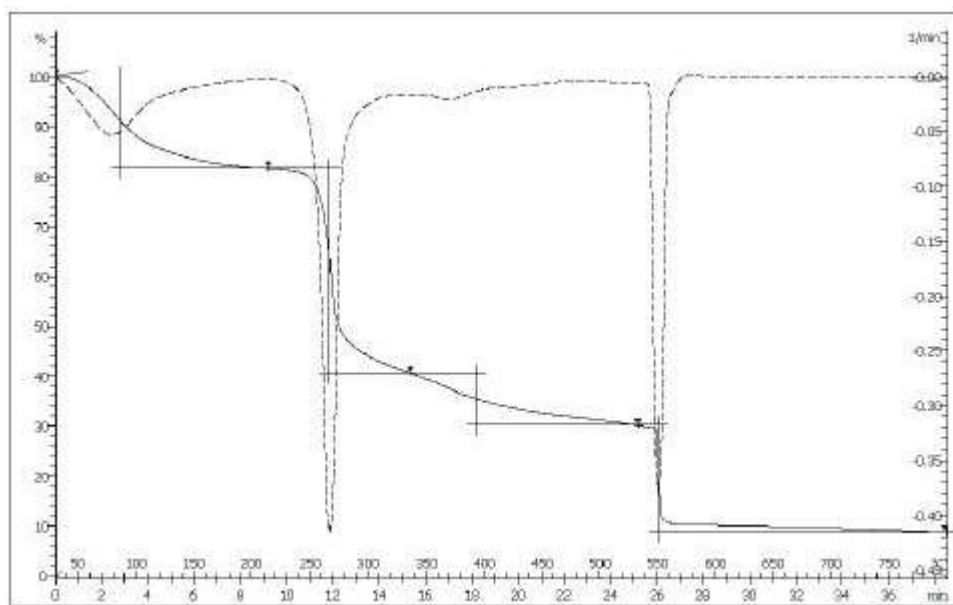


Fig. 2: Thermogravimetry curve for MNG.

Table 1: FTIR wavelength of MNG for photodegradation

0 <sup>th</sup> day, cm <sup>-1</sup>	15 <sup>th</sup> day, cm <sup>-1</sup>	30 <sup>th</sup> day, cm <sup>-1</sup>	Remark
3422	3444	3444	
2936	2924	2925	
2360	2361	2360	
2341	2340	2340	
1617	1624	1623	
1420	1420	1420	
1240	1230	1230	
1148	-	-	Ester stretching
1096	-	-	C-C vibration
1038	1039	1035	
896	897	897	
820	819	819	
669	-	-	C-OH stretching

Table 2: FTIR wavelength of PAM for photodegradation

0 <sup>th</sup> Day, cm <sup>-1</sup>	15 <sup>th</sup> Day, cm <sup>-1</sup>	30 <sup>th</sup> Day, cm <sup>-1</sup>	Remark
3443	3450	3448	
2923	2923	-	C-H stretching
2360	2360	2360	
2340	2339	2339	
1624	1651	1635	
1455	1457	1458	
1409	-	-	C-N stretching
1033	1030	-	C-O stretching vibration
866	849	848	
686	-	-	C-H bending
668	669	669	
653	653	654	

Section 1 of degradation occurs due to depolymerization of simple bonds and desorption of physically absorbed water and water volatilization. Sections 2 and 3 degradation involves complex depolymerization and side group/substituent reaction. The degradation rate is the fastest at Section 2 whereby the weight lost is 41%. This would be the major constituent of MNG. Large numbers of carbon chains and the main chain of the biopolymeric flocculant have been broken down and released CO<sub>2</sub>. At Section 3, there is side group degradation or reaction occurred. Therefore, it decomposes at a higher level and left only 29% residue. Finally, the formation of ash happens at Section 4 at 531.89°C and the analysis ends at 799.12°C.

MNG has fewer complex degradation processes and ash content as compared to PAM in previous study conducted [14]. During the degradation process, PAM released CO<sub>2</sub> and NH<sub>3</sub>. Ammonia gas in the air causes air pollution, acidification in the ecosystem and

respiratory distress to humans. It can be concluded that MNG is easier to degrade thermally and a better material for pollution prevention as compared to PAM.

**Photo-degradation:** Previous photodegradation studies on cassava and corn starches was conducted to examine the stability of chemical structure in the biopolymer, the result showed both polymers depolymerized with the absence of breakage of the C-C bond [15]. Thus, photodegradation study of MNG and PAM was carried out.

**Photodegradation of MNG:** The absorption peak of MNG determined by FTIR technique for 30 days of exposure under natural sunlight is depicted in Table 1. The result shows MNG degraded ester, C-C bonds and C-OH bonds in 15 days.

Photodegradation is a process whereby degradation initiated by absorption of light quanta i.e. ultra violet and visible light. The degradation process involves breaking of the main valence bond in the backbones of the chain or inside groups. Ester, C-C bonds and C-OH bonds are vulnerable under solar light (UV and infra-red) as most commonly chemical bonds broken down are C-C in aliphatic compounds (77-83 kcal/mol) and C-O in aliphatic ethers (76-79 kcal/mol). The break down mechanism is closely related to environmental factors under natural solar exposure. MNG reacts chemically with the atmospheric oxygen and water vapor and UV photons in the sun light. These photochemical changes in the polymer structure causes deterioration in cross-linking and chains scission [16]. Furthermore, the increase in temperature leads to acceleration of deleterious chemical reactions and can speed up the diffusion of low molecular weight components [17].

**Photodegradation of PAM:** Similarly, FTIR technique used to observe the changes in structure of polymer. PAM shows absorption peaks as indicated in Table 2.

The influence of light and other abiotic factors such as water, oxygen, occluded monomer, additives and other impurities cause sunlight-induced oxidation. Under these conditions, weak bond strength could be broken down. With the presence of UV, the structure in PAM underwent photochemical changes and break bonds for C-H (83 kcal/mol), C-N (73 kcal/mol) and C-O (86 kcal/mol) on day 30. The loss of these bonds in PAM is assigned as a primary alcoholic group. As a conclusion, UV broke down single bond both in MNG and PAM with energy around 70-90 kcal/mol during photodegradation.

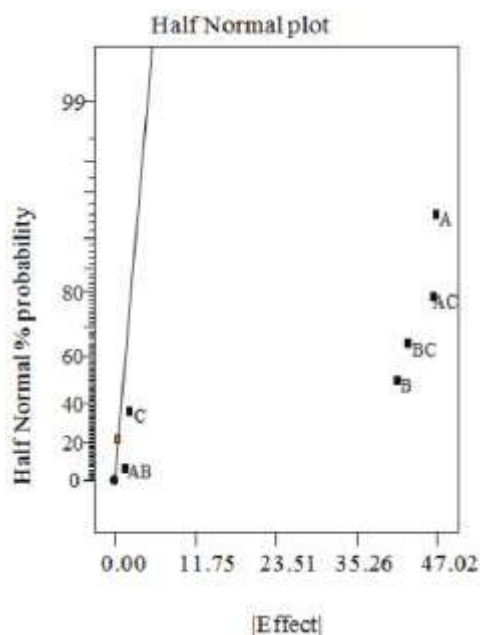


Fig. 3: Half normal plot

**Design of Experiment:** Screening experiment was carried out to identify the influential factors for turbidity reduction where pH, concentration of cation and concentration of MNG denoted for A, B and C, respectively (Fig. 3). Furthermore, results from screening experiment revealed significant factors are factor A (pH), B (concentration of Fe), C (concentration of MNG) and the interaction between AB (pH and concentration of Fe) and AC (pH and concentration of MNG) contributed to the coagulation-flocculation process.

**Interaction between pH and concentration of Fe:** pH influences all the hydrolysis equilibria resulted by addition of the metal cation. When Fe was added into water, hydrolysis reaction takes place. It leads to the formation of various hydrolyzed species as cation. The metal hydroxides have very large surface areas and positive charges. The interaction between pH and cation occurred due to the electrical potential gradient on the particle surface. The electrical potential difference between particle surface and the bulk solution greatly depends on the concentration of potential ionic constituents of the solid particle such as hydrogen ( $H^+$ ) and hydroxyl ( $OH^-$ ) ions. Often, at low pH and low concentration of coagulant, charge neutralization predominant the flocculation process; at high pH and high concentration of coagulant, sweep-floc mechanism may involve in flocculation process.

#### Interaction between pH and concentration of MNG:

It has been noted that a lower concentration of polymers is not enough to bridge particles. Hence, optimum concentration of polymers is preferred for coagulation and flocculation process. At low pH, high concentration of  $H^+$  in water could enhance the bridging mechanism in the presence of acids to form hydrogen-bonding and hydrophobic interactions. The presence of hydrophobic interaction reduces the electrostatic repulsion and promotes attachment on surface of particle. Subsequently, it leads to linking and binding during flocculation. Hence, the concentrations of hydrogen and hydroxyl ions play an important role in flocculation process.

Moreover, MNG has oxygen-containing functional groups, such as,  $OH^-$  (hydroxyl group),  $-COO^-$  (carboxyl groups) and primary amide ( $NH_2$ ) which contains carbonyl ( $C=O$ ) and ester ( $N-C$ ) group. These groups increase hydrogen bonding between surface of particle and MNG. During flocculation, the biopolymeric flocculants expose the electrolyte groups such as negatively charged carboxyl group ( $COO^-$ ) and hydroxyl group ( $OH^-$ ). Then, the chain stretched due to electrostatic repulsion and provides a more effective surface for the kaolin particles to attach. Likewise to polyacrylamide (PAM), it agglomerates the particles via hydrogen bonding with the presence of amide as its functional group [19]. With the presence of  $H^+$  and  $OH^-$  in water, primary amide functional groups in polymers can be extended to produce loops and tails, which lead to the formation of flocs.

Unlike other synthetic coagulants that are widely used in industry, the flocculation mechanism of using MNG as flocculant remains unknown. More than one in four typical flocculation mechanisms could occur in the presence of polymers during flocculation [18]. Based on the properties in MNG, the flocculating mechanism (with the presence of Fe and MNG) is proposed to occur as the following sequence: (1) destabilization of particle by Fe (overcome repulsive electrostatic interaction and neutralize the surface charge), (2) adsorption onto neutralized surface (from MNG functional groups). (3) polymer bridging (MNG extends loops and tails onto surface of particles). Fig. 4 shows the bridging mechanism of adsorption of functional group on neutralized surface of particles. Here, possible flocculation mechanism that may relate to binding and bridging process using MNG is proposed.

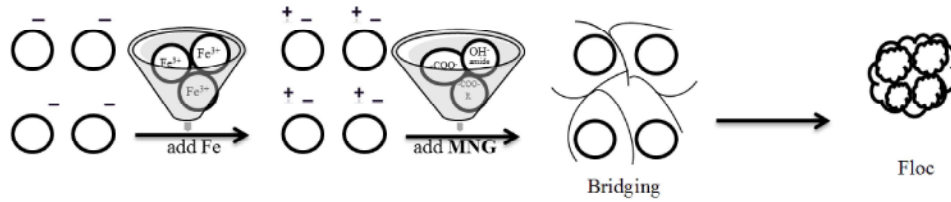


Fig. 4: MNG adsorption on neutralized surface of particle

Table 3: The results of analysis of variance (ANOVA) for turbidity reduction of MNG

Turbidity reduction						
Source	Sum of squares	Degrees of freedom	Mean square	F-value	P-value	
Model	8857.24	9	984.14	25.26	< 0.0001	
pH	3398.86	1	3398.86	87.23	< 0.0001	
Concentration of Fe	1198.15	1	1198.15	30.75	< 0.0002	
Concentration of MNG	742.35	1	742.35	19.05	< 0.0014	
pH×pH	588.97	1	588.97	15.12	< 0.0030	
Concentration of Fe × Concentration of Fe	62.95	1	62.95	1.62	< 0.2325	
Concentration of MNG× Concentration of MNG	124.72	1	124.72	3.20	< 0.1039	
pH× Concentration of Fe	535.63	1	535.63	13.75	< 0.0041	
pH× Concentration of MNG	148.44	1	148.44	3.81	< 0.0795	
Concentration of Fe × Concentration of MNG	216.94	1	216.94	5.57	< 0.0400	
Residual	389.63	10	38.96			
Total	9246.88	19				

**Optimization:** Based on optimization result, the linear effect, contribution of quadratic effect over the linear effect and the interaction terms were significant (P value <0.10) as shown in ANOVA table (Table 3).

It showed the behavior of each variable in the presence of other variables. A second-order model was built and adequately fitted the experimental data. The second-order regression model obtained for turbidity reduction is satisfied since the values of the coefficient of determination ( $R^2$ ) is 0.9579 which is high and close to 1. This indicates that 95.79% of MNG to the total variation was explained by the model and only 4.21% was unexplained. The behaviour of turbidity reduction to optimize the process by finding the best settings of the three variables that maximize the turbidity reduction is described with a model built (Eq. 2).

$$\text{Turbidity reduction} = 96.31 - 18.44 x_1 + 10.95 x_2 + 8.62 x_3 - 14.63 x_1^2 - 4.78 x_2^2 - 6.73 x_3^2 + 8.18 x_1 x_2 + 4.31 x_1 x_3 - 5.21 x_2 x_3 \quad (2)$$

A 3-D optimization plot is shown in Fig. 5 to illustrate the second order quadratic curve. Optimization setting revealed that maximum turbidity reduction (99%) when using MNG as biopolymeric flocculant was achieved at

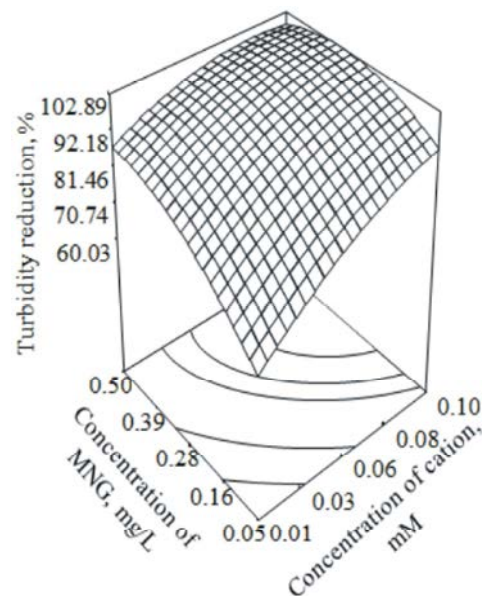


Fig. 5: 3-D optimization plot for MNG

pH 5.77, concentration of cation at 0.05 mM and concentration of MNG at 0.42 mg/L. As compared to PAM in previous study [19\_ENREF\_19], at the 0.05 mM concentration of Fe, the optimal condition for PAM is around pH 8.84 and concentration of PAM required

7.77 mg/L. Therefore, MNG performs better as a flocculant than PAM where it works well at pH near to neutral and requires lower amount of flocculant. Furthermore, less amount of MNG added into water treatment reduces the operating cost. As for PAM, it contains harmful monomers whereby they bring negative impacts to environment and human health. Higher concentration of chemicals added to water may possibly induce secondary pollution and has higher operating cost. From this finding, it can be concluded that, MNG is preferable over PAM as a flocculant in water treatment.

**Validation:** A confirmation experiment was carried out with turbidity reduction from the optimization results in order to validate the developed model. The results of the selected combinations of pH, cation concentration and flocculant concentration display a 98.54% turbidity reduction.

### CONCLUSIONS

The degradation and flocculation study of MNG were presented. MNG requires 17 days to biodegrade whereas PAM shows continuous degradation trend. Next, thermal degradation result revealed that MNG shows fewer residues and stages than PAM when heated and releases only CO<sub>2</sub>. Thirdly, the photodegradation under natural solar exposure shows single bonds chemical structures broken by UV. When using MNG as flocculant in water treatment, the result shows 99% turbidity reduction with pH closer to neutral and lower concentration of flocculant required. In the context of operating a wastewater treatment plant, using MNG is an economically friendly and environmental sound material than that of PAM.

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### Persian Abstract

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#### چکیده

پلی آکریل آمید به عنوان یک خطر جدی برای سلامت انسان شناخته شده است. به تازگی یک لخته‌ساز با پایه گیاهی جدید (MNG) برای تیمار آب و به عنوان یک ماده سبز مورد بررسی قرار گرفته است. این تحقیق به بررسی تجزیه بیولوژیکی، تخریب حرارتی و تخریب نوری MNG و توانایی لخته‌سازی آن می‌پردازد. نتایج نشان می‌دهد MNG میزان تجزیه زیستی بالا و نرخ مصرف اکسیژن پایینی دارد. MNG در روز هفدهم تجزیه زیستی شد، در حالیکه PAM در پایان آزمایش به طور کامل تجزیه زیستی نشد. آنالیز تخریب حرارتی نشان داد که تعداد مراحل تجزیه MNG زیاد نیست و مقداری خاکستر نیز برجای می‌ماند. آنالیز تخریب نوری تغییرات شیمیایی در ساختار MNG و PAM را نشان داد. آزمایشات فاکتوریل نشان داد pH، غلظت کاتیون و غلظت MNG عوامل مؤثر در فرایند انعقاد و لخته‌سازی می‌باشد. مطالعات بهینه‌سازی نشان داد در pH برابر ۵.۷۷، غلظت کاتیون ۰.۰۵ میلی مولار و غلظت MNG ۰.۴۲ میلی گرم بر لیتر کدورت ۹۷٪ کاهش پیدا کرده است. لذا، عملکرد موثر MNG در pH نزدیک خنثی و در غلظت کم است. این نتایج قویاً پیشنهاد می‌کند که MNG یک نامزد مناسب است که می‌تواند به جای PAM، به عنوان لخته‌ساز در تصفیه آب مورد استفاده قرار گیرد.

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