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Nanofibrillated cationic cellulose derivatives as flocculants for domestic wastewater treatment

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ABSTRACT

Natural-based coagulants have emerged as a reliable option to implement more sustainable operations and management of wastewater treatment plants. This work aims at evaluating the use of cationic nanofibrillated celluloses (cNFC) as novel bio-based flocculants to treat domestic wastewaters by the most widely employed treatment process – coagulation/flocculation. Two cNFC samples were prepared with different charge densities and tested as coagulant/flocculants using different water characteristics. The effect of cNFCs was studied by measuring the residual turbidity and dissolved organic carbon. The aggregation mechanism and kinetics of flocculation were also evaluated. Results show that cNFC can be used as an efficient flocculant to treat medium and high DOC waters since they considerably reduce turbidity (turbidity removals varied between 66.0 % and 85.7 % for the waters and cNFCs tested) without increasing dissolved organic carbon. Instead, cNFC removed dissolved organic carbon from domestic wastewaters (between 22.1 % and 65.5 % of DOC removals for the waters and cNFCs tested), which is a novel remarkable finding and a step forward in this knowledge area. High density charged cNFC revealed superior removal capacity at lower doses than the commercial coagulant $FeCl_3$.

1. Introduction

Metal-based coagulants, such as aluminium or iron salts, and more recently synthetic polymers, are usually applied in the coagulation/flocculation (C/F) process in water and wastewater treatment. This treatment process is cost-efficient, simple, and effective, has low energy demand, and targets the removal of suspended solids, colour, organic compounds, microorganisms/pathogens, micropollutants, and some inorganic compounds [1,2]. However, regardless of the recognized efficiency of the aluminium and iron based coagulants, they often require pH and alkalinity adjustments, generate high volumes of sludge, are harmful to human health, especially the aluminium-based ones [3], may have a negative ecotoxicological impact when introduced in the environment through sludges, and some are not biodegradable [4,5]. Therefore, the use of natural coagulants has emerged as a reliable solution to address sustainable operations and management of wastewater treatment plants (WWTP). Natural coagulants are biodegradable and renewable (feedstock can be obtained easily), non-hazardous, cost-effective (sludge handling/treatment and coagulant cost), and of simple operation [3,5]. Natural coagulants can be sourced from various natural feedstock, mostly plant and marine biomass-based [2,5,6]. Among the different possibilities, microbial polysaccharides, starch, cellulose derivatives, gelatine, galactomannans, chitosan, alginate, glues, biowastes, and *Moringa oleifera* have been studied and tested for this purpose [1,5–8]. According to Koul et al. [1], the natural coagulants mechanisms of action are close to those of polyelectrolytes, namely charge neutralization, sweep flocculation, inter-particle bridging, and double-layer compression. Therefore, they can be effectively applied to

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the most widely employed technology in water and wastewater treatment, C/F, with high potential as substitutes for both synthetic metal and polymer-based coagulants. However, these materials can originate an increase in the organic load of the treated water, which is a source of colour, odour, and taste, may be precursors of disinfection by-products, and may induce the enhancement of microbial activity [2,8].

Among natural candidates, cellulose is the most abundant and is recognized by its non-toxicity, biodegradability, hydrophilicity, and broad chemical variability due to the reactivity of the hydroxyl groups [9]. Nanocelluloses can boost the cellulose benefits due to their unique characteristics, such as nanosized scale dimensions (thickness), high surface area, and fibrils morphology [10]. These nanomaterials also exhibit low density, high mechanical strength and stability, chemical inertness, and the ability to modify their surface chemistry [10]. Therefore, nanocelluloses have a broad range of applications, including in wastewater treatment, as adsorbent to remove heavy metals [11], dyes [12], and natural organic matter and pharmaceuticals [13,14], as well as flocculant to remove turbidity and dyes [15–19]. Despite the potential of nanocelluloses in wastewater treatment, the literature is surprisingly scarce. Most of the studies focus on anionic nanocelluloses which, for instance, reveal good turbidity removal (99.5 %) in the treatment of a kaolin suspension with high turbidity, using 300 mg/L of $CaCl_2$ as a coagulant [18]. Using a dye effluent from textile industry, these authors demonstrated that anionic nanocelluoses removed 80.9 % of turbidity and 36.5 % of chemical oxygen demand (COD). These authors also showed the highest carboxylic group content tested provided more active flocculation/adsorption. Suopajärvi et al. [16] showed the good performance of an anionic nanofibrillated dicarboxylated cellulose in the C/F treatment of municipal wastewater. The selected system produced low residual turbidity (removal of 50–70 %) and COD (removal of 30–50 %) in treated water when $Fe(SO_4)_3$ was also added [16]. They considered that the charged ferric ion establishes cationic patches on the particles' surface and the anionic nanofibrils bridge with the particles via these patches. On the other hand, in Suopajärvi et al. [15] sulfonated nanocelluloses were compared with anionic dicarboxylic nanocelluloses, also using $Fe(SO_4)_3$ before adding the sulfonated nanocelluloses. The former flocculant had better performance than the latter in removing turbidity at low flocculant dosages. However, none of these authors studied the solo use of nanocelluloses directly, as flocculants in wastewater treatment, as this work made and which is a novelty. More recently, Haleem et al. [19], addressed that for the pH conditions and optimal cationic cellulose dose, up to 53.3 % of total the suspended solids (TSS) were removed from a dairy wastewater. Despite the relevant findings of these authors [19], they only tested a very specific wastewater (dairy) with distinctive characteristics, different from the much more abundant domestic wastewaters. Moreover, they only analyse pH and TSS, not having carried any dissolved organic carbon (DOC) measurements. This finding served as a promising foundation for our research, where nanofibrillated celluloses are applied to wastewater treatment.

This work aims to evaluate the suitability of cationic nanofibrillated celluloses (cNFC) as coagulant/flocculants, as a proof-of-concept of the use of novel bio-based materials to treat domestic wastewaters. To the best of the authors knowledge, this application of cNFCs has never been addressed. For that propose, two differently charged cNFCs were chosen and used in coagulation/flocculation tests in wastewaters with different organic matter content. The effect of cNFCs was studied by measuring the residual turbidity and DOC. Further insights on the aggregation mechanisms and kinetics were accessed by laser diffraction spectroscopy. The cNFC coagulation/flocculation performance was also compared with a conventional commercial coagulant ($FeCl_3$), serving as a benchmark for its high effectiveness in wastewater treatment.

2. Materials and methods

2.1. Materials

Bleached Eucalyptus kraft pulp (BEKP), containing 80–85 wt% cellulose, 14–19 wt% xylan, 0.3 wt% lignin, and 0.4 wt% extractives, was used as starting material for cCNFs preparation. 3-chloro-2-hydroxypropyl) trimethylammonium chloride (60 wt% aqueous solution), sodium hydroxide and hydrochloric acid (1 M aqueous solutions) were acquired from Sigma Aldrich (Schnelldorf, Germany); sodium hydroxide pellets were obtained from VWR (Carnaxide, Portugal) and isopropanol (IPA) was purchased from Labsolve (Lisbon, Portugal).

Ferrous chloride from Riedel-de-Haën - Fisher Scientific (granulate pure, $FeCl_36H_2O$) was used as a commercial coagulant in coagulation/flocculation experiments.

2.2. Production and characterization of cationic Nanofibrillated cellulose (cNFC)

The production of the cNFCs was performed following an adapted procedure described in Pedrosa et al. [20]. Briefly, industrial unrefined and never dried BEKP was used as raw material to prepare the cNFCs. A direct cationization method, using CHPTAC ((3-chloro-2-hydroxypropyl) trimethylammonium chloride) under alkaline conditions, was used to covalently bond quaternary ammonium groups to the kraft pulp. A 5 % (m/v) solution was prepared dispersing BEKP in a 30/70 (*v*/v) water/isopropanol mixture. Two distinct molar ratios of CHPTAC per anhydroglucose unit (AGU) were used (i.e., 0.5 and 3). The CHPTAC was added to the BEKP (one-third of the volume at 0-, 1- and 2-h reaction) and the medium was alkalinized by adding NaOH equivalent to a NaOH/CHPTAC molar ratio of 2. The cationization was carried out at 70 °C for 4 h, the cationized fibres were vacuum filtered through a filter paper (pore size 11 μm) and washed with distilled water until the conductivity of the washing water was constant. A reaction scheme of the cationization procedure is displayed in Fig. S1 (supporting information). Finally, the cationized fibres were subject to mechanical treatment, consisting of high-pressure homogenization, performed at room temperature. The least cationized sample was subjected to one cycle at 500 bar and two cycles at 1000 bar, while the sample with a higher cationization degree was treated with one cycle at 500 bar and four cycles at 1000 bar. The obtained samples presented a concentration of ca. 1 wt% solids. Two degrees of substitution (DS) were obtained and studied, namely DS = 0.106, coded as cNFC++, and DS = 0.06, coded as cNFC+.

The incorporation of the quaternary ammonium group into the NFC backbone was evaluated by FTIR. A Bruker Tensor 27 spectrometer with a MKII Golden Gate accessory was used to acquire the FTIR spectra, in attenuated total reflectance (ATR) mode with 128 scans, in the range 650–4000 cm^{-1} and a resolution of 4 cm^{-1}, at room temperature. The spectra were treated for noise reduction, baseline slope and normalized.

The produced cNFC were further characterized in terms of their DS, yield of fibrillation (YF), soluble fraction (SF), charge density (CD), and transmittance of the suspensions.

To determine the YF, suspensions with 0.05 wt% solid content were centrifuged at 2400 xg for 30 min and the non-fibrillated fraction that settled in the bottom of the flask was weighed. The YF (%) was estimated by the difference between the original amount of weighted material and the remaining fibres in the supernatant after centrifugation.

The soluble fraction was determined by passing 3 mL of the original cationic cellulose suspension (vacuum filtered) through a cellulose acetate membrane filter (0.2 μm pore size). After completing the filtration, the filter was dried (50 °C for 24 h) and the material retained by the membrane was weighed to determine the percentage of fibres in the filtrate. The material (fine fraction) that was able to pass through the membrane was defined as “soluble fraction 0.2” (SF0.2). The assays were performed in duplicate.

The transmittance of the suspensions containing 0.1 wt% of cNFC

Table 1
Characteristics of the wastewaters collected.

	Low DOC water	Medium DOC water	High DOC water
DOC (mg/L)	12.9 ± 0.1	48.5 ± 1.3	108.5 ± 0.2
TOC (mg/L)	17.7 ± 0.6	53.2 ± 0.8	115 ± 1.1
pH	7.24 ± 0.26	7.03 ± 0.43	6.92 ± 0.50
Turbidity (NTU)	5.73 ± 0.34	9.45 ± 0.49	150.0 ± 5.9
Conductivity (μS/cm)	951 ± 10	1250 ± 8	1300 ± 12
Zeta potential (mV)	n.d.	−15.1 ± 4.8	n.d.

n.d. – not determined

was determined using a UV–Visible UVWin5 Software V5.0.5 spectrophotometer T60, at 600 nm and a medium speed scan of 2 nm/s. The transmittance was measured immediately after stirring the suspensions. Distilled water was used as a blank.

Potentiometric titration was used to determine the charge density, CD and degree of substitution, DS, of the different cNFC. For the CD determination, a given mass of cNFCs suspension (corresponding to 200 mg dry material) was dispersed in 150 mL of distilled water and the pH of the suspension was adjusted to 11 with a 0.1 M NaOH (aq) solution, to convert the cationic quaternary ammonium groups to their OH$^-$ form. The suspension was then titrated at 35 °C with 0.01 M HCl until the inflection point of the pH versus HCl volume curve was reached [20]. The CD value was subsequently estimated using Eq. (1).

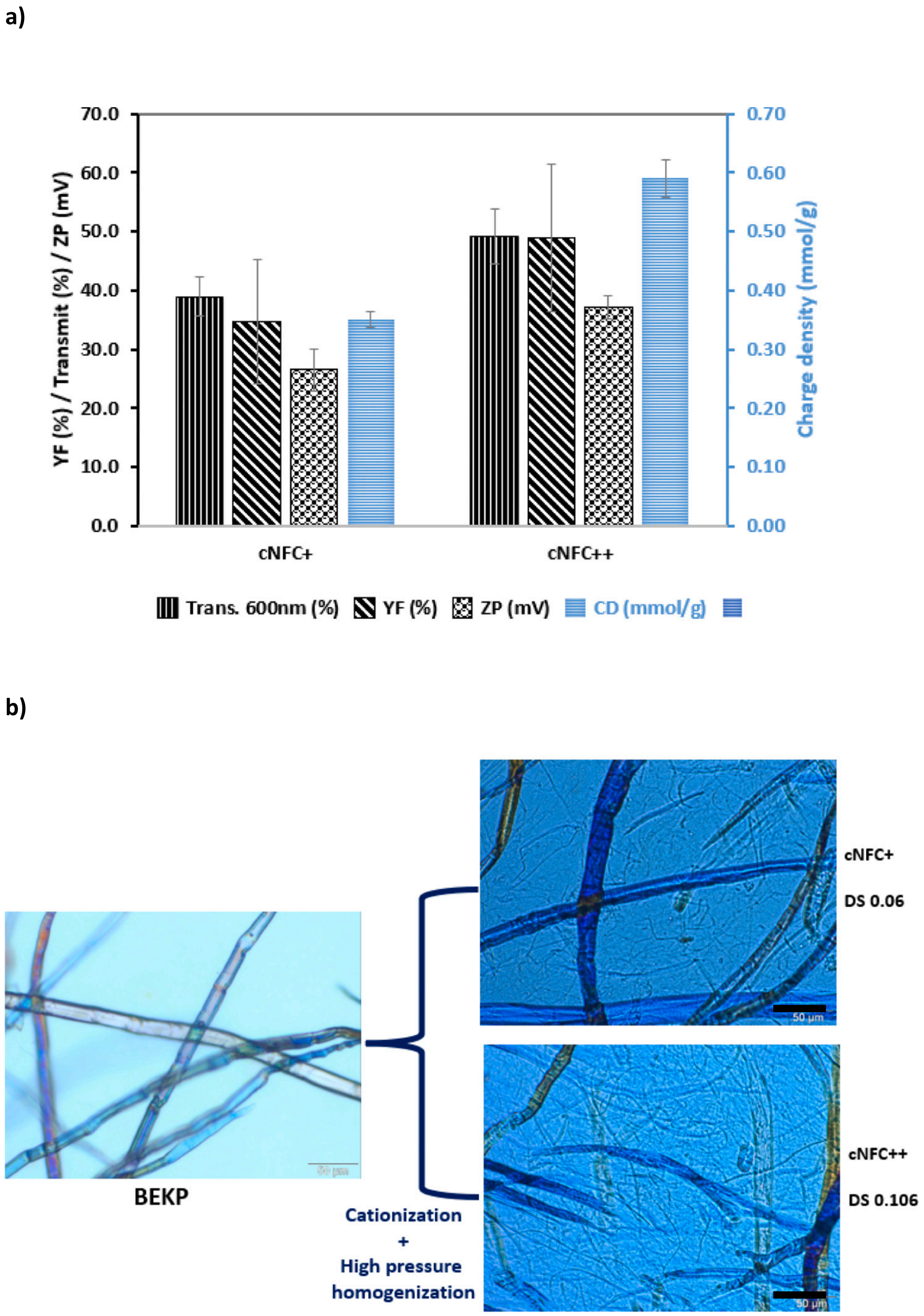

Fig. 1. a) Yield of fibrillation (YF), zeta potential (ZP), cationic charge density (CD), and transmittance of the cNFC produced. b) Optical microscopy images of the starting cellulose fibres (BEKP) and the produced samples of cNFC. The scale bars represent 50 μm.

$$CD = \frac{(V_2 - V_1) \times C_{HCl}}{m_{dry}} \tag{1}$$

where V_1 and V_2 are the volumes (in mL) obtained by the interception of the regression of the lines plotted on the first derivative of the pH versus HCl volume curve, C_{HCl} is the concentration in mmol/mL of the titrant solution and m_{dry} is the mass of cationic cellulose in g. Once CD was obtained, the DS was estimated via Eq. (2) [20].

$$DS = \frac{mol_{sub}}{mol_{AGU}} = \frac{(V_2 - V_1) \times \frac{C_{HCl}}{1000} \times Mw_{AGU}}{m_{dry} - (V_2 - V_1) \times \frac{C_{HCl}}{1000} \times \left(Mw_{sub} - Mw_{lg}\right)} \tag{2}$$

where Mw_{sub} is the molecular weight of substituent cationic group, Mw_{lg} is the molecular weight of the living group, and Mw_{AGU} is the molecular weight of the anhydroglucose (162 g/mol).

To infer the disintegration degree of cellulose fibres, some drops of the cellulose suspensions (BEKP and cNFCs) were placed on glass slides and dried at room temperature. Polarized light optical microscopy images were acquired using an Olympus BH-2 KPA microscope (Olympus Optical Co., Ltd) equipped with a high-resolution CCD colour camera (Olympus ColorView III). Images were processed with the analySIS software (Soft Imaging System GmbH).

2.3. Sampling and characterization of wastewater

Domestic wastewater samples were collected in a conventional wastewater treatment plant located in the Algarve region, south of Portugal, which does not have a C/F process. The grab sampling technique was applied after preliminary treatment. Wastewater samples were used as collected and stored at 4 °C if not possible to use immediately.

Samples were collected in three different periods, which correspond to different water characteristics (Table 1). The waters were classified according to dissolved organic carbon (DOC) content. Wastewater samples were characterized for DOC, total organic carbon (TOC), turbidity, pH, and conductivity (Table 1) using analytical methods described in Standard Methods [21] in triplicate. All samples were analysed for DOC (TOC-5000, Shimadzu, Japan), turbidity (HACH 2100 N, USA), conductivity (Crison GLP 32 conductivity meter, Spain), and pH (Crison Basic 20+, Spain). DOC was determined in filtered samples through a 0.45 μm acrodisk filter and analysed immediately. Zeta potential was measured before and after C/F experiments for medium DOC water. Measurements were performed in a Zetasizer Nano ZS analyser (Malvern Instruments Inc., UK) by electrophoretic light scattering (ELS), as previously described [22].

2.4. Coagulation/flocculation experiments

Coagulation/flocculation (C/F) experiments were made in a laboratory jar-test equipment (Flocumatic, Selecta, Spain) using 800 mL of domestic wastewater. The equipment has four paddles for 1 L capacity beakers (Fig. S3, in supplementary information). The operational conditions used in the coagulation/flocculation/sedimentation process (C/F/S) were the common conditions used in WWTP in these operations, namely a rapid mixing gradient of 743 s^{-1} (200 rpm) for 2 min, a slow mixing gradient of 24 s^{-1} (20 rpm) for 15 min, and a settling time for 20 min [23].

Cationic NFCs were used as coagulant/flocculants in C/F experiments. Three concentrations were tested for each coagulant, namely 30.3, 60.6, and 92.5 mg/L for cNFC++ and 172.1, 241.2, and 310.4 mg/L for cNFC+. These concentrations were defined previously in screening trials using a model wastewater (data not shown). Once these cNFC concentrations were added to the wastewaters, the trials started, first at a rapid mixing gradient and after 2 min at a slow mixing gradient. After 15 min, the paddles stopped and sedimentation started. After sedimentation, samples were collected and analysed for DOC, turbidity, pH, and conductivity, using Standard Methods [21]. For comparison, the same experiments were also performed using $FeCl_3$ as the coagulant. The applied doses covered the range used with cNFC. All experiments were performed in duplicate.

2.5. Laser diffraction spectroscopy flocculation tests

To access the performance of cNFCs and $FeCl_3$ to flocculate particles in wastewaters, the evolution of the particle size as a function of time was accessed using laser diffraction spectroscopy (LDS) in a Mastersizer 2000 device (from Malvern Instruments) equipped with the Hydro 2000 module [24].

In a typical measurement, ca. 750 mL of wastewater was added in a 1 L beaker, the pump speed was set at 1200 rpm and the suspension was initially sonicated for 1 min to disaggregate any preformed particle. Then, the measurement was started and the particle size evolution was recorded. Note that the coagulants were added 2 min after starting the assay at a concentration of 92.5 mg /L. The vigorous agitation continued for more 2 min. Then, the speed was reduced to 200 rpm to allow the flocculation of the suspended particles in wastewaters. The flocculation process was monitored for 25 min.

The structural information (i.e., mass fractal dimensions, dF, and scattering exponent, SE) of the formed flocs was obtained by outputting the raw data of the LDS equipment to an external spreadsheet (provided by Malvern Instruments), for the offline analysis of the data and for computing the angle and the intensity of light at each detector. The data was further computed into log-log plots of scattered light intensity (see Fig. S1 as typical data in supplementary information).

3. Results and discussion

3.1. Characterization of the cationic cellulose nanofibrils (cNFC)

As mentioned in the experimental section, two cationic cellulose nanofibres samples were prepared (i.e., cNFC+ with a DS of 0.06, and cNFC++ with a DS of 0.106), being the main characteristics presented in Fig. 1a). The cationization was probed by FTIR, and the spectra of the original BEKP and produced derivatives (cNFC+ and cNFC++) are depicted in Fig. S2 (in supplementary information). Overall, the spectra recorded for BEKP, cNFC+ and cNFC++ do not display large differences due to the low degree of modification. However, it is possible to observe a slight increase in absorbance at the typical regions of the methyl groups of the quaternary ammonium groups, particularly at 1638 cm^{-1} (peak a) and a "shoulder" at 1473 cm^{-1} (peak b) which might be related to the absorption of water due to the increase in hydrophobicity [29,30]. It is important to highlight that these derivatives were extensively characterized by us and, apart from FTIR, the success of the modification protocol was confirmed by potentiometric titration and elemental analysis [20,26]. The charge density observed in the samples is different (i.e., 0.35 mmol/g for cNFC+, and 0.59 mmol/g for cNFC++), as a result of different conditions used for cationization. This difference in the content of cationic groups also has an impact on the yield of fibrillation (ca. 35 % for cNFC+ and ca. 50 % for cNFC++), because a higher density of charged groups introduced in the cellulose backbone leads to a higher swelling and, consequently, to an easier exfoliation of the nanofibres from the cellulose fibres during high pressure homogenization, i.e., enhance the fibrillation of cellulose in line with the previous results on cellulose cationization [20]. These results are similar to those obtained with TEMPO oxidation of cellulose fibres [31]. Note that the higher level of cationic groups in the cNFC++ sample yields a more positive zeta potential, being the difference between the two samples of ca. 10 mV. A larger fibrillation degree means that the level of large fibres in suspension is reduced. Therefore, since the number of large fibres in suspension is lower, the scattered light from such large fibres is reduced and, consequently, the transmittance of the samples of higher fibrillation degree is superior (i.e., 49.1 % for cNFC++ and 39.0 % for cNFC+).

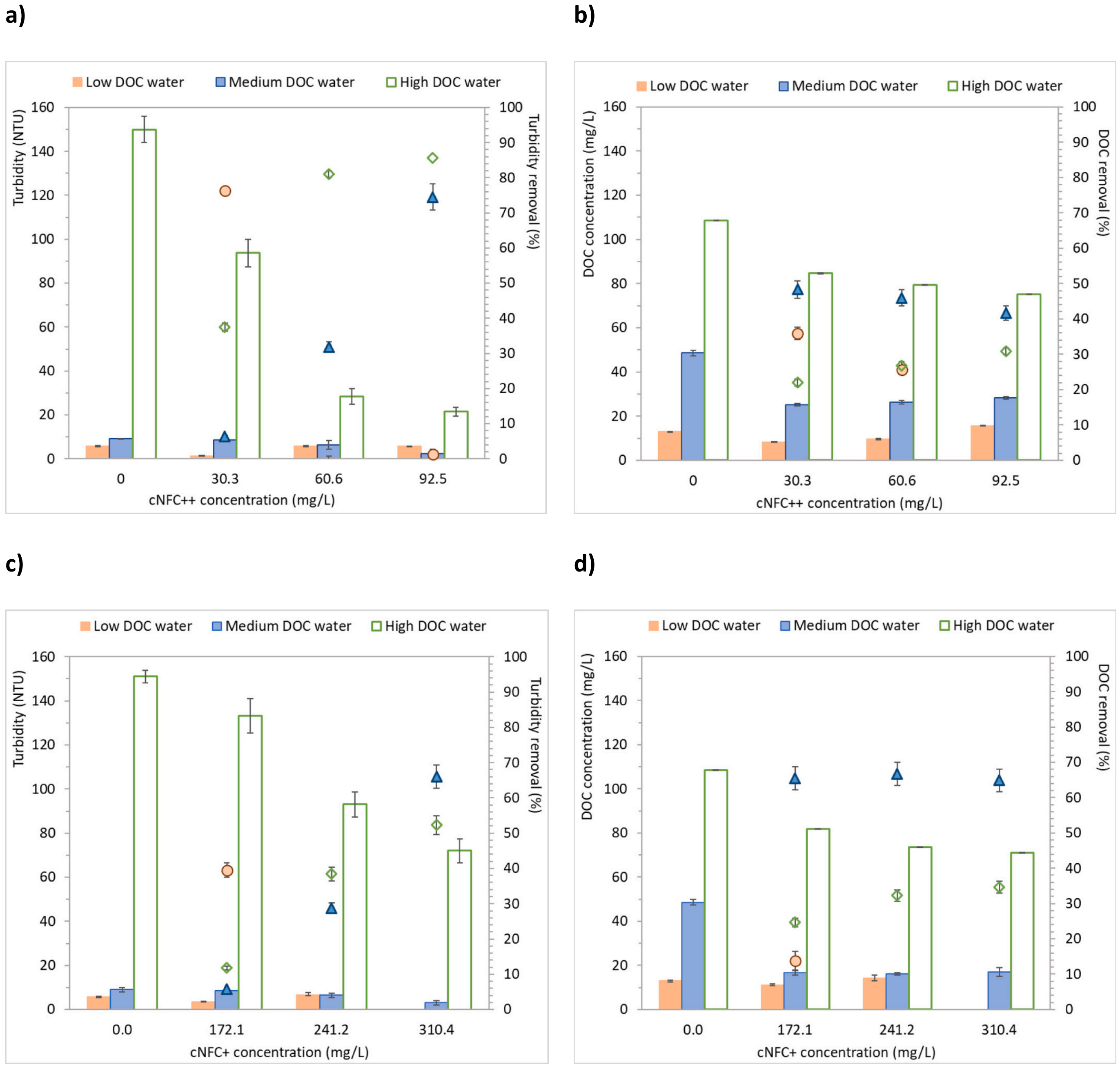

Fig. 2. Turbidity (a,c) and DOC (b,d) of treated water and removal using cNFC++ (a.b) and cNFC+ (b,d) coagulant (Bars: treated wastewater quality; Symbols: removal). No variations of pH were noticed with cNFC addition (n = 2).

These results are supported by optical microscopy (Fig. 1b), where it is possible to observe in the sample with higher DS a smaller number of large cellulose fibres and the presence of more thin cellulosic fibrils. Despite the intense mechanical treatment applied to obtain the cNFCs, no solubilization was detected. Therefore, the soluble fraction (SF) obtained for both samples is considered null, even for the sample with higher DS and yield of fibrillation, indicating no extensive depolymerization of cellulose chains.

3.2. Treated water quality and removal

Fig. 2 shows that, depending on the type of water, both cNFC++, and cNFC+ have an impact on water quality. For low DOC water, turbidity of the treated water shows a decrease for the lowest NFC dose applied in each case (i.e., 30.3 mg/L for cNFC++ and 172.1 mg/L for cNFC+), but an increase for higher flocculant concentrations. However, medium and high DOC waters show a decrease in treated water turbidity with increasing cNFC concentrations (Fig. 2a and c). These results may be attributed to the water DOC content and the nature of NFC fibrils. Low DOC water has both low turbidity and DOC (i.e., 5.73 ± 0.34 NTU and 12.9 ± 0.1 mg C/L). Thus, when using higher concentrations of flocculant, the cNFC long fibrils in excess can remain in suspension, since no charge neutralization or entrapment occurs between cNFC and colloidal particles. However, medium and high DOC waters present sufficient charges to be neutralised, so the higher the cNFC concentrations, the higher the removal efficiency, even for medium DOC waters with similar initial turbidity to low DOC water (Table 1). These observations are in line with those of Edzwald [32] who concluded that, for many waters, natural organic matter controls coagulant dosages instead of turbidity. For the medium and high DOC waters, the highest turbidity removal was

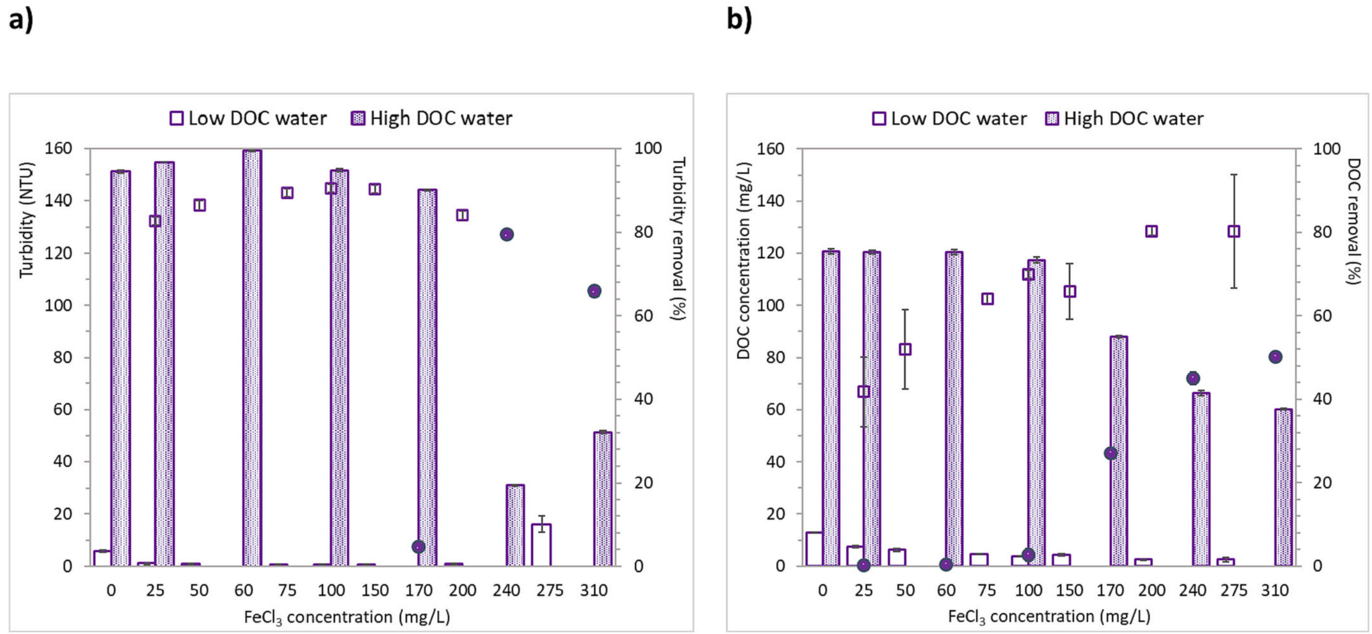

Fig. 3. Turbidity (a) and DOC (b) of treated water and removal using FeCl$_3$ coagulant (Bars: treated wastewater quality; Symbols: removal; n = 2).

obtained with the highest coagulant dose tested (i.e., 74.5 % ± 5.9 and 85.7 % ± 5.9 using 92.5 mg/L cNFC++, and 66.0 % ± 1.0 and 52.3 % ± 2.1 using 310.4 mg/L cNFC+, respectively, Fig. 2a and c). DOC measurements are in accordance with the above observations (Fig. 2b and d). When using low DOC waters (initial concentration of 12.9 ± 0.1 mg C/L), DOC is removed but only using the lowest concentrations of both cNFC++ and cNFC+ (Fig. 2b and d). On the other hand, for medium and high DOC waters there is a decrease in DOC concentrations in treated water with cNFC which was dose-independent for medium DOC water and increased with added coagulant for high DOC water. Removals varied between 48.3 ± 1.1 % and 41.6 ± 1.1 %, and 22.1 ± 1.3 % and 30.8 ± 1.2 %, respectively, for medium and high DOC waters using cNFC++, while varied between 65.5 ± 1.4 % and 64.9 ± 1.7 %, and 24.7 ± 1.3 % and 34.6 ± 1.2 % using cNFC+. These results represent a novel and remarkable finding since it has been reported that one of the drawbacks of natural coagulants is the increase in the organic load of the treated water [2,8].

When comparing the results obtained using cNFC with those of conventional FeCl$_3$ (Fig. 2 vs. Fig. 3), results show that removals are very similar to the best cNFC++ dose tested for low DOC water. Therefore, for these waters and using 30.3 mg/L of cNFC++ removals are 77.0 ± 1.0 % for turbidity and 35.9 ± 2.6 % for DOC, while using 25 mg/L of FeCl$_3$ removals are 82.7 ± 1.9 % and 41.6 ± 8.2 %, respectively (Fig. 1a and b vs. Fig. 2). For high DOC water, the results are slightly different because low FeCl$_3$ concentrations do not remove turbidity but increase it (Fig. 3a). Only from the concentration of 170 mg/L of FeCl$_3$, some removal of turbidity was observed (i.e., 4.73 ± 1.1 %, DOC removal 27.1 ± 1.1 %). When using 92.5 mg/L of cNFC++, the turbidity removal is 85.7 ± 2.8 % and DOC 30.8 ± 1.2 %. In this case, the best FeCl$_3$ concentration for both turbidity and DOC removal is 240 mg/L.

3.3. Mechanisms of coagulation/flocculation with NFC

The coagulation/flocculation performance of the NFCs is dependent on the surface chemical groups. According to Pedrosa et al. (2022), the original BEKP pulp has a negative zeta potential (−23 mV) that shifted to positive (from +24 to +30 mV) upon cationization, which clearly indicates the effective incorporation of positive charges into the cellulose fibres. Therefore, cationized cellulose fibbers when added to wastewater may interact electrostatically with negatively charged particles and, consequently, induce the coagulation/flocculation of suspended particles. Since cNFC++ has a higher degree of substitution than cNFC+, it presents more charged groups and higher absolute zeta potential values (Table 2). Therefore, cNFC++ presents more active sites to coagulate the suspended particles.

Table 2
Zeta potential of the medium DOC water after C/F (pH ≅ 7.03 ± 0.43) (*n* = 5).

cNFC++		cNFC+	
Coagulant dose (mg/L)	Zeta potential (mV)	Coagulant dose (mg/L)	Zeta potential (mV)
30.3	−13.6 ± 0.9	172.1	−11.4 ± 1.2
60.6	−4.6 ± 0.3	241.2	−6.3 ± 1.6
92.5	+2.1 ± 0.7	310.4	−4.4 ± 0.8

The zeta potential of raw medium DOC wastewater before (Table 1) and after the C/F process (Table 2) shows a continuous increase with the addition of the cNCF. It remains negative with cNFC+ in all tested doses, but at the highest cNFC++ dose it becomes positive. Therefore, the cationic coagulant interacts with the oppositely charged particles neutralising their surface charge [33,34]. As cNFCs were used as coagulants, they acted as a destabilizing agent via a charge neutralization-precipitation mechanism, therefore promoting floc nucleation and growth (Fig. 4). Quinlan et al. [17] studied the use of quaternized cationic nanocelluloses to remove an anionic azo dye from an aqueous solution. The authors observed that flocs were formed instantaneously when cationic nanocelluloses with the highest charged density were added to the solution, due to the strong electrostatic interaction resulting in charge neutralization and removal of the dye. This mechanism can explain the different results obtained between low, medium, and high DOC waters. Low and medium waters have similar turbidity, but medium DOC water has a higher DOC concentration than low DOC water (Table 1).

From the data in Table 2, the highest turbidity removal would be expected to occur at a dose between 60.6 and 92.5 mg/L for cNCF++, where the point of zero charge (PZC) would be attained. The highest turbidity removal for the former dose relative to the latter also correlates with the absolute values of zeta potential (4.6 ± 0.3 and 2.1 ± 0.7 mV, respectively). On the other hand, in the experiments with cNCF+, zeta potential has not reached the PZC with the used doses. Therefore,

Fig. 4. Flocs formation using different concentrations of cationic cNFC++.

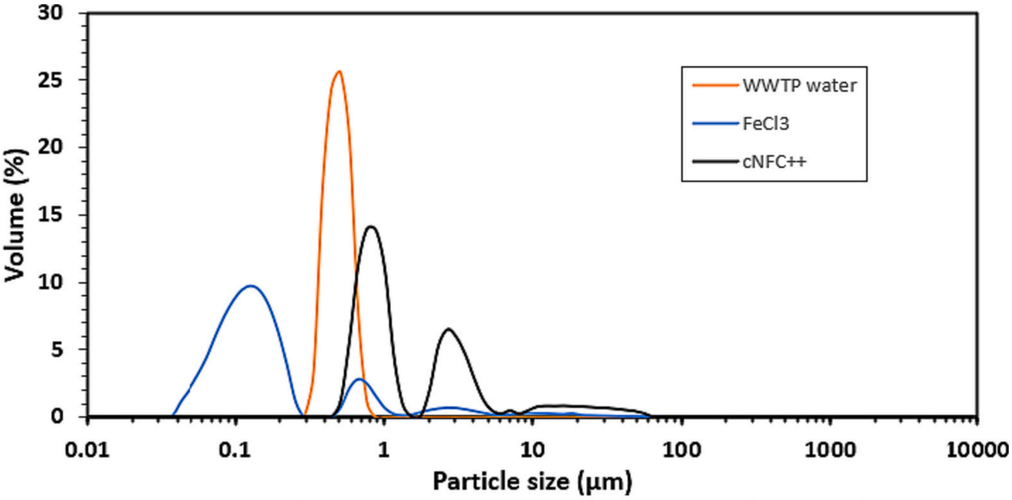

Fig. 5. Laser diffraction spectroscopy analysis of water from WWTP, without treatment (WWTP water – orange line), treated with 92.5 mg/L of $FeCl_3$ ($FeCl_3$ – blue line), and treated with 92.5 mg/L of cNFC++ (cNFC++ – black line). (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

maximal removal would be anticipated at a dose higher than the highest assessed in this trial. These results are consistent with the highest charge density presented by cNCF++ relative to cNCF+.

3.4. Laser diffraction spectroscopy flocculation tests

LDS is a useful technique to follow the nucleation and growth of the aggregates during a flocculation process. Besides the particle size evolution, LDS also allows inferring the structure of the formed flocs using mass fractal dimensions (dF) analysis [25]. The dF parameter can be used as an indicator of the fluffiness/density of the formed flocs because it can be related to the degree to which primary particles fill the space within the nominal volume occupied by an aggregate [26]. Often, a dF value close to 1 indicates a stringier and less dense floc, while a value nearly 3 suggests stronger and denser flocs [25]. According to the Rayleigh–Gans–Debye (RGB) theory, the dF can be calculated from the negative slope of the log-log plot of the scattered light intensity versus the scattering wave vector (q), Fig. S1 (in supplementary information) [24]. The RGB theory is valid if the assumption that the elementary units scatter light independently, being thus more appropriate for spherical particles with low refractive index and sub-micron size [27], and applicable when the flocs size is significantly larger than primary particles and smaller than flocs aggregates [28]. For systems where secondary aggregation, resulting from the aggregation of primary flocs, is observed, the information about the secondary flocs is provided by the scattering exponent (SE) [27]. The SE corresponds to the negative slope of the log-log plot of the scattered light intensity, versus q at large length-scales (low diffraction angles), Fig. S1 (in supplementary information).

The effect of the addition of cNFC++ and $FeCl_3$ (i.e., 92.5 mg/L of each coagulant) to domestic wastewater (low DOC water) was also followed by LDS (Fig. 5).

The starting wastewater presents particles in suspension with ca. 0.5 μm diameter, narrow particle size distribution, and without the presence of large aggregates of primary particles. The addition of $FeCl_3$ enhances the obscuration of the solution, also observed in the formation of larger particles, in the range of 0.8 to 10 μm. On the other hand, smaller particles were also observed probably due to dispersion induced by the presence of the different ions from the ferric salt, which could act as stabilizer of primary particles.

Conversely, the addition of cNFC++, at the same dosage, leads to an increase in particle size, observed in the presence of secondary particles

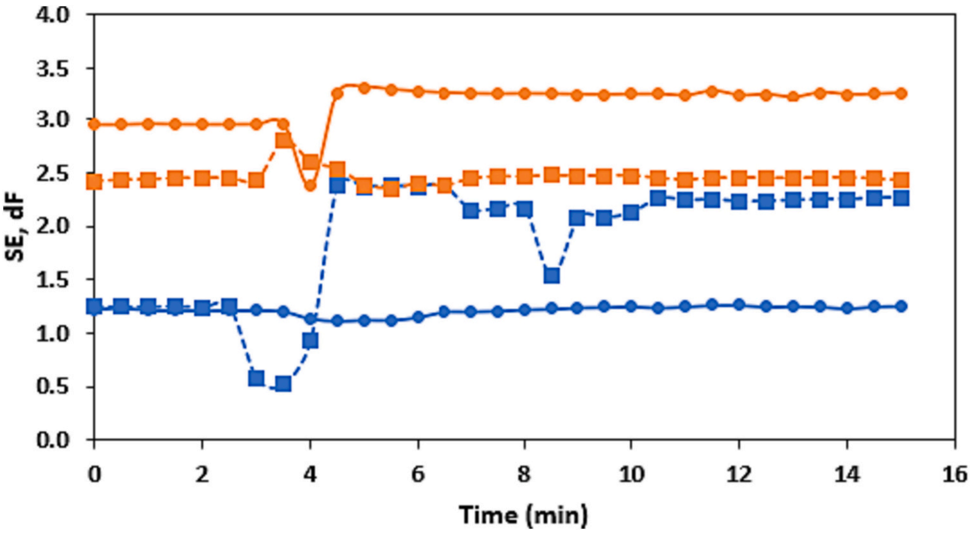

Fig. 6. Evolution of the scattering exponent (SE) and mass fractal dimension (dF) as a function of flocculation time for the different coagulants ($FeCl_3$ and cNFC++). SE (cNFC++) – blue circles; dF (cNFC++) – orange circles; SE ($FeCl_3$) – blue squares; dF ($FeCl_3$) – orange squares. (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

(resulting from the aggregation of primary particles) with dimensions in the range of ca. 0.8 to 50.0–60.0 μm. This larger increment in particle size could be explained by the fact that cNFC++ not only neutralizes the negative charges of the primary particles but can also induce “bridging” among primary particles, resulting in aggregates of larger size and, consequently, improving the removal of contaminants (particles in suspension) from wastewaters.

In addition to the particle size dependence on the coagulant used, the mass fractal dimensions (dF and SE) of the flocs formed were calculated from the scattering data (Fig. 6).

As mentioned before, the dF and SE parameters provide insights into the structure (density) of the primary and secondary aggregates, respectively. The flocs formed by the addition of $FeCl_3$ present a lower density for the primary aggregates (dF is lower when $FeCl_3$ is added in comparison to the addition of cNFC++). On the other hand, the opposite trend is observed for the secondary flocs, which result from the agglomeration of the primary flocs. Denser primary aggregates seem to be as important to reach better stability of the flocs formed, thus leading to good results in the removal of suspended particles of wastewaters when cNFC++ is used as flocculants, which is a new and major finding.

4. Conclusions

Cationic cellulose nanofibres were developed with different charge densities and applied in wastewater treatment. As hypothesized, these nanofibres efficiently coagulate and flocculate suspended particles from wastewaters with different characteristics, without increasing the DOC of the treated waters. Removals such as 74.5 % and 85.7 % using 92.5 mg/L cNFC++, and 66.0 % and 52.3 % using 310.4 mg/L cNFC+, respectively for medium and high DOC waters, were obtained for turbidity. For the same waters, removals varied between 48.3 % and 41.6 %, and 22.1 % and 30.8 %, respectively, waters using cNFC++, while varied between 65.5 % and 64.9 %, and 24.7 % and 34.6 % using cNFC+, respectively. In addition, a better performance than with a commercial coagulant was obtained. The main removal mechanism using cNFCs was charge neutralization, that promotes floc nucleation, growth and sedimentation. However, it can also induce “bridging” among primary particles, resulting in aggregates of larger size and, consequently, improving the removal of contaminants (particles in suspension) from wastewaters. The flocs formed by the addition of $FeCl_3$ present a lower density for the primary aggregates.

This work proved that it is possible to use natural-based cNFCs as coagulant/flocculants, without the need of adding other chemicals, to treat real domestic wastewaters. This is a major contribution to the sustainability of wastewater treatment, reducing the use of chemicals that usually have negative impacts on the environment and human health. The next steps should optimise the operational C/F conditions and test the technology at a higher scale, a step forward the scaled up to industrial/field scale.

CRediT authorship contribution statement

Margarida Ribau Teixeira: Conceptualization, Funding acquisition, Methodology, Supervision, Writing – original draft, Writing – review & editing. **Abdullah Ismail:** Investigation. **Bruno Medronho:** Conceptualization, Funding acquisition, Methodology, Writing – review & editing. **Luís Alves:** Conceptualization, Funding acquisition, Methodology, Writing – review & editing. **Jorge F.S. Pedrosa:** Investigation. **Paulo J. T. Ferreira:** Investigation. **Vânia Serrão Sousa:** Investigation, Writing – review & editing. **Ana M. Rosa da Costa:** Conceptualization, Methodology, Writing – review & editing.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The authors are unable or have chosen not to specify which data has been used.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jwpe.2024.104817.

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