Extraction, characterization and application of malva nut gum in water treatment

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ABSTRACT

In view of green developments in water treatment, plant-based flocculants have become the focus due to their safety, degradation and renewable properties. In addition, cost and energy-saving processes are preferable. In this study, malva nut gum (MNG), a new plant-based flocculant, and its composite with Fe in water treatment using single mode mixing are demonstrated. The result presents a simplified extraction of the MNG process. MNG has a high molecular weight of 2.3×10^5 kDa and a high negative charge of −58.7 mV. From the results, it is a strong anionic flocculant. Moreover, it is observed to have a branch-like surface structure. Therefore, it conforms to the surface of particles well and exhibits good performance in water treatment. In water treatment, the Fe-MNG composite treats water at pH 3.01 and requires a low concentration of Fe and MNG of 0.08 and 0.06 mg/L, respectively, when added to the system. It is concluded that for a single-stage flocculation process, physico-chemical properties such as molecular weight, charge of polymer, surface morphology, pH, concentration of cation and concentration of biopolymeric flocculant affect the flocculating performance.

Key words | biopolymer, malva nut gum, polyacrylamide, response surface methodology, water treatment

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ABBREVIATIONS

MNG malva nut gum
PAM polyacrylamide
RSM response surface methodology

INTRODUCTION

Water and toxic generation and dispersion are examples of major challenges in the sustainability of our planet, where 30% of the world's population will face a water shortage by 2050 and the generation and release of toxic substances into air, water and soil are current global issues. Furthermore, people have a high expectation of a good water quality supply (Hurst *et al.* 2004). Silt, surface runoff, waste discharge, erosion from river banks, excessive algal growth and stir-up from bottom sediments cause turbidity in water (EPA 2013). Turbidity in water provides food and shelter for pathogens that could cause water-borne disease outbreaks and decrease the concentration of dissolved oxygen in water. Moreover, the solids in water may cause problems during the water treatment process (Bratby 2006; Metcalf *et al.* 2013). Therefore, in water treatment, it is a challenge to treat turbid water. Furthermore, the trend is moving forward to conserve energy in water and wastewater treatment plants. Therefore, there is a need to look into energy-saving processes. In a treatment plant, the coagulation–flocculation process is a common and essential treatment process for solid–liquid separation (Ching *et al.* 1994). Flocculation of raw water is essential to affirm good water quality goals (Hurst *et al.* 2004). However, high turbidity water cannot be treated solely by the addition of coagulants such as alum, ferric sulphate or lime. Often, a coagulant aid or flocculant is added into water treatment.

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A renewable material like natural polysaccharide as a flocculant would be a vital and viable alternative in water and wastewater treatment. Moreover, research into renewable resources has increased greatly over the past 10 years (Kerton *et al.* 2013). Undoubtedly, plant-based polymers exhibit the capability to treat industrial wastewater (Carpinteyro-Urban *et al.* 2012). In this study, a new plant-based natural polysaccharide flocculant is extracted as a green material which possesses sustainable properties like abundance, cost efficiency and sustainability. It is extracted from the malva nut and referred to as malva nut gum (MNG).

Use of a natural resource

MNG is extracted from the malva nut, the seed of the *Scaphium scaphigerum* tree. One malva nut tree has the potential to yield up to 40 kg of fruit annually. It produces small glabrous brown-skinned fruit during the dry season from early March to late April (Baird & Dearden 2003). MNG contains a high molecular weight uronic acid. From Fourier transform infrared results, MNG consists of carboxyl (COO^-) and esterified (COO–R) carboxyl groups. Furthermore, alkaline-extracted MNG has a similar monosaccharide composition to arabic gum and flaxseed gum, which are both plant-based flocculants (Somboonpanyakul *et al.* 2006). In addition, it is a plant native to Malaysia, and it is preferable that naturally renewable flocculants are locally grown, as they will be more economical (Kawamura 1991). In contrast to natural flocculants, the price of polyacrylamide (PAM), which is commonly used in water and wastewater treatment plants, is expected to increase over time due to the depletion in natural gas supplies (Piazza *et al.* 2011). Therefore, renewable materials need to be discovered in order to contribute to scientific knowledge and also their behaviour in water treatment. The significance in this study is the capability of a new plant-based flocculant in treating water and the physico-chemical properties which lead to water treatment plant design. The objectives in this study include: (1) to prepare MNG, (2) to characterize MNG, and (3) to determine the physico-chemical parameters in their contribution to water treatment.

METHODS

Preparation of MNG

There are a few methods for MNG extraction and there are pros and cons for the different methods. Wu *et al.* (2007) used Soxhlet apparatus with petroleum ether (30–60 °C) for 5 h, 80% ethanol reflux, hot water extraction (65 °C) and ethanol precipitation. Srichamroen & Chavasit (2011) ground malva nuts into powder and used alkaline extraction followed by ethanol precipitation and freezing at −20 °C. The application of various chemical media and procedures for extraction and high energy consumption to prepare MNG is not preferable in the application of water and wastewater treatment plants. Therefore, in this study, a green production of MNG was slightly modified from the alkaline extraction of Somboonpanyakul *et al.* (2006), as given in Figure 1. The

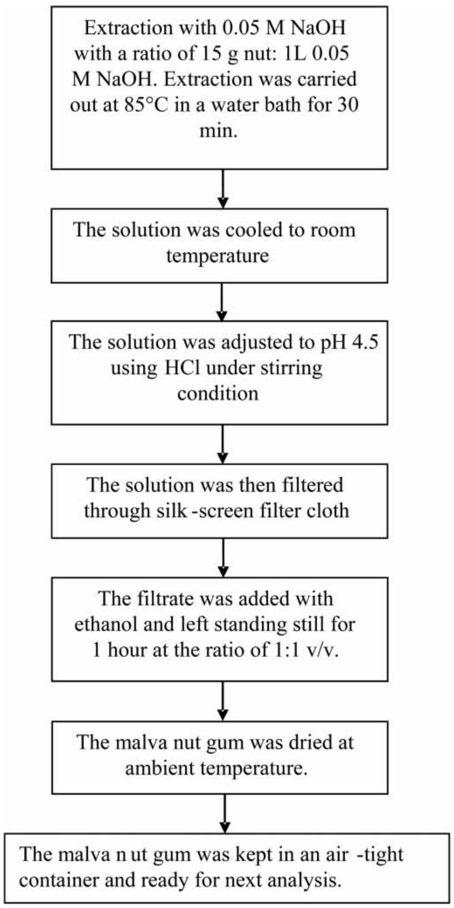

Figure 1 | Schematic diagram of MNG extraction process.

benefits of this extraction process include: (1) only one extraction medium is in use, (2) air-dried extracted MNG saves costs incurred on the energy consumed and the purchase of equipment, and (3) the ratio of nut to volume of alkaline medium is indicated based on the preliminary extraction process carried out to keep the yield to an optimum and, at the same time, reduce the volume of extraction medium used. The extraction process uses chemicals purchased from Qrëc and a water bath from Memmert.

Molecular weight determination

Molecular weight (M_w) was determined using gel permeable chromatography (GPC) supplied by Malvern Instruments, Viscotek (GPCmax) model with TDA305 triple detector array. The column used was an A6000M, general mixed Ag 300 × 0.8 mm. 0.1 mM sodium chloride as a mobile phase was prepared and samples were filtered through a 0.20 μm nylon filter. The result was analysed using OmniSEC software. The method and operating conditions are given in Table 1.

Surface charges determination

MNG was prepared using sodium chloride at 0.1 mM with a sample concentration of 1 mg/mL. The sample was placed in a disposable folded capillary cell and analysed at 25 °C using Malvern Zetasizer (Nano Z) with a measurement angle of 173°. Three runs were performed as replication and there was a minimum of 10 runs in each replication.

Surface morphology examination

Surface morphology was examined using field emission scanning electron microscope, Leo Supra 50VP. Samples were prepared in solid form and underwent coating under vacuum conditions with a thin layer of gold (around 20 nm) in a Sputter Coater Polar SC515 FISONS.

Table 1 | Operating conditions of molecular weight determination by GPC

Criteria	Details
Standard	PolyCAL^{tm} PEO 19k
Verification	Dextran
Flow rate	1 mL/min
Column temperature	25 °C
Sample or injection volume	20 μL

Assay of turbidity reduction

Flocculator (Velp Scientifica type FC6S) was used to study the turbidity in water treatment. Synthetic turbid water was prepared with 0.3 g kaolin clay with 1 L distilled water with initial turbidity at around 400 nephelometric turbidity units (NTU). Fe^{3+} from $FeCl_3$ was chosen as a coagulant and from here onwards it is noted as Fe. It was chosen as it performs in a wide range of pHs. Furthermore, as the costs involved in water treatment plants should be kept to a minimum, a one-stage flocculation process is aimed at achieving maximum treatment yield. In this study, a single mixing mode was applied and was known as the coag–flocculation process. Moreover, the physical parameters, such as mixing speed, mixing time and sedimentation time, were investigated.

$$\text{Percent reduction, } \% = \frac{Ao - A}{Ao} \times 100 \qquad (1)$$

where A_o was the control sample before treatment (NTU) and A was the sample after treatment (NTU).

Design of experiment

The design of the experiment lies within the boundaries as depicted in Table 2. The pH, concentration of cation and concentration of MNG were indicated at low and high limits.

A screening process is crucial as it exhibits the significant factors for the experimental work. The screening of the factors was carried out by using fractional factorial

Table 2 | Design boundaries for screening experiment

Parameter	Low limit	High limit
pH	3	9
Concentration of Fe, mM	0.01	0.1
Concentration of MNG, mg/L	0.05	0.5
Mixing speed, rpm (mixing intensity, G)	30 (51 s^{-1})	150 (116 s^{-1})
Mixing time, min	5	15
Sedimentation time, min	5	10

design. The benefit of this design is in reducing the number of runs (experiments) and maintaining the purpose of the experiment to choose influential factors (Karunakaran 2006). The experimental runs for each replicate to cover all possible combinations for fractional factorial design are 2^{k-1} (Molina *et al.* 2008; Montgomery 2008; Prakash *et al.* 2008). There are six factors involved, namely, pH, concentration of Fe and concentration of MNG, mixing speed, mixing time and sedimentation time. They are denoted as factors A, B, C, D, E and F, respectively, with 2^{6-1} runs.

The optimization in this study was meant to maximize turbidity reduction. Face-centred composite design (FCCD) was employed. FCCD has $\alpha = 1$. In practice, the number of centre runs, $n_c = 2$ or 3, is sufficient to provide good experimental design. However, sometimes more centre runs will be employed to give a reasonable estimate of experimental error (Montgomery 2008).

RESULTS AND DISCUSSION

Effects of molecular weight

Typically, the molecular weight of biopolymers is higher than 10^2 kDa (Yuan *et al.* 2010). In this study, MNG molecular weight exhibited from GPC analysis was 2.3×10^5 kDa. It was much higher than other natural flocculants such as xanthan gum (4 to 25×10^3 kDa) (Corredig & Wicker 2001), pectin (85 to 132 kDa) (Corredig & Wicker 2001), chitosan (around 520 kDa) (Wang *et al.* 2009), to name but a few. Figure 2 shows the refractive index response of MNG for molecular weight determination.

This explains why MNG was only required in low amounts in water treatment to achieve high turbidity reduction. High molecular weight flocculants may have many repeating units that contain functional groups such as hydroxyl and carboxyl groups. In this condition, it is possible for the functional groups to adsorb onto the surface of particles and then induce bridging between particles. It occurs by extending the chain from the surface of particles into the solution and overcoming the electrostatic repulsion between particles. This leads to the formation of a three-dimensional (3D) matrix, which has the capability to form and settle flocs (Salehizadeh & Shojaosadati 2001). By bridging particles using high molecular weight polymers, the flocs formed are stronger than when using salts alone. Due to the strength of the polymer–surface bond being high, the adsorbed polymer chain has attachments at multiple sites on the surface

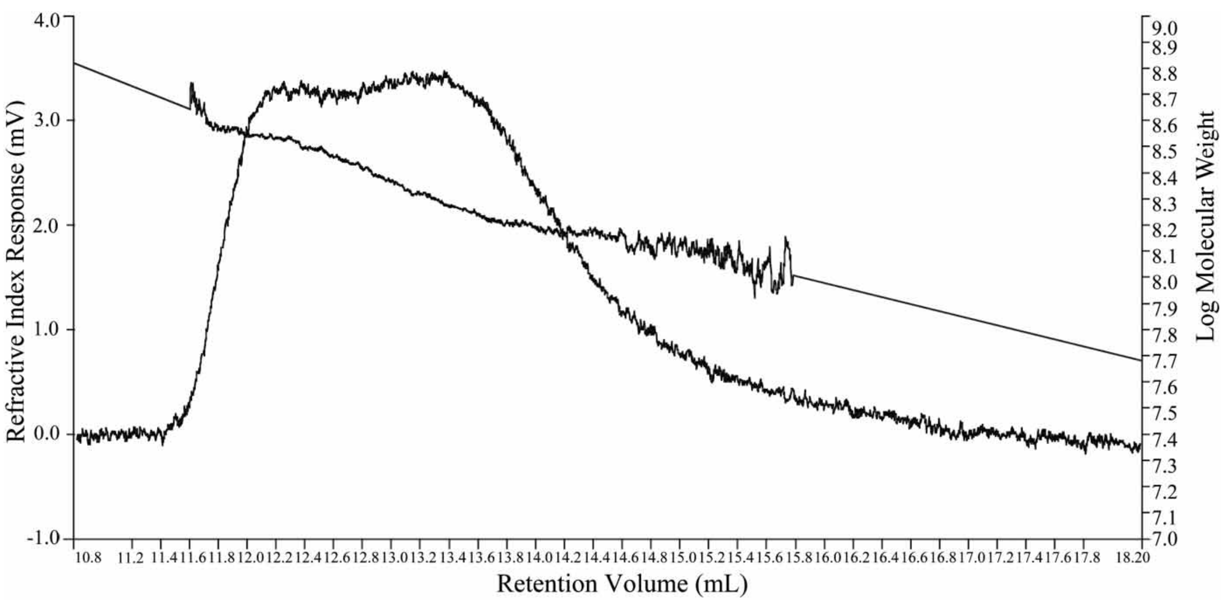

Figure 2 | Elution chromatography (refractive index signal) of MNG analysed by GPC.

(Runkana *et al.* 2006). On the other hand, high concentrations of polymers added may have an adverse impact as this will increase the competition of polymers to adsorb on available sites. Apart from the high molecular weight polymer conformation on the surface of the particles, flocculation may be related to the charge of the polymers.

Effect of polymer charge

Another aspect to examine when determining the strength of a polymer is its charge. Figure 3 shows zeta potential distribution for MNG. The results were analysed using Zetasizer software that applied Smoluchowski's equation. It showed that the charge density of MNG was −58.7 mV and it performed as an anionic polymer or anionic polyelectrolyte flocculant. The small peak near −90 mV is a noise peak. As noted in the section validation analysis on optimized treatment setting, a highly negatively charged polymer like MNG is only required in low concentration in the treatment system. In short, they were inversely proportional. High surface charge in a polymer corresponds to high flocculation capability.

The main reason why MNG has a high negative charge is due to the functional groups in MNG, which consist of hydroxyl and carboxyl groups. Furthermore, as shown earlier, MNG has a high molecular weight where there are many repeating units of negatively charged functional groups. For kaolin particles, like the colloids in water, it has a negative charge on the surface of particles. After the cation is added, it reduces the surface charges on particles with its positive charge, thus forming micro-flocs during collisions. However, the flocs formed may not be dense enough to settle or may form only a small amount of aggregate. Subsequently, MNG further induces flocculation by overcoming the electric potential between the particles and attracts the positive edge of the surface charge of particles and then bridges the flocs to aggregate better by the negatively charged functional groups in MNG. Therefore, MNG exhibits a good role in the flocculation process.

Effect of surface morphology

Investigation of surface morphology is necessary to determine the surface structure likely to conform well with the surface of particles. For example, crystal-linear structure (Xia *et al.* 2008) and lace-like surface (Mishra *et al.* 2011) flocculants were found to exhibit good flocculating performance by effectively adsorbing onto the surface of particles and thus improving the conformation of the flocculant on the surface of particles. Figure 4 shows the surface morphology of MNG. It is seen to have a branch-like structure. The flocculating performance in the section Validation analysis shows that the branch-like structure adequately contributes to the flocculating mechanism.

Design of experiment

Flocculating performance was affected by a few factors. Conformation of flocculant to the surface of particles by adsorption may relate to the molecular weight and surface

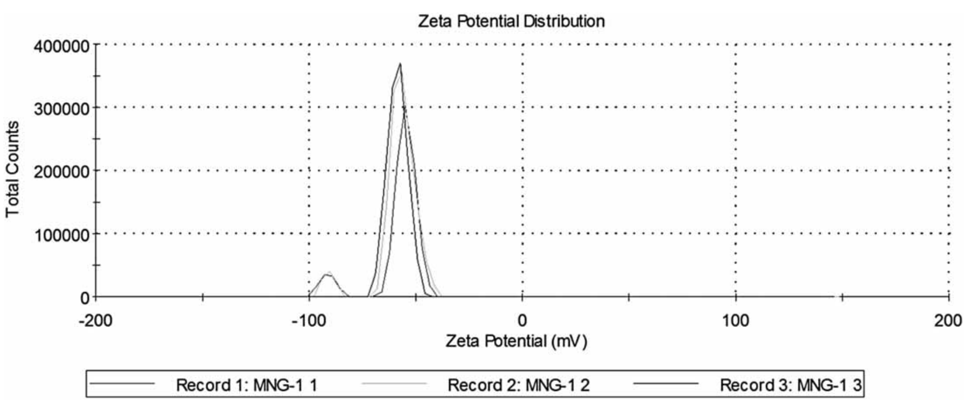

Figure 3 | Zeta potential distribution.

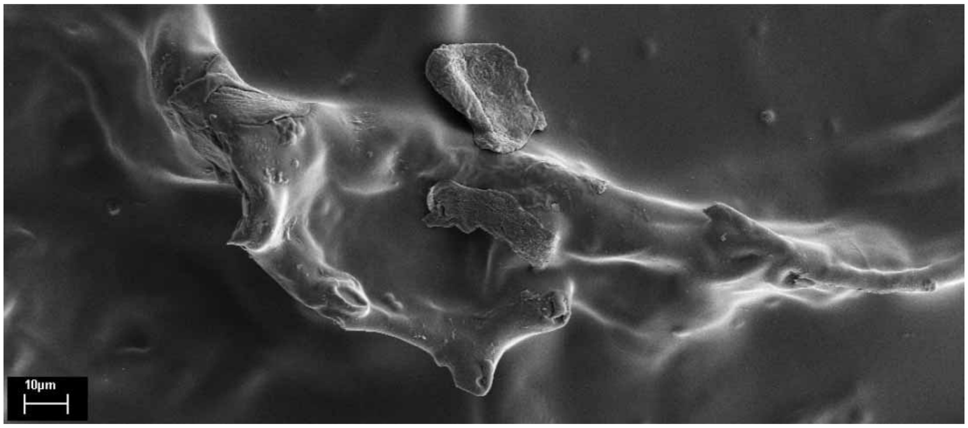

Figure 4 | Scanning electron microscopy micrograph of MNG.

structure of the flocculant, and the surface charge of the particles. However, the characteristics of the solution and physical mixing parameters such as mixing time, mixing speed and sedimentation time require investigation. Next, we evaluated these factors which contribute to the flocculation process by using factorial design. Two-level fractional factorial design was used for a screening experiment including six factors. The total number of runs was 32.

Normal probability plot (Figure 5) shows the significant factors in the flocculation process when using MNG indicating that the factors depend on each other. It can be seen that only three factors, that is, pH (A), concentration of Fe (B) and concentration of MNG (C) were significant.

The screening of factors revealed that factors A, B and C affected the coagulation–flocculation process. Therefore, these three factors are included for optimization to maximize the response output–turbidity reduction. For

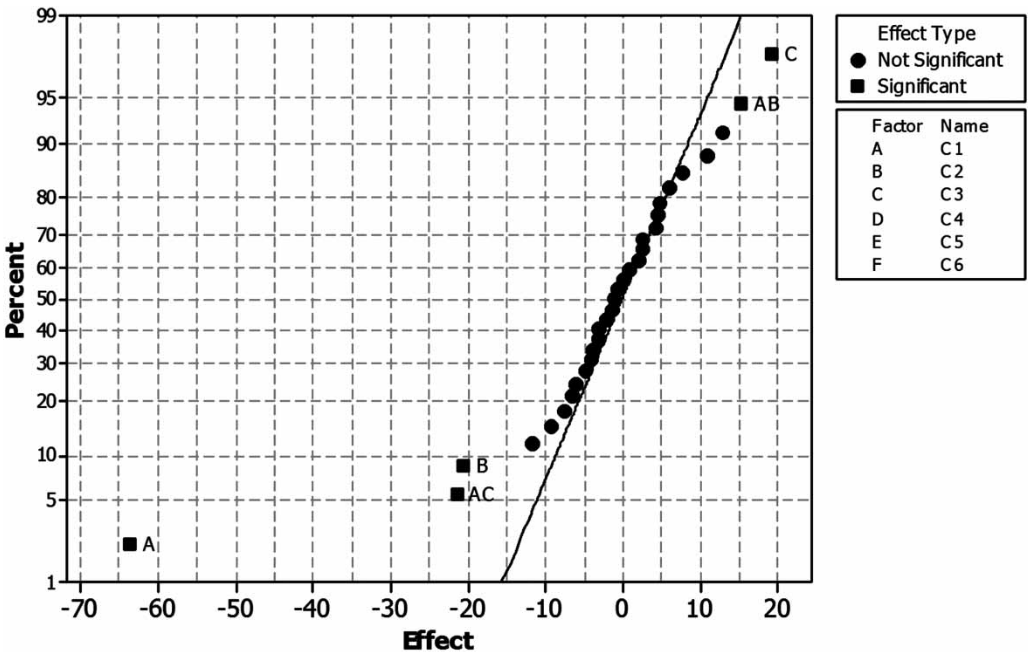

Figure 5 | Normal probability plot of the effects of MNG in single mixing mode. A: pH, B: Concentration of Fe, C: Concentration of MNG, D: Mixing speed, E: Mixing time, F: sedimentation time.

optimization, response surface methodology (RSM) was applied. RSM is used by researchers in the field of engineering and technology to determine the values of the input process parameters that yield the optimum responses (Ho et al. 2010). In this study, FCCD with 20 runs was used to find the best operating condition for the flocculation process. Table 3 shows the FCCD with natural variables for turbidity reduction when using an Fe-MNG composite. There are 20 runs including six centre points used to evaluate the effects of pH, concentration of cation and concentration of MNG towards turbidity reduction.

The optimization process helps in finding the maximum turbidity reduction in this study. Table 4 shows the analysis of variance (ANOVA). Each variable term (linear, square and interaction) showed P values <0.05 and, thus, they are statistically significant. In other words, the main effect of all selected factors was significant. In addition to the main effect, the interaction between different factors was significant as well. The ANOVA revealed that second-order models adequately fitted the experimental data for the flocculants. A regression model was built. The second-order polynomial model in coded unit that correlates turbidity reduction with all the significant variable terms is expressed in Equation (2). The positive coefficients of variable terms indicate their synergistic effects on turbidity reduction, otherwise, the negative coefficients show their antagonistic effects (Chang et al. 2011). The second-order regression model obtained for the optimization was satisfied since the value of the coefficient of determination (R^2) is high and close to 1. The R^2 was 0.9810. It indicates 98.10% of the total variation was explained by the model and only 1.9% was unexplained when using MNG in turbidity reduction. Based on coefficients in Equation (2), x_1 was shown to have the highest influence on turbidity reduction. Figure 6 shows the behaviour of two factors interacting with each other.

$$\text{Turbidity reduction} = 34.26 - 31.95x_1 - 6.13x_2 + 11.91x_3 + 21.65x_1^2 - 20.52x_2^2 + 21.35x_3^2 - 5.22x_1x_2 + 5.50x_1x_3 - 6.40x_2x_3 \quad (2)$$

where x_1 is pH, x_2 is concentration of Fe and x_3 is concentration of MNG.

The optimization analysis showed that the optimum setting was found to be at pH 3.01, and 0.08 mM and 0.06 mg/L for concentration of Fe and concentration of MNG, respectively. In line with the result of high molecular weight and negative charge in MNG, it was shown to be a strong anionic polymer flocculant, and when a low concentration was added to the process, it achieved high flocculating performance (97% turbidity reduction). This shows that MNG works well in single mixing mode where it requires minimum mixing speed. Furthermore, it requires short collision time for one particle to attach to another and settles fast. This may be due to the strong affinity possessed by MNG to attract particles with greater separation distances in solution and to conform to the surface of the particles in the split second it touches their surface, and subsequently it forms flocs quickly, which are dense enough to settle as soon as they aggregate. However, the characterization and optimization results showed that these factors are influencing the flocculation process. Therefore, these factors are interrelated where MNG has many repeating units consisting of negatively charged

Table 3 | FCCD in natural variables with the experimental data values of turbidity reduction for MNG

pH	Concentration of Fe, mM	Concentration of MNG, mg/L	Turbidity reduction, %
3	0.1	0.05	88.7
6	0.055	0.275	95.5
6	0.1	0.275	97.4
6	0.055	0.05	85.6
6	0.055	0.275	96.3
6	0.055	0.275	95.4
3	0.1	0.5	98.7
3	0.01	0.05	79.7
3	0.055	0.275	97.0
6	0.055	0.275	95.7
9	0.01	0.05	5.64
9	0.1	0.05	67.5
3	0.01	0.5	90.4
6	0.055	0.5	95.9
9	0.1	0.5	74.6
9	0.055	0.275	68.7
9	0.01	0.5	53.7
6	0.01	0.275	88
6	0.055	0.275	95.3
6	0.055	0.275	95

Table 4 | The results of ANOVA for turbidity reduction of MNG

Source	Sum of squares	Degree of freedom	Mean square	*F*-value	*P*-value
Model	16,996.77	9	1888.53	57.49	< 0.0001
pH	10,208.02	1	10,208.02	310.76	< 0.0001
Concentration of Fe	375.16	1	375.16	11.42	< 0.0070
Concentration of MNG	1,418.48	1	1418.48	43.18	< 0.0001
pH * pH	1,288.99	1	1288.99	39.24	< 0.0001
Concentration of Fe* Concentration of Fe	1,158.51	1	1158.51	35.27	< 0.0001
Concentration of MNG* Concentration of MNG	1,253.51	1	1253.51	38.16	< 0.0001
pH* Concentration of Fe	218.40	1	218.40	6.65	< 0.0275
pH* Concentration of MNG	242.00	1	242.00	7.37	< 0.0218
Concentration of Fe* Concentration of MNG	327.68	1	327.68	9.98	< 0.0102
Residual	328.49	10	32.85		
Total	17,325.25	19			

F-value, computed *F*-value; *P*-value, probability value, or the actual area under the standard normal distribution curve; DF, degrees of freedom.

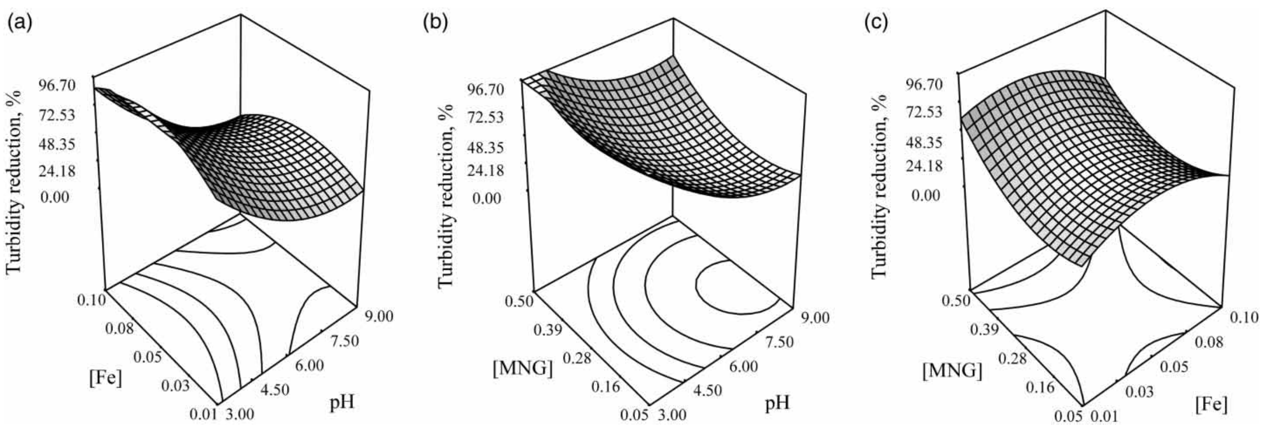

Figure 6 | 3-D response surface plots of (a) pH and concentration of Fe, (b) pH and concentration of MNG, and (c) concentration of Fe and MNG.

functional groups and has a branch structure that easily stretches out and is adsorbed on the surface of particles during the bridging process. Owing to these facts, the flocculation process is easily achieved by single mode mixing with low concentration of MNG added to the system.

Effect of pH and concentration of Fe

pH influences all the hydrolysis equilibria resulting from addition of metal cations. When Fe is added to water, a hydrolysis reaction forms hydrolysed species as cations (Domínguez *et al.* 2005). The metal ions in the hydroxides are positively charged and are influenced by the pH, which causes electrical potential gradient on the particle surface. The difference in electrical potential between the surface of particles and water is greatly influenced by the concentration of hydrogen (H^+) and hydroxyl (OH^-) ions (Stechemesser & Dobias 2005).

Effect of pH and concentration of MNG

Optimum concentrations of polymer and H^+ are preferred for coagulation and flocculation processes. A lower concentration of polymer is insufficient to bridge particles (Krishnamoorthi *et al.* 2007). In this study, the optimum pH was pH 3. The higher concentration of H^+ in water enhances the bridging mechanism to form hydrogen bonding and hydrophobic interactions. The presence of hydrophobic interaction reduces the electrostatic repulsion and promotes attachment on the surface of particles. Subsequently, it leads to linking and binding during flocculation (Tsoga *et al.* 2004). Hence, the concentrations of hydrogen and hydroxyl ions play an important role in the flocculation process.

Moreover, MNG consists of oxygen-containing functional groups, such as OH^- (hydroxyl group) and COO^- (carboxyl group). These groups increase hydrogen bonding between the surface of the particles and MNG. During the flocculation process, high molecular weight and high charge density characteristics in MNG expose the functional groups; the chain stretches out due to electrostatic repulsion and provides a more effective surface for the kaolin particles to attach to.

Effect of concentration of Fe and concentration of MNG

Simple charge neutralization, charge patch neutralization, polymer bridging and polymer depletion are four typical mechanisms for flocculation in the presence of polymers. A combination of a few flocculation processes among the four may occur (Napper 1983; Levine & Friesen 1987). During the coagulation–flocculation process, Fe and MNG are added in different steps. Therefore, the primary mechanism of MNG might be a bridging mechanism. The concentration of Fe added may be insufficient during coagulation. Thus, a higher concentration of MNG is required to achieve the charge patch neutralization and bridging mechanism during the flocculation process. However, if the Fe added is excessive, it will cause a reverse effect or prefer charge patch neutralization as there is attraction of opposite charges.

On the other hand, in the coagulation–flocculation process, Fe and MNG are added together. Therefore, the dominant role of MNG could be determined. The neutralization mechanism was shown to be dominated by Fe where it required a higher concentration to destabilize the opposite charge edge of the surface of particles. Since Fe plays an important role in the neutralization mechanism, the role of MNG in charge patch neutralization may only make a small contribution. Therefore, it was dominated by the bridging mechanism. MNG extended the tails and loops to bridge the neutralized particles and microflocs. Subsequently, when the flocs were denser, it settled. Conclusively, when the Fe-MNG composite is added, (1) Fe destabilizes the surface of particles and possibly forms microflocs and (2) MNG bridges the particles and microflocs.

Validation analysis

As a validation to the optimized treatment condition using kaolin suspension (synthetic turbid water), the treatment setting was applied to surface water as well in order to evaluate the feasibility of using MNG in water treatment. The river water was sampled from the Sungai Pinang River which is located on Penang Island, Malaysia. The river is fed by six tributary rivers, namely, Sungai Dondang, Sungai Air Itam, Sungai Air Putih, Sungai Air Terjun, Sungai Jelutong and Sungai Kecil. All the rivers make up the Sungai Pinang basin. Since 2011, the water quality of Sungai Pinang has been categorized as Class III as per the National Water Quality Standards for Malaysia (DoE 2006). They indicate the polluted water requires extensive treatment before drinking and is suitable for recreational use without body contact. Therefore, a validation analysis was carried out (Table 5) on the river water. It is observed that the treatment settings fit well in river water treatment, in addition to kaolin suspension.

Table 5 | Optimized treatment setting and validation test when using MNG in coagulation–flocculation process using kaolin suspension and river water

			Turbidity reduction, %		
pH	**Concentration of Fe, mM**	**Concentration of MNG, mg/L**	**Predicted (kaolin suspension)**	**Actual (kaolin suspension)**	**Actual (river water)**
5.77	0.05	0.42	99.5	97.4	95.2

CONCLUSION

In summary, a new renewable material for water treatment was presented. Some intriguing features make MNG a green material, as follows: (1) it is extracted from a natural and renewable resource; (2) it possesses high molecular weight, surface charge and a branch-like surface morphology; and (3) it exhibits good flocculation performance at near neutral pH and is only required at low concentrations. The aim is to replace PAM in water treatment for economic and sustainability reasons as well as because of the increasing health concerns of using PAM.

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REFERENCES

Baird, I. G. & Dearden, P. 2003 Biodiversity conservation and resource tenure regimes: a case study from northeast Cambodia. *Environ. Manage.* **32** (5), 541–550.

Bratby, J. 2006 *Coagulation and Flocculation in Water and Wastewater Treatment*. International Water Association, London.

Carpinteyro-Urban, S., Vaca, M. & Torres, L. 2012 Can vegetal biopolymers work as coagulant–flocculant aids in the treatment of high-load cosmetic industrial wastewaters? *Water Air Soil Pollut.* **223** (8), 4925–4936.

Chang, S. H., Teng, T. T., Ismail, N. & Alkarkhi, A. F. M. 2011 Selection of design parameters and optimization of operating parameters of soybean oil-based bulk liquid membrane for Cu(II) removal and recovery from aqueous solutions. *J. Hazard. Mater.* **190** (1–3), 197–204.

Ching, H.-W., Tanaka, T. S. & Elimelech, M. 1994 Dynamics of coagulation of kaolin particles with ferric chloride. *Water Res.* **28** (3), 559–569.

Corredig, M. & Wicker, L. 2001 Changes in the molecular weight distribution of three commercial pectins after valve homogenization. *Food Hydrocolloids* **15** (1), 17–23.

DoE 2006 *National Water Quality Standard for Malaysia*. DoE, Malaysia.

Domínguez, J. R., de Heredia, J. B., González, T. & Sanchez-Lavado, F. 2005 Evaluation of ferric chloride as a coagulant for cork processing wastewaters. Influence of the operating conditions on the removal of organic matter and settleability parameters. *Ind. Eng. Chem. Res.* **44** (17), 6539–6548.

EPA 2013 Turbidity. http://water.epa.gov/type/rsl/monitoring/vms55.cfm (accessed 21 May 2013).

Ho, Y. C., Ismail, N., Alkarkhi, A. F. & Morad, N. 2010 Reuse of fruit waste as biopolymeric flocculant and optimizing turbidity reduction: comparison study with industrial flocculant. *J. Environ. Eng.* **136** (11), 1267–1276.

Hurst, A. M., Edwards, M. J., Chipps, M., Jefferson, B. & Parsons, S. A. 2004 The impact of rainstorm events on coagulation and clarifier performance in potable water treatment. *Sci. Total Environ.* **321** (1–3), 219–230.

Karunakaran, V. 2006 Quality assurance and optimization studies of light weight PU prosthetic foot. *Trends Biomater. Artif. Organs* **19** (2), 63–69.

Kawamura, S. 1991 Effectiveness of natural polyelectrolytes in water treatment. *J. Am. Water Works Assoc.* **83**, 88–91.

Kerton, F. M., Liu, Y., Omari, K. W. & Hawboldt, K. 2013 Green chemistry and the ocean-based biorefinery. *Green Chem.* **15**, 860–871.

Krishnamoorthi, S., Mal, D. & Singh, R. 2007 Characterization of graft copolymer based on polyacrylamide and dextran. *Carbohyd. Polym.* **69** (2), 371–377.

Levine, S. & Friesen, W. 1987 Flocculation of colloid particles by water-soluble polymers. *Flocculation in Biotechnology and Separation Systems*. Elsevier, Amsterdam, pp. 3–20.

Metcalf & Eddy, Tchobanoglous, G., Stensel, H. D., Tsuchihashi, R. & Burton, F. 2013 *Wastewater Engineering: Treatment and Resource Recovery*. McGraw-Hill Education, New York.

Mishra, S., Mukul, A., Sen, G. & Jha, U. 2011 Microwave assisted synthesis of polyacrylamide grafted starch (St-g-PAM) and its applicability as flocculant for water treatment. *Int. J. Biol. Macromol.* **48** (1), 106–111.

Molina, M. A., Zhao, W., Sankaran, S., Schivo, M., Kenyon, N. J. & Davis, C. E. 2008 Design-of-experiment optimization of exhaled breath condensate analysis using a miniature differential mobility spectrometer (DMS). *Anal. Chim. Acta* **628** (2), 155–161.

Montgomery, D. C. 2008 *Design and Analysis of Experiments*. Wiley, New York.

Napper, D. H. 1983 *Polymeric Stabilization of Colloidal Dispersions*. Academic Press, London.

Piazza, G. J., McAloon, A. J. & Garcia, R. A. 2011 A renewable flocculant from a poultry slaughterhouse waste and preliminary estimate of production costs. *Resour. Conserv. Recycle* **55** (9–10), 842–848.

Prakash, O., Talat, M., Hasan, S. & Pandey, R. K. 2008 Factorial design for the optimization of enzymatic detection of cadmium in aqueous solution using immobilized urease from vegetable waste. *Bioresour. Technol.* **99** (16), 7565–7572.

Runkana, V., Somasundaran, P. & Kapur, P. 2006 A population balance model for flocculation of colloidal suspensions by polymer bridging. *Chem. Eng. Sci.* **61** (1), 182–191.

Salehizadeh, H. & Shojaosadati, S. 2001 Extracellular biopolymeric flocculants: recent trends and biotechnological importance. *Biotechnol. Adv.* **19** (5), 371–385.

Somboonpanyakul, P., Wang, Q., Cui, W., Barbut, S. & Jantawat, P. 2006 Malva nut gum. (Part I): Extraction and physicochemical characterization. *Carbohyd. Polym.* **64** (2), 247–253.

Srichamroen, A. & Chavasit, V. 2011 Rheological properties of extracted malva nut gum (*Scaphium scaphigerum*) in different conditions of solvent. *Food Hydrocolloids* **25** (3), 444–450.

Stechemesser, H. & Dobias, B. 2005 *Coagulation and Flocculation*. CRC Press, Boca Raton.

Tsoga, A., Richardson, R. & Morris, E. 2004 Role of cosolutes in gelation of high-methoxy pectin. Part 1. Comparison of sugars and polyols. *Food Hydrocolloids* **18** (6), 907–919.

Wang, J.-P., Chen, Y.-Z., Yuan, S.-J., Sheng, G.-P. & Yu, H.-Q. 2009 Synthesis and characterization of a novel cationic chitosan-based flocculant with a high water-solubility for pulp mill wastewater treatment. *Water Res.* **43** (20), 5267–5275.

Wu, Y., Cui, S. W., Tang, J., Wang, Q. & Gu, X. 2007 Preparation, partial characterization and bioactivity of water-soluble polysaccharides from boat-fruited sterculia seeds. *Carbohyd. Polym.* **70** (4), 437–443.

Xia, S., Zhang, Z., Wang, X., Yang, A., Chen, L., Zhao, J., Leonard, D. & Jaffrezic-Renault, N. 2008 Production and characterization of a bioflocculant by *Proteus mirabilis* TJ-1. *Bioresour. Technol.* **99** (14), 6520–6527.

Yuan, S.-J., Sun, M., Sheng, G.-P., Li, Y., Li, W.-W., Yao, R.-S. & Yu, H.-Q. 2010 Identification of key constituents and structure of the extracellular polymeric substances excreted by Bacillus megaterium TF10 for their flocculation capacity. *Environ. Sci. Technol.* **45** (3), 1152–1157.

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