

# Functionalization of Microcrystalline Cellulose through Integrated Sodium Periodate and TEMPO/Ozone Oxidations

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**Abstract:** Cellulose modification has been a primary focus in developing bio-based degradable functional materials. In this study, an integrated approach combining sodium periodate ( $\text{NaIO}_4$ ) and TEMPO (2,2,6,6-tetramethylpiperidin-1-oxyl)-ozone oxidation process is employed to functionalize microcrystalline cellulose (MCC). The resulting materials in water-soluble and precipitate fractions were characterized to understand the influences of each oxidation method on the reaction mechanisms, structures, and physicochemical properties of the oxidized MCC samples. The results demonstrated that the combination of both methods resulted significantly in chemical structures, morphology, and surface reactivity of the samples. FTIR spectra showed an appearance of oxidized functional groups, i.e., carboxyl, ketone, and aldehydes, confirming a successful oxidation process. The use of ozone as a co-oxidant contributes positively to environmental aspects and process economics due to its availability and low cost. These findings illustrate the considerable potential of ozone utilization in improving cellulose properties for various applications. Consequently, the integrated approach offers an effective and sustainable solution for enhancing the quality and performance of cellulose, paving the way for further research and applications in the field of cellulose materials.

**Keywords:** Microcrystalline cellulose, TEMPO, Periodate, Ozone, Oxidation process

## 1. Introduction

Cellulose, a predominant and versatile biopolymer, plays a significant role in the global economy. It serves as a fundamental resource for human needs, ranging from the construction of buildings to the production of clothing and paper. The material and its derivatives contribute to renewable energy, the food industry, cosmetics, and medical applications. Given its relative affordability and potential, cellulose is a valuable resource. However, a key limitation of cellulose is its insolubility in common solvents, which restricts its use in several applications [1] due to its strong inter and intramolecular hydrogen bonding and crystalline structure. Therefore, it is essential to enhance the properties of cellulose through chemical modifications. This will expand its utility and make it more suitable for a broader range of applications.

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The oxidation reaction of cellulose has attracted attention from many research groups, as it can produce various products based on the reaction site and the reagents used, suitable for different industrial purposes [2,3]. Selective oxidation is a reaction that introduces oxygen atoms into specific molecule positions without affecting the rest of the structures. The reaction is useful for synthesizing new compounds with different properties and functions from the original molecule [4].

Cellulose is frequently modified through oxidation to enhance its suitability for various medical, pharmacy, and industry applications. Different cellulose types, reagents, and conditions can be selected to produce various oxidation products with distinct properties. Oxidizing cellulose can enhance its surface charge density, reducing the energy required to separate fibers into nanocellulose fibrils. The oxidation reactions employing 2,2,6,6-tetramethylpiperidine-1-oxyl (TEMPO) or periodate are advantageous due to their selectivity, efficiency, and environmentally friendly nature [5].

TEMPO is a stable aminoxyl radical that selectively oxidizes primary alcohols to aldehydes. In cellulose, TEMPO-mediated oxidation transforms the abundant primary hydroxyls on each cellulose microfibril surface into negatively charged carboxylate groups. This conversion offers several benefits, especially improved dispersibility and enhanced reactivity. The negative charges from the oxidation induce strong electrostatic repulsion, facilitating easy and fast fibrillation and producing well-dispersed individual nanofibers. Additionally, the carboxylate groups increase the reactivity of cellulose, making it easier to modify and functionalize for various applications [6]. These attributes make TEMPO-oxidized cellulose a promising material for diverse applications, including producing cellulose nanocrystals, bio-based composites, biomedical reagents, and efficient dispersing agents. Most importantly, its ability to create high-value products from renewable resources drives the growing research interest in TEMPO-oxidized cellulose.

Using TEMPO as a selective oxidizing agent for primary hydroxyls in cellulose marks a significant advancement in cellulose oxidation. TEMPO selectively targets the primary hydroxyl groups in cellulose in conjunction with co-oxidants, including sodium hypochlorite (NaOCl) and sodium bromide (NaBr) due to their halogen constituents. However, these co-oxidants raise environmental concerns [7,8]. Ozone, frequently employed for bleaching pulp, lignin removal, and cellulose depolymerization, is a suitable alternative co-oxidant due to its efficient modification of carbohydrates, lower cost than halogen oxidants, non-toxicity, and minimal environmental impact. Additionally, under low acidity conditions, ozone can enhance the solubility fraction of cellulose during extraction [9].

This study investigates the efficiency and mechanisms of microcrystalline cellulose (MCC) oxidation processes, employing TEMPO with ozone as a co-oxidant, in comparison with periodate, and a combined TEMPO/Ozone and periodate mediated oxidation. In the TEMPO/Ozone system, ozone reacts with TEMPO via disproportionation, forming TEMPO<sup>+</sup>, which selectively converts primary hydroxyls to carboxyl groups [8]. Periodate oxidation converts vicinal hydroxyl groups of MCC to aldehyde groups. When the two processes are combined, both reactions occur, and the aldehyde formation enhances the MCC oxidation with TEMPO/Ozone mediation. Insights into the efficiency and mechanisms of these processes are beneficial in optimizing the modifications of MCC for specific applications, especially environmental, packaging, cosmetics, and biomedical fields.

## 2. Methodology

### 2.1 Materials

Microcrystalline cellulose (MCC, average size of 20  $\mu\text{m}$ ) and (2,2,6,6-tetramethyl-piperidin-1-yl) oxyl (TEMPO, 98%) were purchased from Sigma-Aldrich (USA). Sodium periodate (99%) was supplied from Thermo Fisher Scientific. Sulfuric acid and sodium hydroxide were obtained from Carlo Erba Chemical, France.

### 2.2 Sodium periodate oxidation

The oxidation of microcrystalline cellulose using sodium periodate was modified from that reported by Gomez (2007) [10] and Paşcalău et al. (2013) [11]. MCC sample (4 g) was dispersed into 50 mL of ethanol. Sodium periodate (2.6 g in 40 mL distilled water) was added and stirred in dark room conditions for around 6 h (300 rpm, room temperature). After the reaction, 2 mL of ethylene glycol was added to end the reaction by neutralizing excess

sodium periodate. The mixture was further stirred for 30 min. Finally, the resulting solid sample was filtered, washed with ethanol, and dried overnight at room temperature.

### 2.3 Generation of N-oxoammonium cation (TEMPO<sup>+</sup>)

N-oxoammonium cation (TEMPO<sup>+</sup>) was generated from nitroxyl radical (TEMPO) with ozone as a co-oxidant. The process begins with dissolving 0.08 g of TEMPO in 10 mL of deionized (DI) water (0.512 mmol), followed by adding 10 mL of 0.1 M H<sub>2</sub>SO<sub>4</sub> (with a pH of 3-4) and stirring for 5 min before introducing ozone [8,12]. The concentration of TEMPO utilized in this study is based on Le & Opaprakasit (2021) [12], as the maximum solubility of TEMPO in water is approximately 9.7 g/L at 20 °C. Ozone was dispersed using an ozone generator (Enaly series ZO-30 output 200 mg/h from dry air, in the TEMPO solution for 2 h. The amount of TEMPO and TEMPO<sup>+</sup> increased as a function of time in the 2-h disproportionation reaction.

### 2.4 Application of TEMPO<sup>+</sup> in microcrystalline cellulose oxidation

The oxidation procedure was adapted from Le & Opaprakasit (2021) [12]. MCC (0.5 g) was added to the TEMPO<sup>+</sup> solution, and the reaction was conducted for 2 h at 4-5 °C. The solution was adjusted to pH 10 with NaOH to enhance the solubility. Subsequently, the sample underwent dialysis for approximately 2 days with water renewal until the pH became neutral. The resulting products were then centrifuged to separate MCC from the supernatant fraction (6000 rpm, 15 min). The oxidized MCC was collected and dried overnight in an oven at 60 °C for precipitate fraction, while the supernatant fraction was freeze-dried.

### 2.5 Characterization

UV-Vis spectrometer (Thermo Scientific, GENESYS™ 180) was employed to measure the concentrations of TEMPO and the generated TEMPO<sup>+</sup> as a function of ozone dispersion time using the absorbance at 425 and 482 nm, respectively. The data was recorded in the 200-800 nm range with an interval of 0.5.

The chemical structures of MCC samples were analyzed using Fourier transform infrared (FTIR) spectroscopy (Thermo Scientific Nicolet iS5) in an attenuated total reflectance (ATR) mode. The spectra were recorded in the 4000-525 cm<sup>-1</sup> range for 32 scans at a resolution of 4 cm<sup>-1</sup>. Nuclear magnetic resonance (<sup>1</sup>H-NMR) spectroscopy was employed to characterize the structures of MCC and oxidized MCC using Ascend TM 600 MHz/Avance III HD spectrometer, Bruker (Switzerland). Deuterium oxide (D<sub>2</sub>O) was employed as a solvent.

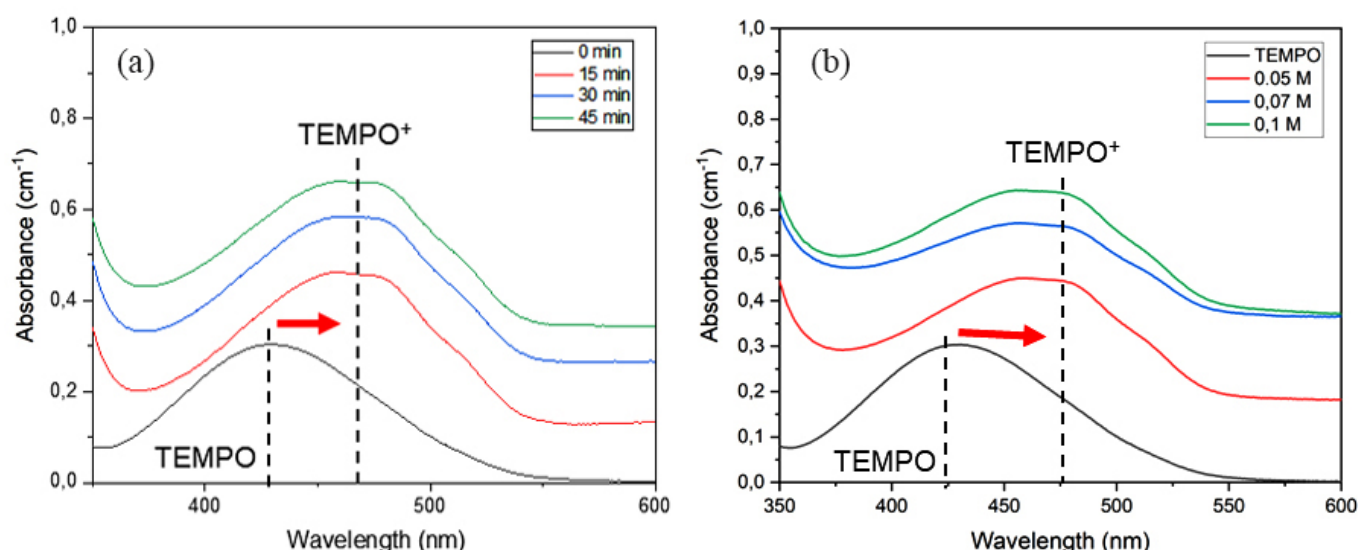
The morphology of MCC and oxidized MCC samples was examined using scanning electron microscopy (SEM) on Hitachi FE-SEM SU5000 (Japan). The sample was coated with a thin layer of platinum before recording SEM images. The samples' Zeta potential, particle size, and particle size distribution were determined using Zetasizer Nano-ZS (Malvern, United Kingdom). The samples were prepared by dispersing them in Milli-Q water.

The potentiometric titration was employed by adapting from that of Tang et al. (2024) to quantify the carboxyl content in MCC samples based on the equilibrium point [13]. The samples (100 mg) were either suspended or dissolved in 20 mL of a 0.01 mol/L hydrochloric acid solution. The titration was conducted using a 0.005 mol/L sodium hydroxide solution delivered via a peristaltic pump. Throughout the titration, the pH of the solutions was recorded at 10-s intervals through a Hanna Instrument HI92000 software. The titration process was concluded when the pH reached 10.

## 3. Results and discussion

TEMPO is a stable nitroxyl radical that is widely used in oxidation reactions. In acidic conditions, TEMPO can undergo a reaction known as disproportionation. This involves 2 TEMPO molecules reacting to form one molecule of the reduced form (hydroxylamine) and one molecule of the oxidized form (oxoammonium cation). The oxoammonium cation of TEMPO, denoted as TEMPO<sup>+</sup>, is the oxidized form of TEMPO. UV-Vis spectra were recorded to observe TEMPO<sup>+</sup> formation during the introduction of ozone, as shown in Figure 1. The TEMPO and TEMPO<sup>+</sup> contents are monitored by the absorbance at 425 and 482 nm, respectively. Figure 1(a) shows UV-Vis spectra of the solution, i.e., the formation of TEMPO<sup>+</sup>, as a function of ozone dispersion time. TEMPO solution without ozone showed a peak at

around 425 nm. When ozone was applied, the peak shifted to around 480-500 nm, whose intensity increased with time. This indicates that ozone successfully converted TEMPO to TEMPO<sup>+</sup>, and the ozonation time strongly correlated with the TEMPO<sup>+</sup> concentration. Because hydroxyl-amine and protonated hydroxylamine are formed at longer reaction times, the effects of acid concentration on the contents of the generated TEMPO were examined. Figure 1(b) shows that the TEMPO<sup>+</sup> concentration increased with the acid concentrations, reflected by the increase in the peak intensity at 480 nm.

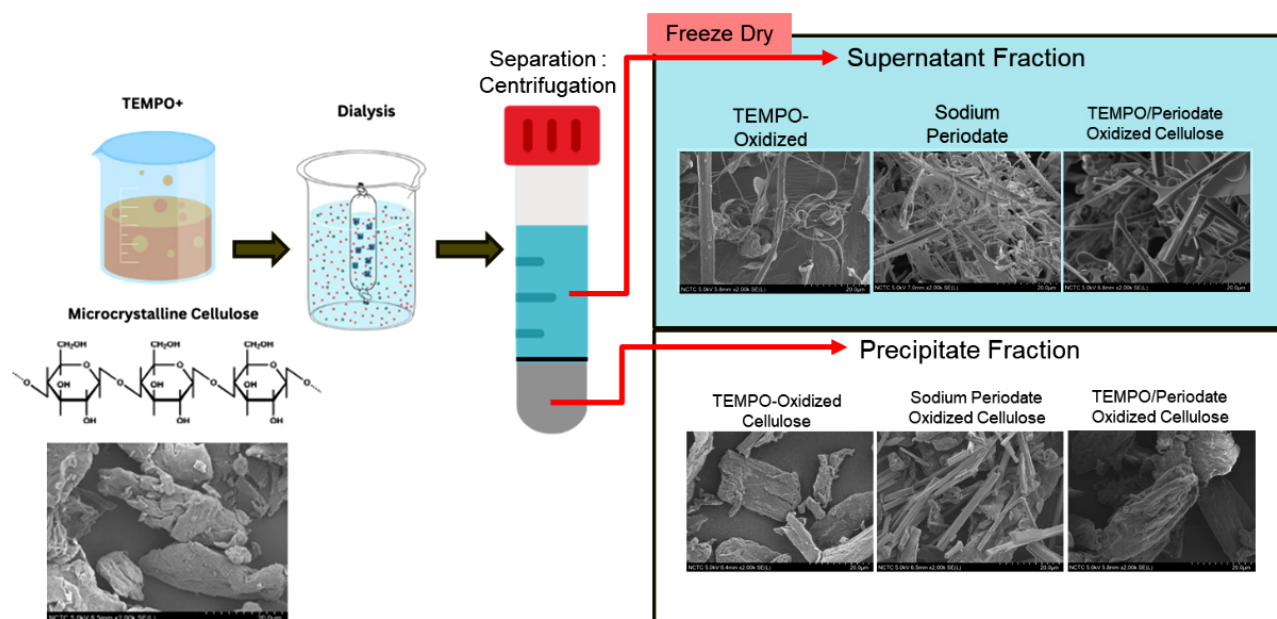


**Figure 1** UV-Vis spectra indicating the conversion of TEMPO to TEMPO<sup>+</sup> as a function of (a) Ozonation time and (b) Acid concentrations.

In TEMPO-mediated reactions, the oxoammonium cation acts as an efficient oxidant for the selective oxidation of primary alcohols, with hydroxylamine (TEMPOH) being a side product. The TEMPO<sup>+</sup> cation exhibits excellent oxidizing reactivity due to its highly positive redox potential and tremendously high kinetics in the redox reaction compared with other redox reagents [12]. Tempo-catalyzed oxidation of polysaccharides enables efficient and selective conversion of primary hydroxy groups in water-soluble and water-insoluble polysaccharides to carboxylate groups under mild conditions. This oxidation system significantly inhibits depolymerization and yields oxidized products that contain no aldehydes [14]. Its unique properties make it a valuable tool in the synthesis of pharmaceutical products, modification of biomass, and the development of high-performance materials [15].

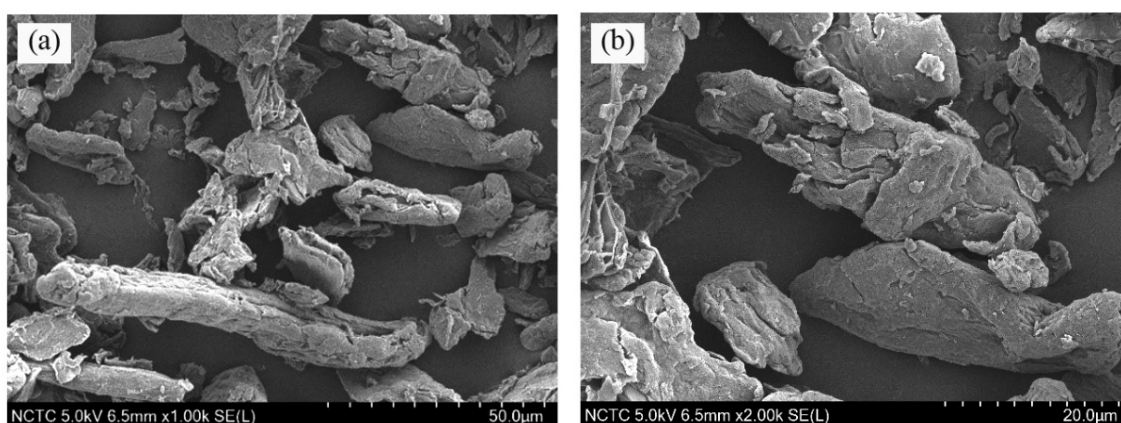
In this study, TEMPO<sup>+</sup> was synthesized using 0.1 M H<sub>2</sub>SO<sub>4</sub> at a 2-h ozonation process. This freshly prepared TEMPO<sup>+</sup> solution was then applied to MCC for oxidation. The initial MCC, with a particle size of 20 μm, exhibited a reduction in size despite the inherent size variability. After the reaction, the mixture was separated as precipitate and supernatant fractions, each displaying unique characteristics attributable to the effects of TEMPO/Ozone-mediated and periodate oxidations. The supernatant fraction consisted of MCC with a higher oxidation degree and smaller particle size. The separation process and SEM images of the products from each fraction are summarized in Figure 2. The results align with Hou et al. (2024) [16], which focused on the structural analyses of supernatant fractions from TEMPO-oxidized pulp/water reaction mixtures separated by centrifugation and dialysis. The authors found that the oxidized cellulose in supernatant fractions had much higher carboxyl contents than those of the precipitate fractions, in which water-soluble β-1,4-polyglucuronic acid was a major constituent.





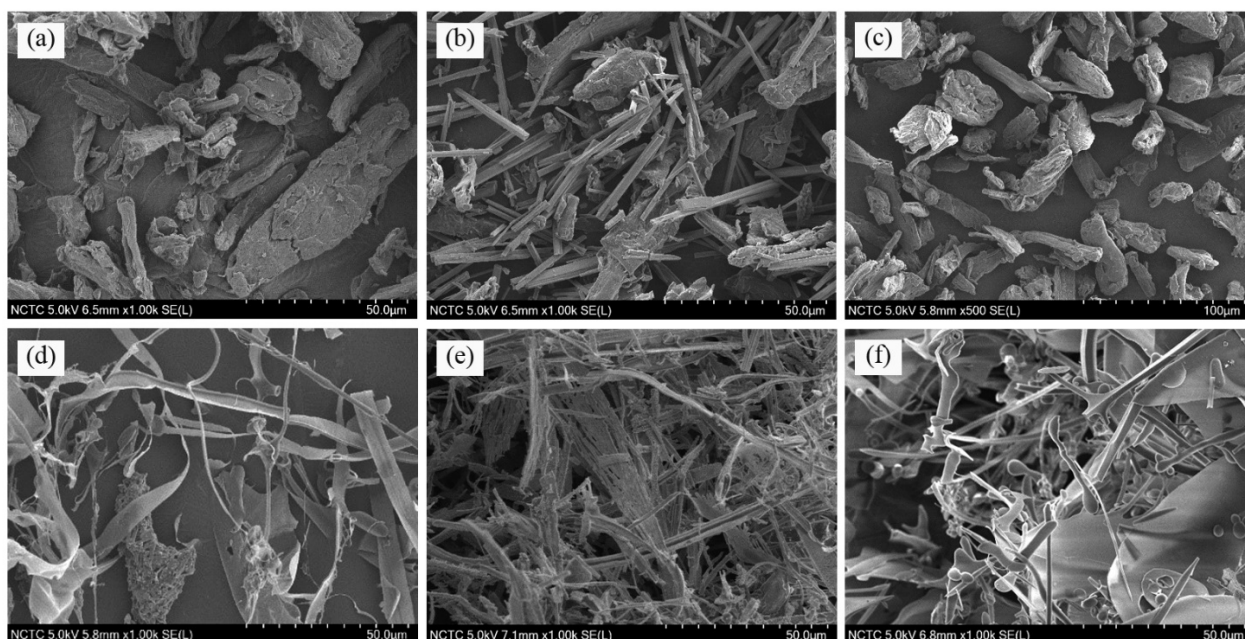
**Figure 2** The separation of precipitate and supernatant fractions of oxidized MCC products and their respective SEM images.

The SEM images of native MCC showed its microstructure before oxidation, as shown in Figure 3. Particles with irregular shapes and rough textures were observed due to the natural formation of cellulose in plant cell walls, which is a semi-crystalline polymer. The size and shape of these particles varied widely, reflecting the natural variability of cellulose sources and the mechanical processes used to isolate the MCC.



**Figure 3** Morphology of microcrystalline cellulose.

The corresponding SEM images of MCC subjected to different oxidation processes are compared in Figure 4. Particles with a higher degree of fiber separation than the native MCC were observed, especially in the supernatant fractions (Figure 4(d), 4(e), and 4(f)), which present as fine, well-separated fibers. The changes in the microstructure are indicative of chemical modifications introduced during the TEMPO/Ozone oxidation. In native MCC, the cellulose fibers are more aggregated and intertwined. After oxidation, the fibers become more individualized and separated, likely due to the introduction of carboxylate groups that increase their dispersibility in water.



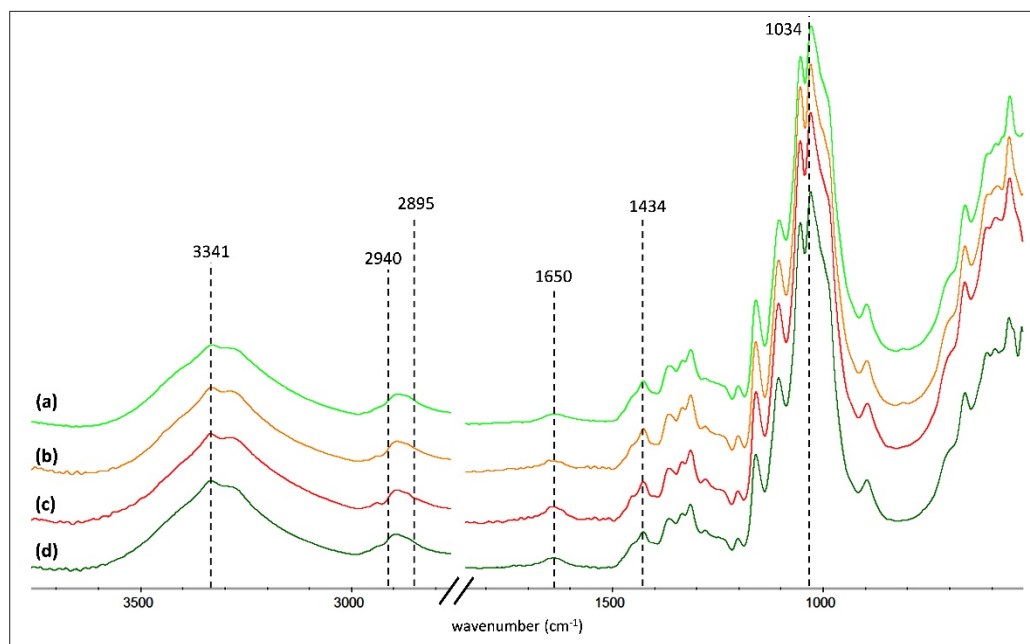
**Figure 4** SEM images showing microstructures of MCC after difference oxidation processes; (a), (d) TEMPO/Ozone-oxidized MCC, (b), (e) Periodate-oxidized MCC, (c), (f) Combination of TEMPO/Ozone and periodate mediated in a separate state, (a), (b), (c) Precipitate fraction, and (d), (e), (f) Supernatant fraction.

Figures 4(a) and 4(d) show the microstructure of MCC after oxidation using TEMPO/Ozone in the precipitate and supernatant fractions, respectively. The precipitate fraction (Figure 4(a)) showed aggregated particles, while the supernatant fraction (Figure 4(d)) displays fine fibers that appear more individualized. This suggests that the TEMPO/Ozone oxidation process can lead to a reduction in particle size and an increase in fiber separation. The precipitate fraction of periodate-oxidized MCC (Figure 4(b)) predominantly showed rod-shaped and thin fibers, while the supernatant fraction (Figure 4(e)) presented fine, web-like fibril structures. This indicates that periodate oxidation can lead to significant fiber elongation and separation. The precipitate fraction of MCC after the combined TEMPO/Ozone and periodate oxidation (Figure 4(c)) showed a mixture of aggregated small particles and some fiber-like structures. In contrast, the supernatant fraction (Figure 4(f)) consisted of fine fibers with some particle aggregation. The results suggest that the combined oxidation process can lead to diverse structural changes and a decrease in the MCC particles' size.

FTIR spectra characterize the sample's chemical structures and functional groups, as shown in Figure 5. Native MCC showed a broad band centered around  $3341\text{ cm}^{-1}$ , attributing to the O-H stretching mode. Strong overlapped bands at  $1034\text{ cm}^{-1}$  are associated with the C-O vibrations of ether linkages [17]. These linkages are responsible for the stability and rigidity of the cellulose structure, which is vital for its function as a structural component in plant cell walls [18]. The bands at  $2940$  and  $2895\text{ cm}^{-1}$  are due to C-H stretching modes, while the broad and weak band at  $1650\text{ cm}^{-1}$  is associated with absorbed water molecules.

The precipitate fraction of MCC, TEMPO/Ozone-oxidized MCC, and the combined TEMPO/Ozone and periodate-oxidized MCC do not exhibit significant changes compared to native MCC. This reflects that oxidations mainly affect the structures of smaller-sized fractions, enhancing their solubility in the supernatant. In contrast, a relatively lower degree of chemical reaction occurs in the bigger-sized fractions, which are precipitated. Nevertheless, there was a slight intensity decrease in the O-H stretching bands for periodate-oxidized MCC. The absorbance bands of periodate-oxidized cellulose below  $1034\text{ cm}^{-1}$  are slightly higher, indicating increased functional groups after oxidation. The sodium periodate oxidation process breaks the cellulose ring by creating two vicinal hydroxyl groups,

resulting in more hydroxyl groups than native MCC. This increase in functional groups suggests enhanced reactivity and modification potential for the periodate-oxidized cellulose [19].



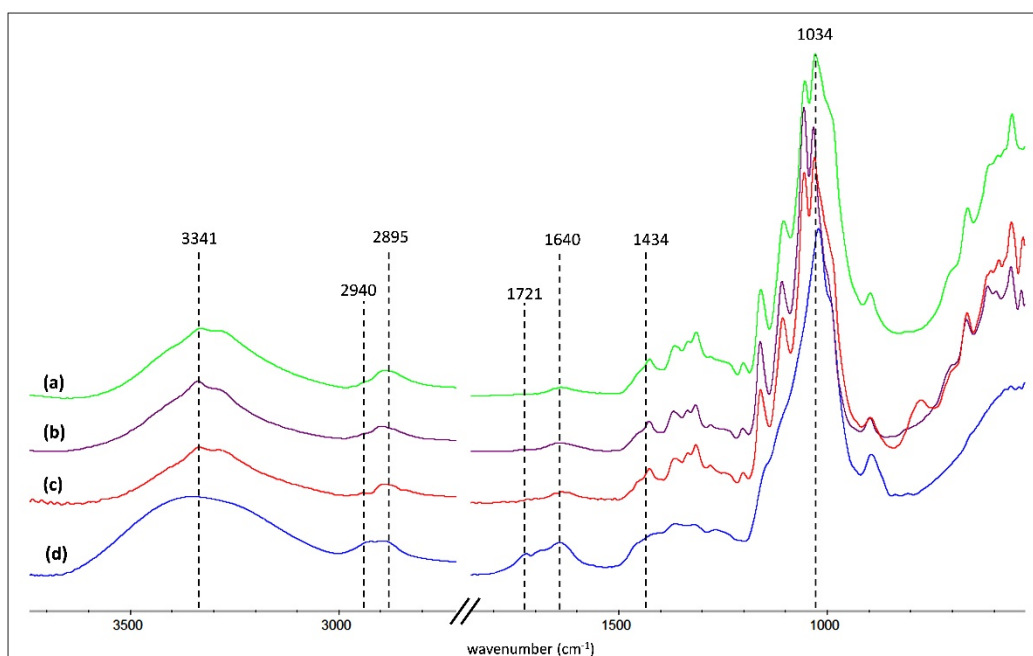
**Figure 5** FTIR spectra of the precipitate fraction of (a) MCC, (b) TEMPO/Ozone-oxidized MCC, (c) Periodate-oxidized MCC, and (d) Combined TEMPO/Ozone and periodate-oxidized MCC.

FTIR spectra of the supernatant fractions of oxidized samples, as shown in Figure 6, exhibit marked differences compared to native MCC and the precipitates from all oxidations. A sharp band at  $1721\text{ cm}^{-1}$ , associated with the C=O stretching of carboxylic acid, appeared, particularly for the one from the combined periodate and TEMPO/Ozone oxidation process. This reflects a high relative content of oxidized functional groups in the supernatant fractions. TEMPO oxidation of cellulose converts hydroxyls to carboxylic groups in the cellulose structures, reflected by an appearance of the C=O's carboxylic acid band. This also leads to an increase in the intensity of the O-H stretching band [20,21]. Sodium periodate oxidation breaks cellulose's rings and generates various oxidized functionalities (aldehyde, keto, and carbonyl), reflected by the bands in the  $1700$  and  $1750\text{ cm}^{-1}$  region [2]. The periodate and TEMPO/Ozone oxidation leads to a combination of the two reaction mechanisms, reflected by more intense bands of the functional groups.

The Zeta potential and average particle size of the supernatant fractions of MCC and oxidized MCC samples were analyzed, as summarized in Table 1 and Figure 7. The results provide information on the impact of the oxidation process on the physicochemical properties of MCC, highlighting the extent of its structural modification. The zeta potential measures the effective charge on a particle in suspension for MCC and its oxidized forms. This plays a crucial role in determining the stability of colloidal dispersions. The sign and magnitude of the zeta potential can provide insights into the surface charge characteristics, which are influenced by the particle surfaces' chemical composition and the surrounding medium's nature. MCC showed a negative zeta potential of  $-13.73\text{ mV}$  due to its hydroxyl groups, which ionize to form negatively charged species in the aqueous medium. The MCC oxidized by TEMPO/Ozone-mediated becomes more negative ( $-32.03\text{ mV}$ ), confirming the selective oxidation of primary hydroxyls to carboxylate groups. These groups ionize in the aqueous medium, increasing the negative charge on the cellulose particles. Periodate-oxidized MCC, however, exhibited a lower negative zeta potential ( $-3.27\text{ mV}$ ). This could be due to the generation of aldehyde groups at the C2–C3 positions of the glucose units. These aldehyde groups can hardly ionize,



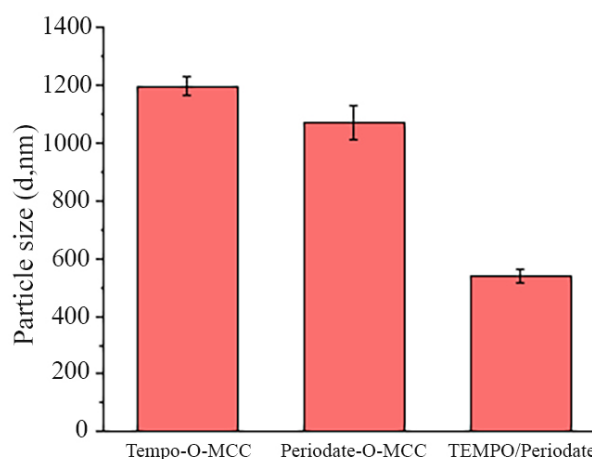
resulting in a less negative zeta potential. When MCC undergoes combined TEMPO/Ozone and periodate oxidations, the zeta potential is -10.77 mV. This suggests that the dual oxidation process balances the functional groups introduced by the individual TEMPO/Ozone and periodate oxidation processes.



**Figure 6** FTIR spectra of the supernatant fraction of (a) MCC, (b) TEMPO/Ozone-oxidized MCC, (c) Periodate-oxidized MCC, and (d) Combined TEMPO/Ozone and periodate-oxidized MCC.

**Table 1** Zeta potential of MCC and oxidized MCC samples

| Sample                | Zeta potential (mV) |
|-----------------------|---------------------|
| MCC                   | -13.73 ± 0.31       |
| Tempo-O-MCC           | -32.03 ± 0.60       |
| Periodate-O-MCC       | -3.27 ± 0.13        |
| TEMPO/Periodate-O-MCC | -10.77 ± 0.38       |



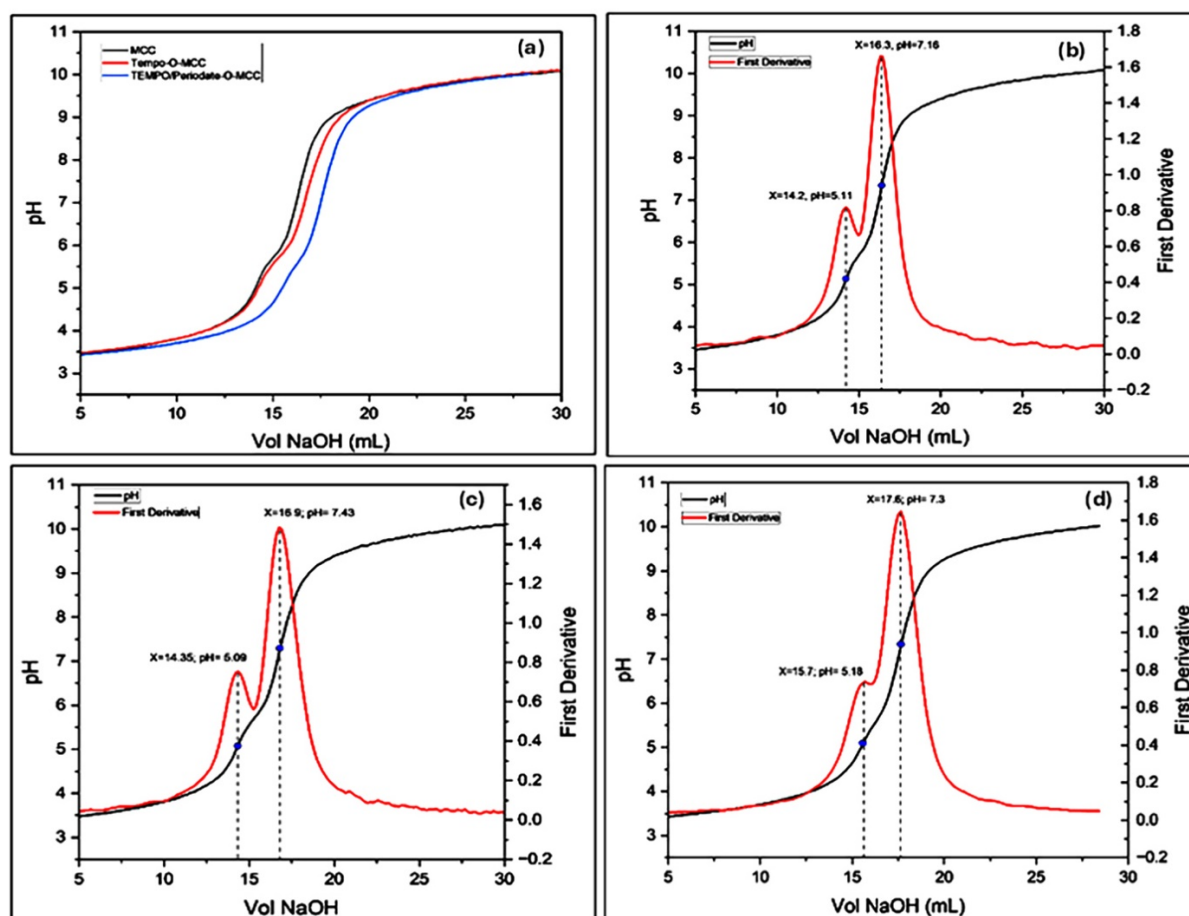
**Figure 7** The average particle size of oxidized MCC derived from different processes using native MCC with an average size of 20  $\mu\text{m}$ .

The average particle size of MCC and oxidized MCC are compared in Figure 7, providing insights into the physical changes induced by these treatments. The particle size of TEMPO/Ozone-oxidized MCC is 1.2  $\mu\text{m}$ , smaller



than native MCC (20  $\mu\text{m}$ ), indicating significant modification on its fibrillar structures. The periodate-oxidized MCC exhibited a smaller particle size of 1.1  $\mu\text{m}$ , which may reflect greater variability due to uneven oxidation and subsequent fragmentation of the cellulose particles. The most substantial reduction in particle size was observed in the combined TEMPO/Periodate-oxidized MCC (540 nm). This significant decrease in particle size suggests that the dual oxidation process induces extensive fragmentation of the cellulose particles. The size reduction can be attributed to the synergistic effect of TEMPO and periodate oxidation, which enhances the cleavage of glycosidic bonds and oxidation of primary hydroxyls, promoting the breakdown of the cellulose structures into smaller particles.

The carboxylate contents of oxidized MCC samples were examined by potentiometric titration, whose profiles are shown in Figure 8. The pH titration curves of 3 different samples illustrate a slight shift in the equilibrium points, indicating differences in their carboxyl contents. The first derivative of the titration curves was used to identify the peaks, i.e., the equilibrium points. For MCC (Figure 8(b)), the equilibrium point was observed at pH 7.16, with 16.3 mL of NaOH titrant. Compared to TEMPO/Ozone and periodate-TEMPO/Ozone oxidized MCC samples (Figures 8(c) and 8(d)), the equilibrium points shifted slightly to pH 7.43 with 16.9 mL of titrant and pH 7.3 with 17.6 mL of titrant, respectively. This confirms that TEMPO/Ozone oxidation increased the concentration of acidic groups, particularly carboxyls, in MCC. The combined TEMPO/Ozone and periodate oxidation introduces further changes, reflected by the peak position and shape alterations, indicating the formation of more complex oxidized structures. This is likely because the sample contains carboxyls and aldehyde groups.



**Figure 8** (a) Comparison of potentiometric titration graphs of MCC and oxidized MCC samples, and (b) Titration graph and first derivative of MCC, (c) TEMPO/Ozone-oxidized MCC, and (d) Combined TEMPO/Ozone and periodate-oxidized MCC in the supernatant fraction.

The TEMPO/Ozone oxidation method for cellulose modification presents a selective and efficient alternative to traditional oxidation techniques. Unlike ozone alone, which breaks down the cellulose structure [22] but lacks the selectivity necessary for targeted functionalization [23-25], the TEMPO-mediated approach allows controlled oxidation of primary hydroxyls to carboxyl groups. This selectivity is vital for applications requiring specific functional groups, e.g., emulsion or nanomaterial synthesis. The TEMPO-mediated selective oxidation emphasizes its importance in producing cellulose fibers with desired functionality [26]. In contrast to harsher chemical oxidants like sodium hypochlorite, which can generate environmentally harmful by-products and require more extreme reaction conditions, the TEMPO/Ozone method operates under milder conditions. It minimizes reliance on toxic reagents detrimental to aquatic life and contributes to environmental pollution. For instance, sodium hypochlorite generates chlorinated by-products that pose risks to human health and the environment, necessitating careful handling and disposal [27]. Integrating ozone, an easily generated and cost-effective co-oxidant, into the TEMPO system enhances oxidation efficiency while aligning with "green" chemistry principles. Consequently, the TEMPO/Ozone system emerges as a more sustainable and selective option for cellulose oxidation than other methods documented in the literature.

#### 4. Conclusion

The process for oxidizing microcrystalline cellulose (MCC) has been optimized to enhance the properties and functionalization of the material. The A process for generating N-oxoammonium cation (TEMPO<sup>+</sup>) from nitroxyl radical (TEMPO) with ozone as a co-oxidant is successfully developed. The reagents selectively oxidize primary hydroxyls of microcrystalline cellulose (MCC) to carboxylic acid. The oxidation efficiency is compared with a periodate oxidation system and a combined TEMPO/Ozone and periodate oxidation. The cellulose in the supernatant fraction exhibited more significant changes than the precipitate fraction. This discrepancy is likely due to the incomplete oxidation of the cellulose. Nonetheless, the TEMPO/Ozone-mediated oxidation demonstrates considerable potential for optimization. Improving the efficiency of this oxidation process could enhance the extent of modification across all cellulose fractions, making it a promising approach for cellulose functionalization. This result can be used as preliminary research for the oxidation method using TEMPO, which is more environmentally friendly for advanced application in the future.

This study investigates the efficiency and mechanisms of microcrystalline cellulose (MCC) oxidation processes, employing TEMPO with ozone as a co-oxidant, in comparison with periodate, and a combined TEMPO/Ozone and periodate mediated oxidation. In the TEMPO/Ozone system, ozone reacts with TEMPO via disproportionation, forming TEMPO<sup>+</sup>, which selectively converts primary hydroxyls to carboxyl groups [8]. Periodate oxidation converts vicinal hydroxyl groups of MCC to aldehyde groups. When the two processes are combined, both reactions occur, and the aldehyde formation enhances the MCC oxidation with TEMPO/Ozone mediation. Insights into the efficiency and mechanisms of these processes are beneficial in optimizing the modifications of MCC for specific applications, especially environmental, packaging, cosmetics, and biomedical fields.

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