

Review Article

Seaweed cellulose nanocrystals: A promising bioresource for sustainable food application

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Cellulose Nanocrystals (CNCs), derived from the natural polymer cellulose, have emerged as a promising bio-based nanomaterial with unique properties, including high crystallinity, non-toxicity, biocompatibility, and a large specific surface area. CNCs can be effectively isolated using acid hydrolysis, TEMPO-mediated isolation, enzymatic isolation, or microwave-assisted isolation followed by mechanical treatment. This review highlights the potential of CNCs in food emulsion stabilisation, functional foods, food packaging, food thickeners and sensors. It also discusses their effectiveness as emulsifiers in pickering emulsions, their role as functional ingredients that enhance food quality, and their use in developing biodegradable packaging materials. Additionally, CNCs are recognised for their thickening properties and ability to improve the stability and quality of starch-based foods. CNCs function as active components or value-added ingredients, contributing to waste reduction through sustainable applications. The study concludes that CNC are prominent natural biomaterial that can be tailored for a variety of food applications.

[**Keywords:** Cellulose, Cellulose nanocrystals, CNCs, Food packaging, Functional foods, Retrogradation]

Introduction

Recently, there has been a shift toward green, sustainable, and eco-friendly materials derived from naturally occurring, biodegradable sources, due to growing environmental concerns. In particular, cellulose is considered a natural polysaccharide, an abundant biopolymer, and an influential bio-based material that serves as a fundamental building block in the structural hierarchy. It acts as the structural cell walls of plants and algae. Cellulose is composed of $\beta(1-4)$ linked β -glucopyranose unit. The derivatives of cellulose have attained widespread popularity for producing innovative products from various sources. These derivatives include Cellulose Nanofibers (CNF), Microcrystalline Cellulose (MCC), Cellulose Nanocrystals (CNC), and bacterial cellulose².

Among cellulose derivatives, CNCs are considered promising nanomaterials for various food applications due to their high crystallinity, biocompatibility, non-toxicity, and large specific surface area³. CNCs appear to be needle-to-rod-like structures⁴ (Fig. 1) with a crystallinity index of up to 88 %^(ref. 5). These nanocrystals are economically viable because they replace some petrochemical-based products while delivering high performance. It is considered the best

biomaterial developed from traditional plant resources such as wood⁶, bast fibre⁷, hemp fibers⁸, sugar cane⁹, cotton¹⁰, garlic skin¹¹, etc.

Seaweed, a marine macroalgae, has been used for centuries in various food and allied sectors due to its unique characteristics and potential applications. It is considered a promising source of cellulose due to its high abundance, fast growth rate, and minimal environmental impact. They are classified into three groups: Green algae (Chlorophyta), brown algae (Phaeophyta) and red algae (Rhodophyta). The average cellulose percentage on a dry weight basis of green seaweed is 9.6, brown seaweed is 7.8, and red seaweed is 4.75^(ref. 12). CNCs were isolated from *Laminaria japonica*, *Sargassum fluitans*, *Ulva lactuca*, *Palmaria palmata*, and *Porphyra umbilicalis*¹³ and found that the seaweeds can be utilised for food applications, such as packaging. Similarly, red algae waste was isolated to develop a polymer nanocomposite¹⁴. Thus, the valorisation of seaweed can be utilised to develop CNC and apply them in distinguished applications.

This review provides an overview of the progressive development of cellulose nanocrystals in different food applications and their safety effects (Fig. 2).

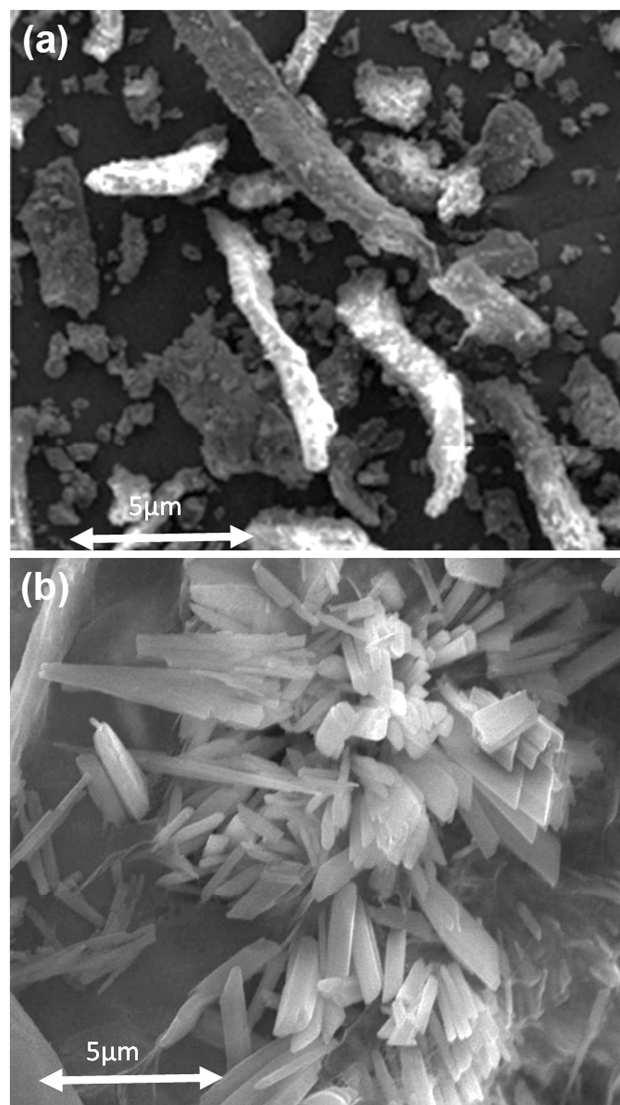


Fig. 1 — SEM images of cellulose nanocrystals sourced from (a) Cotton *Gossypium herbaceum* (Pandi *et al.*, 2021); and (b) Seaweed *Sargassum myriocystum* (work of authors, article in press)

Food emulsion stabilisers

An emulsion is a mixture of two immiscible liquids that requires a stabiliser, typically a surfactant. Pickering emulsion is a type of emulsion in which the water-oil phase boundary is stabilised by solid particles, for example, colloidal silica. They are generally safer than ordinary emulsions and exhibit superior stability and biodegradability¹⁵. Conventional surfactants used as stabilisers contain both hydrophilic and hydrophobic groups. However, nanocelluloses are generally hydrophilic, making them effective stabilisers for oil-in-water emulsions. It has been demonstrated that CNCs obtained from

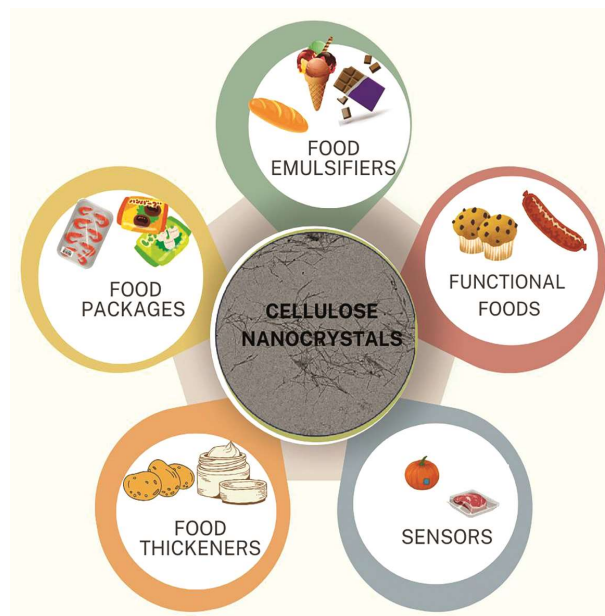


Fig. 2 — Cellulose nanocrystals food applications

various sources, such as algae, cotton, and bacteria, exhibit oil-in-water emulsions by irreversible adsorption^{16,17}. Also, a water-in-water emulsion stabilised by CNC, with adjustable stiffness and gelation rate, allows regulation of the CNC continuous phase rate by varying NaCl^(ref. 18).

Unmodified CNCs, obtained from cotton-linter by acid hydrolysis using H_2SO_4 stabilised canthaxanthin/coconut oil/ water pickering emulsion, and were comparatively more stable than CNC obtained from bacteria¹⁹. Similarly, unmodified CNC was able to stabilise corn oil-in-water pickering emulsion²⁰. Recently, CNCs were modified using food-grade Octenyl Succinic Anhydride (OSA) to enhance their emulsifying capabilities². Pickering emulsion made with OSA-CNC had oil droplets that were extremely resistant to coalescence, even though they were likely to flocculate when the pH or ionic strength was changed to reduce the electrostatic repulsion between them. Even in high-energy microfluidisation, using unmodified CNC, the resulting emulsion was successfully isolated as a Pickering emulsion²², and creaming stability increased as the mean droplet size was less than 1 μm . It was also found that adding seaweed polyphenols maintained the chemical stability of encapsulated β -carotene during spray drying and rehydration²³, establishing a 3D network of antioxidant activity droplets to prevent β -carotene²⁴. Acid-free preparation of nanocellulose using seaweed/kelp (*Laminaria japonica*) by using TEMPO-mediated isolation of CNFs (TEMPO-CNF)

exhibited good stability even after 30 days of storage and emulsified 50 % (v/v) of sunflower oil²⁵. Similarly, nanocellulose isolated from *Undaria pinnatifida*, when fabricated with astaxanthin (AXT), showed good stability in an AXT-loaded pickering emulsion at 50 °C and during storage for up to 14 days²⁶.

Functional foods

Functional food is said to be minimally processed food that is fortified or enhanced with essential ingredients that provide potential health benefits. Nanocellulose acts as a functional ingredient mainly as a food additive²⁷ and fat replacer. Evident use of cellulose as a fat substitute was found in numerous zero-calorie items, such as muffins²⁹, sausage³⁰, and cream³¹. Probiotics are considered as potential functional foods. CNC is said to be the first and most innovative cellulosic nanomaterial with potential efficacy for probiotic encapsulation³². Alginate-CNC-lecithin microbeads serve as an alternative for generating dried microbeads upon application to non-refrigerated, long-shelf-life foods. Biscuit cream stabilised by CNC food additive was made with a substantial amount of oil that met the necessary fatty acid profile³³.

Food packaging application

Most food products are developed using non-renewable and non-biodegradable materials, posing environmental threats. Nanocellulose offers eco-friendly packaging with its superior physical and chemical properties, thus demonstrating great potential for use in food packaging³⁴.

Cellulose nanocrystals have mechanical, thermal, optical and barrier properties, and they are used as reinforcing fillers to aid in the construction of polymer-based nanocomposite films, thus enhancing the characteristics of such polymers to produce the desired materials^{34,35}. They can be combined with poly(3-hydroxybutyrate-co-3-hydroxyvalerate) (PHBV)³⁶, polyvinyl alcohol (PVA)³⁷, polylactic acid (PLA)³⁸, and starch³⁹, which are considered traditional polymers. Incorporation of CNC as fillers into these polymers improves barrier properties such as oxygen barrier, mechanical and biodegradable properties, and increase Young's modulus and tensile strength³⁸.

CNCs from various origins were incorporated into alginate-acerola puree film, improving the film's overall tensile properties except elongation and water vapour barrier³⁹. The modified CNCs by acetic

anhydride, used as a reinforcing agent in linear low-density polyethylene, increased the nanocomposite between CNCs and the matrix⁴⁰. A PVA/chitosan nanocomposite film from acid-hydrolysed CNC from rice straw⁴¹ showed strong hydrogen bonding and electrostatic interactions, resulting in a 130 % increase in tensile strength⁴². The combination of PET/CNC film has increased the water vapour barrier properties, as normal PET film has a water transmission rate of 37 gm⁻² day⁻¹, compared to that of PET/CNC film, which has a water transmission rate of 10 gm⁻² day⁻¹(ref. 43). CNCs isolated from *Enteromorpha prolifera* were used as a promising reinforcement nanofiller to develop a water-soluble polysaccharide bio-nanocomposite film to enhance mechanical properties with a high aspect ratio. Similarly, CNCs obtained from green, red, and brown seaweeds showed higher thermal properties, crystallinity, and aspect ratio than CNCs obtained from traditional resources⁴⁴. Moreover, CNC is used as a food freshness indicator and as intelligent packaging⁴⁵. Herein, a pH-sensing film was developed using hydroxypropyl guar, 1-butyl-3-methylimidazolium chloride, and anthocyanin, with the addition of a CNC-promoted compact film structure that improved the sensitivity of pH, colour, durability, and low-temperature resistance. Whey protein films improved mechanical properties, antioxidant activity, and water permeability when coupled with CNC-coated bergamot oil⁴⁶. Moreover, Konjac glucomannan/carrageenan films (KCNTO) incorporating *Litsea cubeba* essential oil pickering emulsion stabilised by CNC/tannic acid showed enhanced antioxidant, antimicrobial, and barrier properties, which effectively preserved pork quality during cold storage⁴⁷. Highly crystalline, thermally stable CNCs isolated from *Dictyota bartayresiana* showed promising nanofiller for the biodegradable films⁴⁸. Additionally, seaweed-based cling films reinforced with CNCs from *Sargassum wightii* demonstrated superior mechanical, barrier, and degradability properties for sustainable food packaging⁴⁹. Edible films made from *Ulva lactuca* extract with cellulose, CNC, or their combination showed improved barrier and optical properties, especially in the C/CNC blend, demonstrating strong hydrogen bonding, reduced moisture and gas permeability, high light-blocking ability, and notable antioxidant activity influenced by food simulants⁵⁰. Similarly, seaweed-derived nanocellulose isolated via electron-beam-assisted methods produced high-performance biodegradable films with reduced environmental impact⁵¹.

Food thickener and starch foods

The rheological properties of CNs make them effective thickening agents. CNCs entangled with other forms contribute to high viscosity, which helps in the thickening effect⁵². Chitosan/guar gum in combination with 3 % CNC developed a nanocomposite suspension that increased viscosity by 27 %, attributed to the hydroxyl groups⁵³. Nevertheless, CNCs with a high aspect ratio exhibit superior viscosity and thickening capabilities due to their enhanced ability to form polymer matrices⁵³.

Likewise, CNCs have been explored to enhance the properties of starch food matrices. Usually, food quality is often compromised by the gelatinisation of starch. The CNCs source, shape and processing parameters determine the precise impact on starchy food matrices⁵⁴.

The addition of CNC was found to inhibit/reduce starch retrogradation⁵⁴. CNCs from various origins such as mango seed shells, sugarcane bagasse and rice straw can be incorporated into starch-based films to improve barrier properties⁵⁴⁻⁵⁶. Sodium hexametaphosphate cross-linking agents inhibited retrogradation due to the formation of bonds between starch and CNC^(ref. 57). Amylase and CNC interaction revealed that incorporating CNC reduce Rapidly Digestible Starch (RDS) and Slowly Digestible Starch (SDS), while simultaneously increasing the content of Resistant Starch (RS). Particularly, starch in corn, pea and potato increased to 30.4, 23.13, and 20.30 %, respectively⁵⁸.

Sensors

CNCs can serve as a matrix for various types of sensors to detect quality during food processing. CNC-based sensors have been developed to detect glucose levels by creating an electrochemical glucose sensor via pyrrole polymerisation on the surface of CNCs⁵⁹. Similarly, a CNC matrix was fabricated for a pH-sensing film, exhibiting a clear colour change to indicate pH variations⁶⁰. Colourimetric sensors help identify the onset of food spoilage by detecting the gas released. For instance, polydiacetylene-based film sensors demonstrated a colour change from blue to red when exposed to spoiled or rotten meat below freezing temperatures⁶¹. Additionally, colourimetric sensors modified with amine groups on CNC changed colour when exposed to aldehyde gases and discriminated them from non-aldehyde gases⁶¹. Surface-Enhanced Raman Scattering (SERS) smart

sensors developed with cellulose can detect contaminants at trace levels, exhibiting good performance for targeting complex molecules in food systems⁶².

Toxicological characterisation and safety aspects

According to the European Food Safety Authority (EFSA), *in vivo* and *in vitro* studies found that none indicated toxicity following oral or simulated oral exposure. However, CNC developed is similarly safe to that of conventional cellulose, *i.e.*, 2058.3 (males) and 2682.8 (females) mg/kg/day⁶³. Nanocellulose was passed through the mouth, stomach, and small intestines in fasting and non-fasting conditions, and cytotoxicity was studied, demonstrating only minimal changes in factors, including oxidative stress^{64,55}. However, excessive inhalation of CNC can cause pulmonary inflammation due to nanodegradation and the aggregation of nanocellulose⁶⁵.

The long-fibre structure of CNF can sometimes cause more toxicity than CNC, leading to cytotoxicity and oxidative stress⁶⁶, potentially showing carcinogenic effects⁶⁶. Some studies also found that the persistence of CNF can aggravate the immune system when impaired and present at higher concentrations⁶⁷. An experiment conducted with carboxymethylated CNF with a dosage of once/day for 8 weeks showed no toxicity and no concentration dependence⁶⁸.

In contrast, conventional forms of cellulose, like methylcellulose (MC), are considered “Generally Regarded As Safe” (GRAS) and can be incorporated into food ingredients according to the US Food and Drug Administration (FDA). The studies so far have not found any toxicological effects of CNCs and can therefore be used as a food additive at low concentrations.

Conclusion

In conclusion, CNCs offer promising potential for developing sustainable and eco-friendly materials for food applications. The potential of CNCs as a reinforcing filler in food packaging is evident from their mechanical, thermal and barrier properties. Since a reinforcing filler for polymer-based nanocomposite film has demonstrated promising mechanical, thermal, and barrier properties, providing a sustainable substitute for traditional packaging, the promise of CNC in food packaging is clear. In addition, CNC's versatility and potential health benefits are

highlighted by its use as a functional component and fat substitute in functional foods like probiotics. It has also improved the overall quality and shelf life of starch-based food matrices by preventing retrogradation, aligning with the growing desire for healthier and more functional options. Besides, it serves as a sensor to detect pH changes and food spoilage, guaranteeing food safety and quality control. Furthermore, it enhances upcoming nutraceutical applications by serving as a carrier agent to encapsulate the active components. Overall, the future of sustainable materials hinges on cost-effective, eco-friendly solutions from food packaging to biomedical advancements, CNCs hold the potential to revolutionise industries while minimising environmental impact. To fully harness their benefits, rigorous toxicological assessments must accompany development to ensure their safe and widespread adoption.

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Conflict of Interest

All the authors declare that there is no conflict of interest.

Author Contributions

NPN: Conceptualization, data analysis, investigation, writing original manuscript; and SRRR: Conceptualisation, supervision, review, and editing.

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