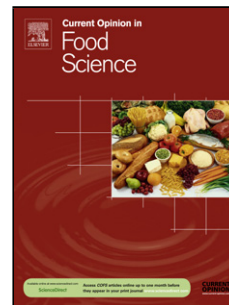


# Journal Pre-proof

Use of nanocellulose in meat products

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## Use of nanocellulose in meat products

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### Highlights

- Raw meat batters with nanocellulose showed a predominantly gel-like elastic behavior
- Nanocellulose emulsifying and water-binding properties increase product stability
- Nanocellulose addition upgrades the hardness of different meat systems
- A more dense network, with few and small cavities, was formed
- Nanocellulose addition did not affect flavor and improved the sensorial texture

### Abstract

In healthier reformulated meat products throughout animal fat reduction or its replacement with emulsified oil, hydrocolloids have been extensively studied and used as stabilizers. Among them, vegetable and bacterial nanocellulose are available to be employed in meat products. Each one has similar but particular characteristics, as high water- and oil- binding capacities, crystallinity, and rheological properties, and could act as an emulsifying and stabilizing ingredient. Therefore, they must be studied and selected according to the necessities of the different reformulated matrixes to improve their characteristics. Several studies have evaluated the nanocellulose addition in meat products with different fat levels or pre-emulsified oil as emulsion stabilizer or fat-replacer. This review aims to discuss the main applications of nanocellulose types in healthier meat products.

**Keywords:** Nanocellulose; meat products; applications; emulsion stability, texture, rheology.

## **Introduction**

Meat and meat products are considered fundamental components of the human diet since they are good sources of high-quality proteins, essential amino acids, B-group vitamins, and minerals [1]. But, like any other foods, they also have constituents that, when consumed in inappropriate amounts may enhance the risk of some of the major degenerative and chronic diseases (ischemic heart disease, cancer, etc.). The growing understanding of the relationship between diet and health combined with consumer interest in healthier nutritious foods with additional health-promoting functions is driving the development of functional foods, a challenge for the meat industry future [2]. In order to respond, extensive research has been done in different options of "fat

replacers/analogs" [3], or the totally or partially fat replacement by pre-emulsified oil, from vegetal or marine sources, where it is mandatory to employ hydrocolloids such as rice bran fiber, carrageenan, pectin, or cellulose to obtain stable meat emulsions [4]. Cellulose fibrils with widths in the nanometer range are nature-based materials with unique and potentially useful features due to their high surface area and hence the powerful interaction of these celluloses with surrounding species, such as water, organic and polymeric compounds [5]. According to Gómez et al. [6], the potential use of vegetable nanocellulose, extracted from wood, cotton, natural fibers, and lignocellulosic materials, as a food additive, natural emulsifying and stabilizing ingredient, was first described in the 1980s. It is based on the ability of the dispersed nanocellulose to adsorb at the oil-water interface and to form steric barriers around the emulsion droplets, which prevent coalescence due to its better affinity for water than for oil [7,8]. Therefore, nanocellulose has high potential as a stabilizing agent for emulsions and food such as crushes, soups, gravies, puddings, dips, toppings, frozen desserts, and meats. Also, nanocellulose is a dietary fiber that plays a beneficial role in the overall health of adults; consequently, it could be considered a functional food ingredient [6].

Vegetable nanocellulose (VNC) was first produced in 1977 by chemical treatment of wood pulp with HCl. The acid hydrolyzed the hemicelluloses and pectin by breaking down the polysaccharides to simple sugars and hence released the cellulose fibers.

Afterward, NaOH was employed to separate alkali-soluble structures. It was followed by a different mechanical treatment and a high-pressure homogenizer to deconstruct the hierarchical structure of fibers obtaining considerable quantities of nanocellulose [6].

The first scientific article on this topic was presented at the Ninth Cellulose Conference in 1981 and later published in the conference proceedings in 1983 [9].

Bacterial nanocellulose (BNC) was first described in 1886 by A. J. Brown [10]. It was produced by *Acetobacter xylinum* (nowadays known as *Komagataeibacter xylinus*, or *Gluconacetobacter xylinus*) as a polymer and nanomaterial by biotechnological assembly processes from low-molecular-weight carbon sources, such as D-glucose, in the presence of oxygen using a bottom-up method [11]. To large-scale production, agro-industrial by-products like cane molasses from manufacturing and refining of sugarcane, glycerol from biodiesel, vegetable oils, methanol, and hydrocarbons, have been proposed as non-conventional cheaper carbon sources [12].

Figure 1 depicts a simple schematization of both VNC (Fig. 1a) and BNC (Fig. 1b) productions. These processes may have modifications and downstream post-treatment of the hydrocolloid, according to emerging technologies, available raw material, and/or techno-functional needs of nanocellulose.

- Insert Figure 1 here -

BNC chemical composition is identical to plant-derived cellulose. However, it can be obtained with higher purity since it does not contain hemicellulose, lignin, and pectin; therefore, their removal with harsh chemicals is not required with a reduction in the purifying costs and the environmental damages [13]. When compared to VNC, BNC presents a higher degree of polymerization (4000-10000 units), crystallinity (80-90%), water holding capacity and longer drying time, and mechanical and thermal stability [5,14,15,16]. In addition to the techno-functional characteristics above described of vegetal and bacterial nanocelluloses, a cost analysis should be also consider comparing the options in order to be selected as an ingredient in a meat product.

Therefore, different nanocellulose types would provide slightly diverse properties to the matrixes, so they could be selected according to the targeted meat product through a deep study in each case.

All forms of nanocellulose are usually chemically treated after production and resulted in slurries, pellicles, or droplets. At this point, it can be sterilized and employed as a food additive but owing to its low dry-weight, the proper amount of nanocellulose results difficult to add without adding excessive water [17]. Drying processes can be employed but could affect their properties and must be studied. Recently, Balquinta et al. [18] studied different post-synthetic treatments (ground, acid hydrolysis, and ultrasound) on bacterial nanocellulose sheets observing modifications in its crystallinity and water-binding properties that would allow the post-synthetic process selection to satisfy the matrix requirements in which the nanocellulose fibers will be incorporated. Therefore, this review aims to examine the main studies related to the application of different types of nanocellulose as a fat replacer and/or binding agent to produce healthier meat products, which are summarized in Table 1.

- Insert Table 1 here -

### **Effect of nanocellulose inclusion on raw meat batters**

The first step to obtain an emulsified meat product such as sausages, bolognas, luncheon meats, pâtés, among others, consists of the batter preparation. Briefly, emulsion-type meat products are made by chopping the meat in a cutter after the addition of salt, phosphates, and water, and converting the chopped meat mass along with all extracted proteins into a viscous mass [23]. This process is responsible for proper fat

emulsification and water binding in the meat emulsion matrix. In the last step during chopping, fat is added to form an oil-in-water emulsion [3].

Different studies had evaluated the nanocellulose addition in the preparation of emulsified meat products and its batter rheological behavior or consistencies. Marchetti et al. [11] analyzed the effect of bacterial nanocellulose (BNC) addition (levels up to 0.53 g/100 g) in low-fat low-sodium meat emulsions with pre-emulsified high oleic acid sunflower oil on batter rheological characteristics. The elastic modulus ( $G'$ ) at 1 Hz at 25 °C, which reflects the behavior of the batters, was increased when BNC was included in the systems. BNC level of 0.27 g/100 g resulted in similar  $G'$  values to a regular-fat/regular-sodium sausage formulation, being for 0.53 g BNC/100 g 70% higher than the low-fat low-sodium formulation without nanocellulose and slightly higher than the regular-fat one. Besides, Zhao et al. [4] informed similar trends with 0.4 to 1.2 g/100 g of the hydrocolloid in emulsified model meat systems finding that their viscoelastic properties  $G'$ ,  $G''$ , and viscosity increased with the concentration. On the other hand, Qi et al. [20] informed that 2 g/100 g of nanocellulose of different sources in meat batters slightly decreased their elastic characteristic at 25°C. When a high nanocellulose level at constant water is employed in a matrix, it would show a negative effect on texture properties due to a decrease in water availability, and eventually, meat protein-water interactions will be affected, leading to a weak meat-protein gel [11]. Changes in elastic modulus ( $G'$ ), loss modulus ( $G''$ ), and loss tangent ( $\tan \delta = G''/G'$ ) of the batters could be studied during the heating (cooking) and cooling process as a function of time. In batter formulations with vegetable or bacterial nanocellulose  $G'$  values were always over  $G''$ , and loss tangents were less than 0.25 [11,20], showing a predominantly elastic behavior of gel-like structures during the whole temperature ramp. When batters were heated from 25 to 75 °C (simulating a cooking process), the

hydrophobic interactions were established reinforcing the gel-like characteristics of the systems as the higher elastic moduli at 75 °C of meat batter with bacterial [11] or vegetal nanocellulose (VNC) [19,20] were reached. This could be attributed to the crosslinking of solubilized proteins in the meat batter upon heating. Some researchers reported that the increase in  $G'$  was related to the denaturation and aggregation of myosin structures when the batters were subjected to high temperatures. Panagopoulou et al. [24] reported that nanocellulose could interact with proteins by hydrogen bonds and van der Waals' force, thus, attaching to the protein network structure, and, as a result, a more compact 3D-network structure was formed. These findings confirmed that nanocellulose had a significant impact on the emulsified meat-gel structure.

### **Effect of nanocellulose addition on cooked meat products**

#### ***Emulsion stability and water-binding properties***

The batter rheological characteristics could be related to cooked meat-emulsion stability. The stability and functionality of a protein gel in meat systems are critical to the quality of the meat products, which is largely determined by the ability to bind and retain water [25]. When emulsified-meat batters are cooked, denaturation of meat proteins occurs, and a tridimensional gel-like network is established. This process can lead to water and/or oil release and is at this point when hydrocolloids such as nanocellulose should act, reducing water/oil losses. Generally, the strong entangled and disordered networks of nanocellulose fibers can crosslink with protein and form a 3D-network that may contribute to preventing the loss of fat and water and decreasing the cooking loss. It was observed through cryo-SEM micrographs of O/W systems that regenerated cellulose nanofibers were not only presented in the continuous phase but also adsorbed on the surface of oil droplets [4]. Furthermore, their hydrophilic nature



gives them adequate water-retaining properties like other hydrocolloids [19]. The cooking loss, or in the other way of measuring, the process yield, can reflect the water-holding ability or oil-binding capacity of the meat products during the cooking process, and therefore the emulsified stability of meat batters [20].

Parés et al. [7] found that 0.5 g/100 g of nanofibrillated cellulose was successfully employed as a fat binder in the stabilization of meat emulsions without added phosphates and starch (finding similar composition and WHC than the control) but was not able to replace the sodium caseinate. Qi et al. [20] pointed out that the addition of 2 g/100 g of vegetable nanocellulose of different sources to pre-emulsified soybean oil meat sausages provided adequate emulsion stability. The authors attributed these results to their emulsifying properties that also improved the water-binding capacities throughout their physical entanglement, aggregation, and supramolecular interactions. Due to the excellent emulsifying properties of the hydrocolloid, the pre-emulsifying process improved the emulsion stability of the sausages. Besides, the higher water-binding capacities were achieved when nanocellulose was included. Meanwhile, Wang et al. [19] informed that nanofibrillated cellulose addition to fat-replaced sausages produced lower cooking losses than the full-fat control. Furthermore, bacterial nanocellulose (0.12-0.53 g/100 g) added in low-fat low-sodium sausages resulted in excellent process yields with a significant increase in water content and a decrease in water activities concerning the control, improving the water-binding properties of the systems [11]. Besides, water holding capacity (WHC), which reflects the strength of water-matrix interactions, was enhanced when BNC was included, reaching the highest with 0.267 g BNC/100 g. This increased WHC is important to achieve in a formulation with a fat mimetic since solid fat replacement with oil strongly diminishes the binding capacity of a cooked meat emulsion [26]. Besides, Zhao et al. [4] demonstrated that

reconstituted cellulose fiber (0.8 g/100 g) and myofibrillar proteins had a synergistic effect on the stability of the emulsion model system. The authors explained it by the fiber's good thickening and gelling properties that led to a satisfying 3D-network in the continuous aqueous phase, a higher viscosity, and a steric barrier that immobilizes the oil droplets and inhibits creaming.

Other studies that employed nanocellulose slurry as a binder (level 0.24-0.63g/100 g) in hamburgers reported that cook losses were reduced with a higher content of the hydrocolloid [17]. Also, in comparison with potato starch, the nanocellulose was more efficient to retain liquid and gave a better sensorial response.

#### ***Texture and rheological behavior***

The most important parameters to define and differentiate the texture of emulsified meat products like sausages are hardness, juiciness, and those relating to them. A medium hardness and high juiciness sausage were considered by the consumers to have the best texture [27]. When traditional emulsified meat products are reformulated to obtain low-fat alternatives, texture profile must be taken into account to avoid changes that could affect consumers' acceptance. Because of its high gelling properties, bacterial nanocellulose (0.27 g/100 g) results in an excellent alternative to attend this issue in low-fat low-sodium meat sausages through reinforcement in the protein matrix [11]. Additionally, Parés et al. [7] were able to compensate for the elasticity imparted by starch in the conventional formulation by its replacement with 0.5 g of nanofibrillated cellulose/100 g, while Qi et al. [20] informed that different sources of vegetal nanocellulose (2 g/100 g) could promote hardness, springiness, and chewiness of sausages but no significant differences were observed in the cohesiveness nor adhesiveness. Wang et al. [19] studied pork sausages with 30 or 50 % fat replaced by

nanofibrillated cellulose and its palm oil Pickering emulsion (0.12-0.15 g/100 g) founding an increase in hardness and springiness while cohesiveness was lower concerning the full-fat control.

Frequency sweeps of storage ( $G'$ ) and loss ( $G''$ ) moduli after the cooling stage at 25 °C of the products could give information about their structure. Marchetti et al. [11], Wang et al. [19], and Qi et al. [20] found that in all nanocellulose-added emulsified meat products  $G'$  was higher than  $G''$ , no crossover with increasing angular frequency was observed, and a lower slope of  $G'$  and  $G''$  was noticed. Moduli values were increased with the concentration level of reconstituted nanocellulose (in the range 0.4 -1.2 g/100 g) or up to 0.267 g/100 g of bacterial nanocellulose.

The addition of nanocellulose led to a structural reinforcement due to the formation of a dynamic network in unheated meat batters that was enhanced upon heating. These indicated elastic gel-like behaviors of the meat systems.

### ***Microstructure***

Changes in rheological behavior, water-binding properties, and texture profile of nanocellulose-added meat emulsions could be correlated with their microstructure. Several studies [7,11,19,20] concluded through the microscopic observations of the different nanocellulose-added meat sausages that the matrixes were compact or dense homogeneous networks, with few and/or small cavities, and could be related to their textural and rheological behavior. These rigid networks correlated with their high hardness or viscoelastic properties and with a crosslink between the nanocellulose and the protein matrix, which helps to entrap more fat droplets and form a more homogeneous structure that leads to better emulsion stability with higher yields and WHC.

Model meat emulsion systems with cellulose nanofibers (0.4-1.2 g/100 g) were observed with confocal laser scattering microscopy (CLSM) and cryo-scanning electron microscopy (cryo-SEM) [4]. The authors informed that a level of 0.8 g/100 g resulted in a more compact structure with smaller oil droplets. Also, the interfacial protein on the membrane of oil droplets increased with increasing nanofiber concentration probably due to their contribution to enhancing the interactions between meat proteins and fat. Finally, they proposed that the main mechanism for the emulsion stabilization by nanofiber is through network formation and Pickering emulsion.

### ***Consumer's acceptance***

Consumers associate cooked meat products with a bright and characteristic pink color [28]. So from the consumer's point of view, a brighter and pinker product is preferred. Considering that most nanocellulose forms are tasteless, odorless, and colorless, the main effect on sensorial attributes would be related to texture, palatability, and/or juiciness of the meat product.

Nanofibrillated cellulose was effectively employed by Wang et al. [19] to include pre-emulsified canola oil in fat-replaced products, which were brighter than a full-fat control. This effect could be attributed to higher water content and the emulsification process included leading to brighter and a smoother surface [26].

Regarding the texture, the incorporation of pre-emulsified palm oil produced high texture scores as much harder products were obtained compared with control due to the structure was enhanced by the nanofibrillated cellulose addition. Besides, no differences were found in the overall acceptability. Microfibrillated cellulose employed in hamburgers gave no off-flavors, the same texture, and mouthfeel as hamburger without additions, and held more water without side effects as watery taste [17]. Also, bacterial

nanocellulose, in the form of nata, had some detrimental effects on Chinese-style meatball formulation as a protein/meat replacer [21]. With over 10% of nata, lowered springiness scores were reported by trained panelists. A similar tendency was observed for firmness, although juiciness increased with increasing nata levels. Nevertheless, no statistical difference was found among 0% (control), 10%, and 20% of nata in the overall product acceptability. Considering springiness and firmness, 2 of the foremost Chinese-style meatball eating-quality parameters, panelists found that the addition of 30% of nata led to unacceptable products.

Besides, Okiyama et al. [22] decreased the energy content of hamburger patties by replacing one-third of beef with BNC paste without impairing tenderness and juiciness. A similar effect was also observed in sausage, where lard was partially replaced with BNC paste and judged to be as good as the control by expert panels. The authors believed that because these minced meat products were heterogeneous and rather fragile, the addition of BNC did not affect the texture greatly.

## **Conclusions**

Several studies had shown that nanocellulose of both origins, vegetal or bacterial, could be useful to act as a stabilizing agent in meat products with different fat levels or pre-emulsified oil. Most nanocellulose additions have improved its water- and oil- binding properties, rheological behavior, and texture, due to the development of matrixes with cohesive and homogeneous microstructures. Nevertheless, more research is necessary to exhaustively evaluate all the nanocellulose possibilities, from different origins and in appropriated levels, to improve a particular meat product throughout specific studies that cover nanocellulose effect on its main properties. Besides, further experiments and advances are needed to find the more adequate industrial options of the diverse

nanocellulose and their state that would adapt better to the industrial production processes and needs of the specific meat product.

### **CRedit authorship contribution statement**

Lucas Marchetti: Investigation, Writing -original draft, Writing - review & editing.

Silvina Cecilia Andrés: Conceptualization, Investigation, Writing - review & editing, Supervision, Project administration, Funding acquisition

### **Declaration of interests**

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.

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\*This work evaluated the bacterial nanocellulose (BNC) application at different levels to low-lipid low-sodium meat emulsions with pre-emulsified high-oleic sunflower oil. Results showed that water-binding properties, hardness, cohesiveness, and chewiness increased when up to 0.267 g of dry BNC/100 g batter was added, while further



additions were harmed. The addition of BNC was not detrimental to shelf-life since quality parameters remained stable under cold vacuum storage for 45-days.

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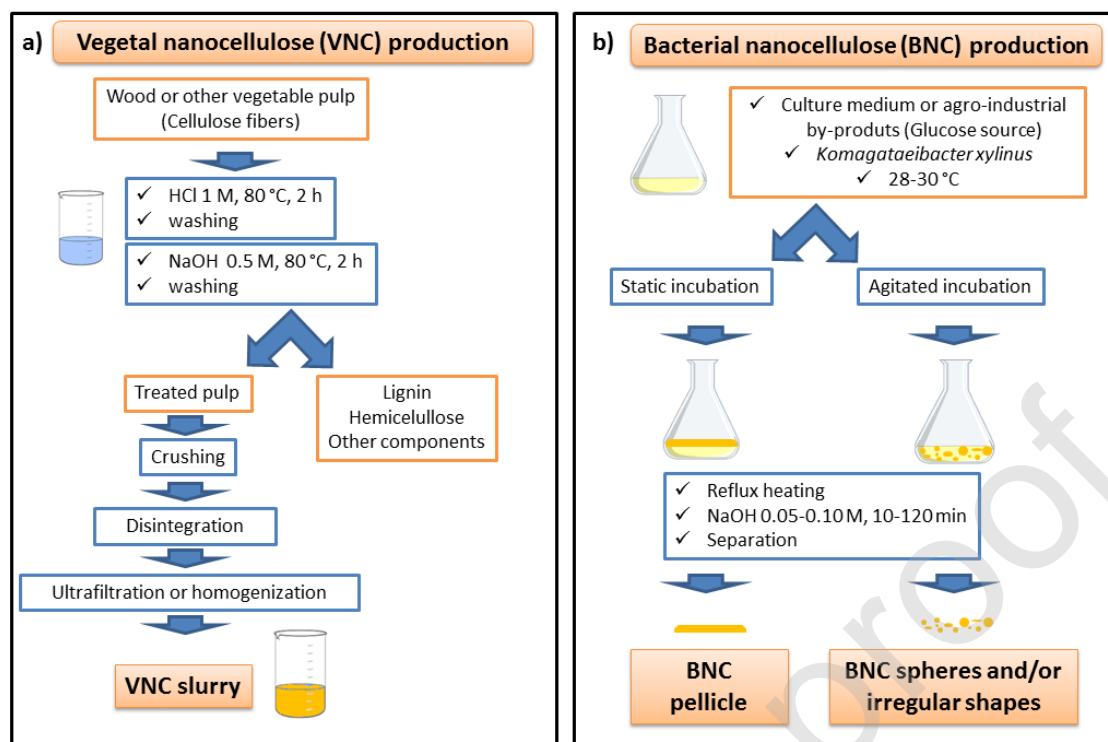
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## Figure Captions



**Figure 1.** Process scheme for the production of a) Vegetal nanocellulose (VNC) and b) Bacterial nanocellulose (BNC).

**Table 1.** Recent studies on nanocellulose addition in **meat products**.

Meat product	Lipid added (g/100 g batter)	Nanocellulose		Reference
		Type	Level (g/100 g batter)	
Low-fat low-sodium meat sausages	5	Bacterial nanocellulose	0.12-0.53	[11]
Pork meat sausages	30	Nanofibrillated cellulose	0.09-0.18	[19]
Meat sausages	24	Nanofibrillated cellulose	0.5	[7]
Emulsion model systems	20	Regenerated cellulose fiber	0.4-1.2	[4]
Meat sausages	10	Vegetal nanocellulose	2	[20]
Chinese-style meatball	20	Nata (Bacterial nanocellulose)	10-30	[21]
Hamburger	-	Microfibrillated cellulose	0.24-0.63	[17]
Hamburger patty/ sausages	10-12	Bacterial nanocellulose	0.70-0.20	[22]