Full Paper

Optical Properties of Self-Assembled Cellulose Nanocrystals Films Suspended at Planar-Symmetrical Interfaces.

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Keywords: nanocellulose, nanorods, chiral, confinement, gelation

Hierarchically structured materials comprising rod-like, chiral, nanoparticles are commonly encountered in nature as they can form assemblies with exceptional optical and mechanical characteristics. These include cellulose nanocrystals (CNCs), which have a large potential for the fabrication of bio-inspired materials mimicking those advanced properties. Fine-tuning the opto-mechanical properties of assemblies obtained from CNCs hinges on the transformations from suspensions of liquid crystals to long-range order in the dry state. So far, associated transitions have been studied using trivial interfaces such as planar substrates. Herein, we explore such transitions as

This is the author manuscript accepted for publication and has undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the [Version of Record](#). Please cite this article as [doi: 10.1002/sml.201702084](#).

they evolve onto meshed supports. The meshed substrate offers a complex topology, as is encountered in nature, for the formation of CNCs films. The CNCs self-assembly occurs under confinement and support of the framework bounding the mesh openings. This leads to coexisting suspended and supported nanoparticle layers exhibiting nematic and/or chiral nematic order. Optical microscopy combined with crossed polarizers indicate that the formation of the suspended films occurs via intermediate gelation or kinetic arrest of CNCs across the mesh's open areas. The formation of self-standing, ultra-thin films of CNCs with tunable optical properties, such as selective reflections in visible range (structural color), is demonstrated by using the presented simple and scalable approach.

1. Introduction

In the endeavor of forming materials mimicking multilayered, super-structured materials encountered in nature, cellulose nanocrystals (CNCs) are a promising building block.^[1-3] CNCs are a principal component of cells in plants and several organisms. They are rod-like crystals of cellulose with characteristic dimensions that range between ~3 and 70 nm in width and ~35 to 3000 nm in length, depending on the source.^[4] CNCs possess a right-handed chiral twist, enabling them to form liquid crystals above a given concentration, depending on their aspect ratio, crystallinity and surface charges, which can be tethered through their isolation process.^[5-7] Beyond their appeal for the formation of environmentally friendly materials, CNCs bear a unique set of properties. For example, they display a very high dielectric moment resulting in a high piezo-electrical response.^[8] They are also biodegradable and are low-to-nontoxic.^[9] Additionally, they display outstanding thermo-mechanical properties^[10-12] and their surface can be easily modified synthetically.^[13,14] Importantly,

CNCs are birefringent and self-assemble into nematic or chiral nematic orders that enable versatile optical properties.^[5, 15-17]

The combination of their biomass origin and inherent properties, make CNCs ideal candidates for the formation of sustainable, advanced materials. For this purpose, the mechanical toughness of CNCs films is expected to be coupled with their transmission and reflection properties. Associated properties of CNCs films are highly dependent on the transfer of the long-range order of the liquid crystalline suspension to the solid, dry films. In order to achieve controlled opto-mechanical properties, two orders of CNCs are sought: (i) nematic order whereby the CNCs are aligned along a given axis and parallel to each other within stacked planes^[18-21] or, (ii) chiral nematic order, whereby CNCs are aligned within a plane along a given axis, which rotates with respect to neighboring planes, forming a 3D structure with CNCs helically arranged across planes.^[1, 3, 19, 22, 23] For optical properties of materials formed from CNC, a nematic order translates into layers or structures with a reduced opacity. On the other hand, chiral nematic order allows control over the reflectance and transmittance of the material, imbuing it with tunable colors.^[5, 15-17, 24] The nematic order is observed when a film forms from a rapidly drying CNC suspension or under shear; in contrast, chiral nematic order requires a longer equilibration time. Control over the chiral nematic order can be achieved by adjusting the interfacial forces between CNCs^[25-29] or by controlling variables during drying such as rate,^[30] shear,^[19] temperature,^[31] or the surface chemistry of the substrate.^[32]

Typically, studies on the formation of single-component, planar CNC materials with long-range order formed by evaporation-induced self-assembly (EISA) have focused on films obtained after drying the suspension on flat solid surfaces. In order to extend the applicability of CNCs films, an understanding of the transition from liquid crystal suspensions to dry films on complex interfaces and on surfaces

with non-planar topologies is required. Herein we address EISA of CNCs onto non-trivial systems such as those comprising alternating curved and open, discontinuous interfaces. Specifically, we use geometries analogous to the homogeneous air-water-air interfaces observed in foams, which promote close packing of spherical and rod-like nanoparticles.^[33, 34] Although CNCs do not display strong interactions at the air-water interface, we show that kinetic arrest of CNCs (i.e. gelation at ca. 10 % CNC solids content) enables the formation of self-supported, suspended, thin films (see **Figure 1** for CNCs films formed within micro-scaled quadrilateral openings of meshes). Here, the suspended films of CNCs were formed across supporting flexible meshes with quadrilateral openings with four different lateral sizes, ranging from 48 to 522 μm . The filaments forming the meshes were composed of polyamide (Nylon 66) making the surface hydrophilic and susceptible to H-bonding with CNCs, leading to strong adhesive interactions. We specifically focused on the characterization of the factors affecting the formation of anisotropically ordered films that bear distinctive optical and mechanical properties. We first studied the film formation as a function of opening size, CNC concentration, cast suspension volume and the presence of surfactants or electrolytes. Following this, we quantified their optical properties as a function of the suspension composition by transmittance measurements and polarized microscopy. We also studied the orientation of CNCs within the suspended thin films formed across the cast area as a function of the suspension composition. The long-range order of CNCs within the films was further studied by imaging fractured films via scanning electron microscopy (SEM). Finally, we show how the employed flexible meshes allowed the formation of structured suspended films across larger areas and how nematic or chiral nematic orders were obtained. Our principal findings indicate that (1) gelation was paramount to the formation of suspended CNCs films across open areas, (2) additives prevented chiral nematic order

of the CNCs films and, (3) the deposition method significantly affected whether the films were in a nematic or chiral nematic order in the dry state. We expect this work to provide a fundamental basis for the formation of thin CNCs films onto complex interfaces with tailorable optical properties while maintaining the mesogen amount to a minimum. This is ideal in a number of application such as liquid crystals based sensing,^[35,36] porous membranes,^[4,37] iridescent materials^[37] and impact-resistant materials.^[38] Furthermore, the proposed setup was particularly interesting under high shear, e.g., under fast drying, because the effect of the rod-like nature of the nanoparticles dominated over the long-range order obtained in the dry state. In contrast, the chiral nature of CNCs was predominant in the absence of shear, e.g., for films obtained after slow drying. Therefore, this investigation bears meaning for disciplines where rod-like particles or liquid crystals are used to synthesize advanced thin films. Furthermore, the fact that the films were suspended within the mesh allowed the formation of thin substrate-free layers that otherwise could not be isolated or separated from the commonly used solid supports.

Figure 1 Process of formation of CNC thin films on a flexible mesh. A CNC suspension is first cast and suspended across the hydrophilic nylon mesh (top, left frame, with the inset showing an image of a drop cast across a micro-scaled mesh). Drying at constant contact points with varying contact angle then occurs as a function of time t (top, right frame). Upon drying, particle concentration increases, which leads to CNC self-assembly into larger structures, i.e., tactoids that form the suspended film (bottom right frame). Upon complete drying, film consolidation occurs across the openings where the CNC suspension was initially cast (bottom, left frame that includes an SEM image of a CNCs film obtained in this manner, inset) (scale bar is 20 μm).

2. Results and Discussion

2.1. Formation of CNCs films across mesh boundaries

CNCs essentially have no affinity for the air-water interface.^[39, 40] Nevertheless, planar films can be formed across the mesh openings after solvent evaporation (**Figure 1 and 2a**). In this case, the formation of the CNC standing films resulted principally from the gelation of the aqueous suspension upon drying and after reaching a critical concentration (at ca. 10 % volume fraction).^[5] Although the zeta potential of the CNCs used in this study was -34 ± 10 mV, above the threshold necessary for colloidal stability ($\sim \pm 21$ mV^[41]), the high aspect ratio of CNCs led to entanglement and gelation at high concentrations.^[5,42] The supporting mesh consisted of nylon, providing a relatively hydrophilic surface that enabled H-bonding interactions with CNCs, thus constraining film formation within the cast areas. Such areas were defined by the volume of deposited CNC suspension, which exhibited a decreasing, receding contact angle during drying. As a result, the dried CNC suspension formed a rim, comprising films suspended across the mesh openings as well as CNC layers adsorbed atop the surface of the mesh's filaments (**Figure 2**).

Interestingly, as illustrated in **Figure 2a₁**, from low initial CNC concentrations, a defect-free, continuous rim of CNCs films was formed, following the coffee ring effect. This occurred under the influence of capillary flow induced by a high evaporation rate at the outer edges of the drop.^[43] This was in contrast to Yunker et al. who found that fibrillar, high aspect ratio particles prevented the formation of "coffee rings".^[44] Thus, it is possible that the filaments comprising the woven structure may have contributed to the creation of the CNC rim since they acted as a barrier that essentially trapped pockets of gelled CNCs, as is evident in **Figure 2**. Importantly, despite the interwoven structure of the mesh support, the suspended CNCs films were flat and were not distorted across the mesh openings (48 μ m side in the case of **Figure 1 and 2**). Empty areas in the mesh (carrying no CNCs films) were apparent inside and outside of the rim.

In the presence of anionic sodium dodecyl sulfate (SDS) at 1/10 of the critical micelle concentration (CMC = 8.2 mM), the coffee ring area appeared thinner if compared to that for systems with no surfactant added. This followed the trend expected for capillary flow because the deformation of the interface was facilitated by the low surface tension induced by SDS molecules that adsorbed at the air-water interface (**Figure 2b₁**).^[44] Interestingly, in the presence of a cationic surfactant, cetyltrimethylammonium bromide (CTAB) at 0.1xCMC (CMC = 0.1 mM), fragmented films formed across the mesh openings all over the cast area (**Figure 2c₁**). This may be due to the fact that CTAB acted as a molecular bridge between CNCs in the medium and at the air-water interface,^[45] favoring early aggregation, interaction with the surface of the mesh's filaments and dispersion of the CNCs across the air-water interface.^[45] Thus, cooperative interactions between CNCs and CTAB increased the deposition of CNC onto the mesh. This can be seen in **Figure S1** where few CNCs were observed on the mesh in the presence of SDS whereas the nylon filament was not visible when CNCs were deposited from suspensions in the presence of CTAB and also in the absence of surfactants (**Figure S1d-f**). Furthermore, when looking at the surface of the CNCs films, the nanoparticles were oriented in the presence of SDS. In contrast, no specific orientation on the surface of the films was observed in the absence of surfactants or with CTAB (**Figures 2 a₂, c₂ and S1**).

Figure 2 SEM images of a nylon mesh displaying rims of CNCs films that formed upon casting a 0.2 % CNC suspension in the absence ($a_{1,2}$) and in the presence of 0.1xCMC SDS ($b_{1,2}$) or 0.1xCMC CTAB ($c_{1,2}$). Scale bars are 100 μm . The SEM images in a_2 , b_2 , c_2 correspond to respective CNCs films formed within an open square of the mesh. Note that individual CNC particles can be identified within each film. Scale bars are 1 μm .

2.2. Effect of mesh morphology on the formation of ordered suspended CNCs films.

In transmission or in reflection, in the absence of polarizers, the films appeared colorless. This can be the result of (i) the formation of a chiral nematic pitch in the solid state that was too large to induce optical effects in the visible spectrum or, (ii) a shear-induced nematic order that was predominant over the chiral nematic order.^[46,47] Accordingly, we evaluated the presence of ordered CNCs films by imaging in transmission mode between crossed polarizers, herein referred to as polarized microscopy (POM). In this case, nematically ordered CNC layers appeared as bright, white areas, whereas isotropic domains were black (colorless). When observing the films formed onto the mesh by POM, bright white areas were apparent, highlighting a long-range order of oriented CNCs (**Figure**

3a). Within and across each suspended film, continuous black lines appeared, likely, originated from drying shear or thinning when the films transitioned from a gelled to a dried state. These features are observable in **Figure 3a** and most evident in **Figure 3b**, across openings in given areas, denoting films locations that were thinner and appeared to be isotropic.

The effects of the opening size and thickness of the mesh filament on film formation were qualitatively assessed by POM. In the following sections, lateral opening size α and filament diameter β' were used to refer to the respective mesh, thereafter denoted as α - β' . For instance, a mesh with square side of 48 μm and comprising filaments with a diameter of 40 μm was labeled as 48-40. The meshes used in our experiments were (α - β') 48-40, 48-80, 157-160 and 522-200. The respective open area density was calculated to be 30, 14, 24.5 and 58.5 %. Higher CNC concentrations were required for the films to be formed across the openings of increased sizes. In the case of 157-160 and 522-200 meshes, the films formed across the openings of the outer rim area only above an initial CNC concentration of 1.2 % and 1.92 %, respectively (**Figure 3c,d**). Whereas for 48-40 and 48-80, films formed across the entire cast area from a CNC concentration of 1.2 %. For 157-160 or 522-200, a CNC concentration of 1.92 % was not enough for films to completely cover the openings (**Figure 3**). Interestingly, even though the open area fraction was lower for the 157-160 mesh (24.5%), when compared with 48-40 mesh (30%), CNCs films formed more homogeneously when they were cast on the latter mesh. This suggests that the size of the opening was paramount for film formation, with smaller openings favoring more homogeneous films. This was further confirmed by the fact that the coverage by CNCs films on 48-40 and 48-80 meshes was comparable. Interestingly, whereas colors could not be observed by POM with 48-40 or 48-80 (**Figure 3a,b**), clear colors were observed for 157-160 and 522-200 (**Figure 3c,d**). This is the result of thicker suspended

films formed upon drying in the larger open areas.^[47,48] Indeed, for 522-200, CNCs films of a thickness as large as 85 μm were observed (SEM) whereas for the other opening sizes, the film thickness was below 40 μm (Figure S2). Therefore, although CNCs films were more homogeneously formed across smaller open areas, the presence of large openings favored thicker, ordered films at higher CNC concentrations.

The effect of CNC concentration, addition of surfactants and mesh characteristics were qualitatively assessed in **Figures 1 to 3**, an in depth assessment of the effect of these variable is required for a better understanding of the formation of suspended CNCs films. This is covered in the following sections.

Figure 3 Films obtained by casting 10 μL of CNC suspensions at concentrations of 1.2 % (a,b) and 1.92 % (c,d) on different meshes, 48-40 (a), 48-80 (b), 157-160 (c) and 522-200 (d) as observed between by POM. Scale bars are 200 μm .

2.3. Effects of CNC suspension volume and composition on the formation of suspended anisotropic films

The factors affecting the formation of anisotropic films across the cast area and the optical properties of the films were investigated using the 48-40 and 48-80 mesh systems. The number of areas containing, bright, anisotropically ordered films as observed by POM were counted and normalized with respect to the dimension of the cast area. This was performed as only anisotropic films of CNCs bear tough mechanical properties and valuable optical properties, applicable in photonics. The transmittance at 600 nm was also measured to assess those optical properties. For a

10 μL suspension of $< 1.2\%$ CNC concentration, a coffee ring type film formation was observed. Further increasing CNC concentration or volume resulted in continuous CNCs films that formed across the cast area (**Figure 3a-b, Figure 4a**). The transmittance measured at 600 nm decreased linearly to 53 % at 1.2 % CNC concentration after which it reached a plateau (**Figure 4b**). This suggests that although further addition of CNCs may have increased the thickness of the layer, it did not increase the opacity of the film. The film was thus more structured with additional CNCs. This may be a consequence of the fact that additional CNCs were better supported and thus with a better long-range order.

The effect of surfactants on the formation of the film was evaluated by using anionic SDS and cationic CTAB. Their respective interactions with cellulose are very distinct; whereas CTAB has a strong interaction with CNCs, leading to aggregation from a low relative surfactant concentration, SDS displays negligible interaction with CNCs and predominantly adsorbs at the air-water interface.^[45, 49]

The effect of SDS addition on film formation at CNC concentrations of 0.2 % (**Figure 4c**) and 1.2 % (**Figure 4d**) was studied by POM and by UV-Vis spectroscopy at 600 nm, respectively. Addition of SDS favored the formation of continuous films in the mesh openings across the whole cast area (**Figure 4c scheme**). Even at a concentration of CNC as low as 0.2 %, addition of SDS at 2xCMC resulted in the formation of films over the entire cast area. It can also be seen that in the presence of SDS the formation of the film was favored in the mesh openings but not on the filaments of the mesh (**Figure S1b₁₋₂**).

To study the effect of SDS on the film anisotropy we looked at the transmittance for CNC concentrations where the film already covered the whole cast area (1.2 %). At this concentration, a decrease in transmittance was observed (**Figure 4d**). With or without addition of SDS, at a CNC concentration of 1.2 %, films covered the entire cast area. Therefore, the decrease in transmittance suggested a decrease in the long range-order of CNCs in the films, reducing their overall transparency (**Figure 4d scheme**).^[21, 50] The addition of SDS should not have affected CNCs assembly directly but principally stabilized the suspension across the openings, even at low concentrations. In turn, this stabilization of the interface may have affected significantly the drying shear in the final drying stages and thus the long-range order of CNCs within individual films.^[18, 51] The decrease in transmittance observed highlights that the presence of SDS promoted isotropic arrangement of the nanoparticles upon drying and increased the opacity of the films. In addition, this may have been the result of SDS acting as a salt and promoting the isotropic aggregation of CNCs.

The volume of the cast suspension affected the total area covered by CNCs films (normalized to the total initial cast area) at constant CNC concentrations of 0.2 % and 0.8 %, (**Figure S3c**). Whereas the concentration of CNCs did not affect the total size of the cast area (data not shown), an increase in concentration resulted in a linear increase of the coverage of the cast area by CNCs films. For a cast volume of 20 μL and compared to results at a CNC concentration of 0.2 %, a four-fold increase in the area covered by the films formed from a CNC concentration of 0.8 % (**Figure S3c**).

When looking at the effect of the addition of CTAB on film formation, at lower concentrations, cationic surfactants reduced the film anisotropy (**Figure S3a**) but promoted the formation of discontinuous films across the cast area (**Figure 2c**). Further addition of CTAB resulted in a more limited formation of anisotropically ordered CNCs films, within the cast area, followed by a slight

increase in coverage at high CTAB concentrations (**Figure S3a right**). At those high concentrations, large CNCs aggregates sedimented.^[42, 45] For nanocellulose it has been shown that small amounts of CTAB increased aggregation and reduced the gelation concentration.^[40] The reduction of the formation of suspended films at intermediate concentrations was likely the result of an increased interaction between CNCs and the mesh filaments composed of nylon while the reduction in anisotropy of the films was the result of aggregation. Noticeably, areas partially covered as shown in **Figure 2c**, appeared completely isotropic (dark) when observed by POM, further supporting the hypothesis of a promoted adhesion of isotropic CNC aggregates to the filaments (**Figure S3a**).

Similarly, increasing amounts of NaCl significantly reduced anisotropically ordered film formation across the openings (**Figure S3b**). The presence of electrolytes in colloidal suspensions significantly reduces the Debye length of the electrostatic double layer, thus, increasing their aggregation behavior.^[41] This reduction in electrostatic double layer thickness significantly decreased the gelation concentration of the CNC suspension and the anisotropic fraction in the suspension.^[28,38,42,52,53] The increase in the gelation resulted in more films formed across the open areas of the mesh but their long range order was significantly decreased, i.e. the film did not appear between crossed polarizers but only in the absence of polarizers in transmission or reflection mode. Additionally, those films cracked and randomly arranged around the filaments (**Figure S3b right**). This suggests that an early onset of aggregation, as was the case with CTAB, strongly reduced the long-range order of films formed from CNCs. In the process, this favored the formation of isotropically ordered films across open areas of the mesh. This phenomenon was observed partially with SDS but the concentrations remained considerably lower, thus the stabilizing effect of the interface was predominant over any contribution of charge shielding. This highlights the importance of the initial CNC concentration, i.e.,

starting from a higher concentration could reduce the effect on the long-range order of CNCs significantly.

Overall, the films formed in a more ordered manner in the absence of any additional component but over smaller areas. SDS promoted spreading of the film within the cast area but reduced its anisotropic order. CTAB had a negative impact on the formation of anisotropic domains and films across openings, whereas NaCl strongly affected the anisotropy of the films formed but promoted the formation of films across open areas. The analyses provided in section III give an in depth understanding of film formation and long-range order of CNCs within films across the cast area. In the following section, we study the homogeneity of the long-range order of films within the cast area as a function of SDS addition.

Figure 4 Formation of oriented CNCs films from 10 μL CNC suspension cast onto the mesh as measured by POM and transmittance as a function of CNC concentration in the absence of SDS, (a) and (c), or in the presence of varying amounts of SDS, (b) and (d).

2.4. Long-range order of CNCs assembled within pinned, suspended films.

We now aim to obtain deeper insights into the long-range order of CNCs within films formed in the presence of SDS and in the absence of any additive. We compared images obtained by POM in transmittance, with and without a first order wave plate (+540nm). Adding a first order wave plate between the polarizer and the analyzer resulted in the appearance of colors. These colors can be

attributed to the given orientations of CNCs within the films. Blue and yellow resulted from long-ranged orientations that were perpendicular to each other (**Figure 5** top-right, magnified areas from **Figure 5b,d**).^[47,54,55] The colors observed depended on the orientation of the sample, as expected for this setup.

In this optical setup, when the cast CNC suspension did not contain SDS, we noted the presence of continuous orientation gradients of blue and yellow areas across individual suspended films and across the areas covered by multiple suspended films (**Figure 5b**). CNC orientation within films varied significantly, depending on the location of the film within the rim of the coffee ring. The CNCs uniformly aligned across openings on the outer or inner rim (coffee ring). The inner rim had CNCs principally oriented north-west to south-east (*yellow*) whereas the inner ring and side of the coffee ring possessed two merging, perpendicularly oriented, domains of north-west to south-east and north-east to south-west (*blue*) orientations (**Figure 5** top-right, magnified areas from **Figure 5b,d**). The transition from those two orientations across openings was somewhat continuous, forming a gradient when going along the coffee ring length or across its width (**Figure 5b,f**). This gradient can easily be correlated with the darker lines observed within films between cross-polarizers in **Figure 3a**, thus further confirming that those lines were the result of drying shear and that the shear was inhomogeneous across the drop when the gelled CNCs reached the final stage.

When the formation of films was studied in the presence of SDS, homogeneous orientation and reduced transparency were observed, as expected from the reduction in transmittance and the increase in coverage previously reported in **Figure 4d and 4c**, respectively. For the films formed in the presence of SDS, when the first order wave plate was inserted, a strong uniform color was observed across the corner of the covered areas. A smoother color gradient towards the edges was

also observed, suggesting that the shear was consistently more uniform across the openings during the latter stages of the transition between liquid and solid. This highlights that the stabilization of the interface induced by SDS promoted a uniform drying shear across the openings (**Figure 5d**). This is in contrast with observations in the absence of SDS, where the drying shear within individual opening was as important as across the openings (**Figure 5b**). Therefore, the differences in opacity observed in **Figure 3** in the presence of SDS resulted principally from a reduced overall anisotropic orientation. With SDS, the orientation across the cast area was more homogeneous across openings but the long-range order was reduced, on average, when compared with CNCs dried in the absence of SDS.

In order to study the internal structure of the film and to relate it to the results obtained thus far, fractured films were analyzed by SEM. To prepare these samples, the mesh filaments were cut prior to casting of the CNC suspension. The films were then fractured following the path imposed by the pre-cut filaments. Fractures highlighted the path of least mechanical resistance and can bring forward the internal structure of the CNCs films.^[1,47,56] Specifically, the transformations of tactoids from the liquid crystalline phase to the dry state could be readily observed. As can be seen in **Figures 5e and S4a** (48-40 and 157-160 meshes, respectively), a deformed chiral nematic phase appeared, suggesting the stretching of drying tactoids in the latter stages of the film formation.^[47] Fracture lines regularly followed an inflexion, suggesting their alignment and deformation along the shear line. In contrast, fracture lines were clean and sharp in the presence of SDS (**Figure 5f and S4b**), suggesting that the presence of SDS induced (i) shear stronger than in its absence, and (ii) possibly, a liquid crystalline order with larger pitches.

Lastly, film cross-sections were examined in relation to the casting protocol or the presence of surfactants. When the drop was cast across the mesh, the films formed were thin and tended to become thinner towards the ring's center (**Figure S5a**). In contrast, when the drop was cast atop of the mesh, the film was supported by a large amount of CNCs deposited on the mesh and thus appeared to be more homogeneous in thickness (**Figure S5b**). When the films were formed from CNC suspensions containing SDS, the thinning effect was more pronounced, supporting the observations reported thus far (**Figure S5c**). The results of this section confirm the structures suggested in the previous ones for the assembly of CNCs within individual suspended films.

Figure 5 Images obtained under cross-polarizers highlighting the position-dependent orientation of CNCs in the absence and in the presence of SDS (a, b) and (c, d), respectively onto a 48-40 mesh system. Blue areas correspond to North-East to South-West orientations while yellow/red areas correspond to orientations perpendicular to blue areas.^[47,54,55] Scale bars are 200 μm . Cross-sections obtained by fracture of films formed in the absence of surfactant (e) and in the presence of SDS (f) are also included. Scale bars are 2 μm .

2.5. Tethering reflections in the visible range of CNCs with increased crystallinity and effect of shear.

In addition to the fact that shear prevented the formation of chiral nematic pitches across the thin, pinned films, the absence of colors in transmission or in reflection, in the absence of polarizers, was also related to the intrinsic properties of the CNC. For instance, a more extensive degree of hydrolysis of the cellulose source, yielded CNCs with a higher crystallinity and smaller sizes, which self-assembled at pitches reflecting wavelengths in the visible range.^[57,58] Therefore, in order to obtain chiral nematic order with colors reflected within the visible range, we prepared a batch of CNCs using more severe hydrolysis conditions (120 min instead of 45 min), thus reducing the size of CNCs and further increasing their crystallinity in order to reduce the size of the helical pitch.^[6,59,60] Whereas no structural colors could be observed when drying concentrated CNC suspensions (8%) obtained from short hydrolysis times (45 min, data not shown), films produced from CNCs (5.5%) obtained from more severe hydrolysis (120 min) displayed bright colors (when observed by reflection microscopy in the absence of polarizers, **Figure 6**).

Additionally, the formation of the film from concentrated suspension was further slowed down by drying the suspensions onto the meshes at 4 °C and over 1 day, which limited the effect of the shear experienced by the films upon drying. In reflection and in the absence of polarizers, the edge of the cast area was opaque and, while going towards its center, the observed colors decreased in reflection wavelengths, which is principally attributed to a decrease of the pitch.^[26] For droplets of CNCs suspension dried onto solid, planar substrates, a color gradient (as we observed in Figure 6) has been reported and attributed principally to the drying kinetics.^[29] This is consistent with the edge of the drop initially drying at a higher rate while the center of the drop took longer time to dry and thus resulted in a blue/purple reflectance corresponding to the shortest pitch. Nevertheless, for iridescent colors arising from self-assembled CNCs, the color/wavelength observed depends on the

angle between the light source, the CNCs film and the observer as well as the cholesteric pitch.^[17] It is therefore possible that the different colors arise partially from the complex orientation of the CNCs resulting from the topography of the film formed on the mesh.

The films formed on the 522-200 mesh described in Figure 3 and, particularly in Figure 6, were extremely brittle and readily broke with minor twisting. This is likely to be a result of the presence of thicker films as highlighted previously. In contrast, the films formed onto the 157-160 mesh were flexible and well embedded within the woven network (**Figure S7**). This is possibly a result of thinner films that were well framed within the boundaries of the filaments that are comparatively larger. It is therefore reasonable to assume that the mesh's filaments size and opening sizes play an important role on the mechanical properties of films formed onto the meshes. Additionally, compared to the films formed onto 522-200, the colors of the suspended films formed onto 157-160 were dim when observed by the naked eye. Possibly, this is a result of the significantly smaller fraction of open area of the 157-160 mesh (24.5%) when compared with the 522-200 mesh (58.5%). This suggests some challenges in the applicability of meshes as canvas for the preparation of CNCs films with a hierarchical, multilayered structure. Nevertheless, the filament thickness and surface properties could be tailored for that aim.

The CNCs used to form the films observed in **Figure 6** were considered to demonstrate the scalability of the process and to further study the effect of the shear on drying. Films were formed over large areas of the mesh by immersing and then withdrawing strips of the mesh (dipping). At a CNC concentration of 5.55 %, the film could form across the openings over any given area for 48-40, 48-80 and 157-160 whereas this was insufficient for 522-200. For 522-200 the film only formed at the bottom of the strip where the solution accumulated, prior to drying. This is due to gravity-induced

flow (Figure S6). This highlights the application of a solution on the film by casting rather than dipping as a preferred method for 522-200 meshes, since continuous films could be formed into the openings in this manner. Additionally, when the films were formed by dipping, no colors were observed in reflection mode, in the absence of polarizers. In contrast, they were readily observed when films were prepared by drying cast CNC suspensions. This is possibly due to a faster drying rate resulting in higher shear. This highlights the fact that when CNC suspensions are dried on solid surfaces, as is commonly encountered, the slow evaporation of the lower sections of the films contribute principally to the colors observed. The upper layer, on the other hand, may not lead to strong color reflection. This has been confirmed by the observation of higher pitches and more isotropic orders for CNCs in the layers closer to the air-water interface for films formed in such manner.^[47]

Figure 6 Reflection colors observed by optical microscopy in the absence of polarizers. A decrease of the pitch from the edge of the dried cast solution (left) to the center of the cast

solution (right) is visible. The decrease in pitch is illustrated at the bottom of the micrograph. Scale bar is 400 μm .

3. Conclusions

We have shown the self-assembly of CNCs onto complex interfaces present on meshes to form suspended films. Drying occurred from the upper and lower air/water interfaces of the film, evolving from the suspension cast onto the mesh. The formation of anisotropically ordered films occurred rapidly and was promoted by higher CNC concentrations and by the presence of an anionic surfactant. Electrolytes prevented the formation of anisotropic films but promoted isotropic structures across open areas. Hypothetically, this was because the presence of electrolytes increased the gelation and the isotropic aggregation of CNCs. With addition of CTAB, the film formation in open areas was reduced and anisotropy within the suspended films was also reduced. We speculate that aggregation and an enhanced interaction at the air-water interface and with the threads in the mesh were responsible for this phenomenon. SDS was shown to efficiently promote the formation of homogeneous films across large areas but reduced their anisotropic order. Moreover, addition of SDS reduced the presence of chiral nematic order within the suspended films. Using larger mesh opening sizes resulted in thicker and less even films. CNCs obtained from more severe hydrolysis resulted in the formation of suspended films with reflection wavelengths in the visible region. The colors were less pronounced when drying at room temperature, or by dipping, further highlighting the importance of the drying rate and drying shear. The results point towards the role of the gelation step in the formation of long-range order in single-component CNCs films. Additives limited the long-range order only when they significantly interacted with the CNCs. This was the case, for instance, of CTAB and NaCl. Other results presented in this study confirmed the self-assembly of CNCs from the

liquid to the dried state. Specifically, we demonstrate the importance of drying-induced shear at the interface.

The results presented herein pave the way towards a significantly higher understanding of the factors affecting the long-range order of CNCs in single component films and, most importantly, for the formation of thin CNCs films at complex interfaces and topographies. Furthermore, our findings may have important implications for the formation of composite and meta materials via EISA. In mixtures of CNCs and other particles' types, the particles interplay may have synergistic effects on the formation of functional superstructures.^[61] The respective particles affinities with the air-water interface or with the substrate may present another handle for the superstructuring of composites.

4. Materials and Methods

Materials: Sodium dodecyl sulfate, cetyltrimethylammonium bromide and sodium chloride were purchased from Sigma-Aldrich and used as received. High-grade precision Nylon 66 (poly(hexano-6-lactam)) meshes (NITEX) were obtained from Finntex, Finland and used as received. Cellulose nanocrystals (CNCs) were prepared by acid hydrolysis of filter paper (Whatman 1). Briefly, ground paper was first acid hydrolyzed with 65 wt. % sulfuric acid at 45 °C for 45 min or 50 °C for 120 min. The resulting dispersion was poured into ~500 g of ice cubes and washed with distilled water until the pH was neutral by successive centrifugation at 12000 rpm (10 °C for 20 min). Finally, dialysis for 1 week against deionized water with a 12000 molecular weight cut-off membrane (Spectrum Laboratories, Inc.) was performed to remove trace amounts of residual sulfuric acid from the suspension. The obtained CNC suspension was kept refrigerated at 4 °C until it was used. The

concentration of CNCs in the suspension was determined gravimetrically. Throughout the manuscript, the concentrations are given in weight fraction.

Casting of CNC suspension onto meshes: The CNC solutions were prepared prior to casting and left for several hours to equilibrate the single component CNC solution and the mixtures. The solution was then cast across flat meshes at a given volume. The meshes were then covered to prevent contamination and dried over several minutes to several hours depending on the solution and cast volume. Meshes covered with CNCs films were then directly used for further characterization.

UV-Vis Spectroscopy: The transmittance of films on meshes was recorded by using a Shimadzu UV-2550 spectrophotometer (Shimadzu Corporation, Kyoto, Japan) in the range of 300-900 nm. All experiments were carried at room temperature. Three individual samples were tested three times.

Zeta Potential Measurement: CNC suspension at a concentration of 0.1 wt. % in 5 mM NaCl was used to measure the zeta potential using a dip cell on a Malvern, Zetasizer ZS.

Scanning Electron Microscopy: The morphology of the CNCs films obtained by drop casting onto meshes were examined using a field emission scanning electron microscope (FE-SEM) (Zeiss SigmaVP, Germany) operating at 1.6 kV and a working distance 4 mm. A small piece of the mesh was fixed on a carbon tape and then sputtered with Pt (3 nm).

Polarized, Reflection and Transmission Microscopy: Images were obtained on a Nikon Leica microscope in transmission mode. Crossed polarizers were used at 90° and the first order wave plate was inserted after obtaining images between crossed polarizers. The camera was a JVC KY-F55BE with a resolution of 752×582. To quantify coverage of the films, the amount of openings containing partial or continuous films showing anisotropy by POM were counted and divided by the total amount of openings counted within the cast area. A constant intensity was set for this quantification assay. For the images obtained in reflection mode, the light source and the camera were parallel to each other and normal to the mesh' surface.

Supporting Information

Supporting Information is available from the Wiley Online Library or from the author.

Acknowledgements

We acknowledge funding support by the Academy of Finland under the Project SIRAF on foam phenomena and the Center of Excellence "Engineering of Biosynthetic Hybrid Materials Research", HYBER (2014-2019). The Finnish Funding Agency for Technology and Innovation (TEKES) and industrial partners are also acknowledged for their commitment under the CLIC program.

Received: ((will be filled in by the editorial staff))

Revised: ((will be filled in by the editorial staff))

Published online: ((will be filled in by the editorial staff))

References

- [1] B. Wang; A. Walther, *ACS nano* **2015**, *9*, 10637-10646.
- [2] S. Nikolov; M. Petrov; L. Lymperakis; M. Friák; C. Sachs; H. O. Fabritius; D. Raabe; J. Neugebauer, *Adv. Mater.* **2010**, *22*, 519-526.
- [3] J. D. Fox; J. R. Capadona; P. D. Marasco; S. J. Rowan, *J. Am. Chem. Soc.* **2013**, *135*, 5167-5174.
- [4] Y. Habibi; L. A. Lucia; O. J. Rojas, *Chem. Rev.* **2010**, *110*, 3479-3500.
- [5] J. P. Lagerwall; C. Schütz; M. Salajkova; J. Noh; J. H. Park; G. Scalia; L. Bergström, *NPG Asia Materials* **2014**, *6*, e80.
- [6] L. Chen; Q. Wang; K. Hirth; C. Baez; U. P. Agarwal; J. Zhu, *Cellulose* **2015**, *22*, 1753-1762.
- [7] A. Isogai; T. Saito; H. Fukuzumi, *Nanoscale* **2011**, *3*, 71-85.
- [8] L. Csoka; I. C. Hoeger; O. J. Rojas; I. Peszlen; J. J. Pawlak; P. N. Peralta, *ACS Macro Lett.* **2012**, *1*, 867-870.
- [9] M. Roman, *Industrial Biotechnology* **2015**, *11*, 25-33.
- [10] V. Favier; H. Chanzy; J. Y. Cavaille, *Macromolecules* **1995**, *28*, 6365-6367.
- [11] G. Siqueira; J. Bras; A. Dufresne, *Biomacromolecules* **2008**, *10*, 425-432.
- [12] A. Šturcová; G. R. Davies; S. J. Eichhorn, *Biomacromolecules* **2005**, *6*, 1055-1061.
- [13] Y. Habibi, *Chem. Soc. Rev.* **2014**, *43*, 1519-1542.

- [14] M. Börjesson; G. Westman, **2015**.
- [15] D. Bordel; J.-L. Putaux; L. Heux, *Langmuir* **2006**, 22, 4899-4901.
- [16] W. Y. Hamad, Photonic and Semiconductor Materials Based on Cellulose Nanocrystals. In *Cellulose Chemistry and Properties: Fibers, Nanocelluloses and Advanced Materials*, Springer: 2015; pp 287-328.
- [17] T. Abitbol; E. D. Cranston, *HANDBOOK OF GREEN MATERIALS: 3 Self-and direct-assembling of bionanomaterials*, World Scientific: **2014**; pp 37-56.
- [18] J. A. Diaz; X. Wu; A. Martini; J. P. Youngblood; R. J. Moon, *Biomacromolecules* **2013**, 14, 2900-2908.
- [19] M. Tatsumi; Y. Teramoto; Y. Nishio, *Cellulose* **2015**, 22, 2983-2992.
- [20] X. Xu; F. Liu; L. Jiang; J. Zhu; D. Haagensohn; D. P. Wiesenborn, *ACS Appl. Mater. & Interfaces* **2013**, 5, 2999-3009.
- [21] M. S. Toivonen; S. Kurki-Suonio; F. H. Schacher; S. Hietala; O. J. Rojas; O. Ikkala, *Biomacromolecules* **2015**, 16, 1062-1071.
- [22] D. Liu; J. Li; F. Sun; R. Xiao; Y. Guo; J. Song, *RSC Adv.* **2014**, 4, 30784-30789.
- [23] G. Guidetti; S. Atifi; S. Vignolini; W. Y. Hamad, *Adv. Mater.* **2016**, 28, 10042-10047.
- [24] D. G. Gray, *Nanomaterials* **2016**, 6, 213.
- [25] R. Bardet; N. Belgacem; J. Bras, *ACS Appl. Mater. & Interfaces* **2015**, 7, 4010-4018.

- [26] K. E. Shopsowitz; J. A. Kelly; W. Y. Hamad; M. J. MacLachlan, *Adv. Funct. Mater.* **2014**, *24*, 327-338.
- [27] S. Beck; J. Bouchard; R. Berry, *Biomacromolecules* **2010**, *12*, 167-172.
- [28] X. M. Dong; D. G. Gray, *Langmuir* **1997**, *13*, 2404-2409.
- [29] X. Mu; D. G. Gray, *Langmuir* **2014**, *30*, 9256-9260.
- [30] H. Tang; B. Guo; H. Jiang; L. Xue; B. Li; X. Cao; Q. Zhang; P. Li, *Cellulose* **2013**, *20*, 2667-2674.
- [31] S. Beck; J. Bouchard; G. Chauve; R. Berry, *Cellulose* **2013**, *20*, 1401-1411.
- [32] T.-D. Nguyen; W. Y. Hamad; M. J. MacLachlan, *Chem. Comm.* **2013**, *49*, 11296-11298.
- [33] B. Madivala; S. Vandebril; J. Fransaer; J. Vermant, *Soft Matter* **2009**, *5*, 1717-1727.
- [34] Z.-Z. Gu; D. Wang; H. Möhwald, *Soft Matter* **2006**, *3*, 68-70.
- [35] S. Shiyankovskii; O. Lavrentovich; T. Schneider; T. Ishikawa; I. Smalyukh; C. Woolverton; G. Niehaus; K. Doane, *Mol. Cryst. Liq. Cryst.* **2005**, *434*, 259/[587]-270/[598].
- [36] C. Smith; D. Sabatino; T. Praisner, *Exp. Fluids* **2001**, *30*, 190-201.
- [37] J. A. Kelly; M. Giese; K. E. Shopsowitz; W. Y. Hamad; M. J. MacLachlan, *Accounts Chem. Res.* **2014**, *47*, 1088-1096.
- [38] C. Honorato-Rios; A. Kuhnhold; J. R. Bruckner; R. Dannert; T. Schilling; J. P. Lagerwall, *Frontiers in Materials* **2016**, *3*, 21.
- [39] Z. Hu; S. Ballinger; R. Pelton; E. D. Cranston, *J. Colloid Interf. Sci.* **2015**, *439*, 139-148.

- [40] B. L. Tardy; S. Yokota; M. Ago; W. Xiang; T. Kondo; R. Bordes; O. J. Rojas, *Curr. Opin. Colloid In.* **2017**.
- [41] J. N. Israelachvili, *Intermolecular and surface forces*. Academic press: **2011**.
- [42] Y. Boluk; R. Lahiji; L. Zhao; M. T. McDermott, *Colloid Surface A* **2011**, 377, 297-303.
- [43] R. D. Deegan; O. Bakajin; T. F. Dupont; G. Huber; S. R. Nagel; T. A. Witten, *Nature* **1997**, 389, 827-829.
- [44] P. J. Yunker; T. Still; M. A. Lohr; A. Yodh, *Nature* **2011**, 476, 308-311.
- [45] N. Dhar; D. Au; R. C. Berry; K. C. Tam, *Colloid Surface A* **2012**, 415, 310-319.
- [46] M. Ličen; B. Majaron; J. Noh; C. Schütz; L. Bergström; J. Lagerwall; I. Drevenšek-Olenik, *Cellulose* **2016**, 23, 3601-3609.
- [47] J. H. Park; J. Noh; C. Schütz; G. Salazar - Alvarez; G. Scalia; L. Bergström; J. P. Lagerwall, *ChemPhysChem* **2014**, 15, 1477-1484.
- [48] B. E. Sørensen, *Eur. J. Mineral.* **2013**, 25, 5-10.
- [49] R. Prathapan; R. Thapa; G. Garnier; R. F. Tabor, *Colloid Surface A* **2016**, 509, 11-18.
- [50] D. H. Kim; Y. S. Song, *Carbohydr. Polym.* **2015**, 130, 448-454.
- [51] I. Hoeger; O. J. Rojas; K. Efimenko; O. D. Velev; S. S. Kelley, *Soft Matter* **2011**, 7, 1957-1967.
- [52] S. Shafiei-Sabet; W. Hamad; S. Hatzikiriakos, *Cellulose* **2014**, 21, 3347-3359.
- [53] D. Liu; X. Chen; Y. Yue; M. Chen; Q. Wu, *Carbohydr. Polym.* **2011**, 84, 316-322.

- [54] M. Roman; D. G. Gray, *Langmuir* **2005**, *21*, 5555-5561.
- [55] D. G. Gray; X. Mu, *Materials* **2015**, *8*, 7873-7888.
- [56] P.-X. Wang; W. Y. Hamad; M. J. MacLachlan, *Nat. Comm.* **2016**, *7*.
- [57] J. Pan; W. Hamad; S. K. Straus, *Macromolecules* **2010**, *43*, 3851-3858.
- [58] T. Abitbol; E. D. Cranston, *Handbook of Green Materials: World Scientific* **2014**, 37-56.
- [59] S. Beck-Candanedo; M. Roman; D. G. Gray, *Biomacromolecules* **2005**, *6*, 1048-1054.
- [60] X. M. Dong; J.-F. Revol; D. G. Gray, *Cellulose* **1998**, *5*, 19-32.
- [61] Q. Liu; M. G. Campbell; J. S. Evans; I. I. Smalyukh, *Adv. Mater.* **2014**, *26*, 7178-7184.

Keywords: nanocellulose, nanorods, chiral, confinement, gelation

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Optical Properties of Self-Assembled Cellulose Nanocrystal Films Suspended at Planar-Symmetrical Interfaces

Evaporation-Induced
Self-Assembly

Author A

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Title:

Optical Properties of Self-Assembled Cellulose Nanocrystals Films Suspended at Planar-Symmetrical Interfaces.

Date:

2017-12

Citation:

Tardy, B. L., Ago, M., Guo, J., Borghei, M., Kämäräinen, T. & Rojas, O. J. (2017). Optical Properties of Self-Assembled Cellulose Nanocrystals Films Suspended at Planar-Symmetrical Interfaces.. Small, 13 (47), pp.1702084-1702084. <https://doi.org/10.1002/smll.201702084>.

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