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Cellulose gelation in NaOH(aq) by CO₂ absorption: Effects of holding time and concentration on biomaterial development

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ABSTRACT

We address the limited solubility and early onset of gelation of aqueous sodium hydroxide to position it as a preferred green solvent for cellulose. For this purpose, we expand the concentration window (up to 12 wt%) by using a CO₂-depleted air and adjusting the time the dope remains in the given atmosphere, before further processing (holding time) and regeneration conditions. Cellulose solutions are extruded following characteristic rheology and extrusion parameters to yield aligned filaments reaching tenacity up to 2.3 cN⋅m⁻¹, similar to that of viscose. Further material demonstrations are achieved by direct ink writing of auxetic biomedical meshes (Poisson’s ratio of –0.2, tensile strength of 115 kPa) and transparent films, which achieved a tensile strength and toughness of 47 MPa and 590 kJ m⁻³, respectively. The results suggest an excellent outlook for cellulose transformation into bioproducts. Key to this development is the control of the gelation ensuing solution flow and polymer alignment, which depend on CO₂ absorption, cellulose concentration, and holding time.

1. Introduction

The traditional textile industry, fast-fashion markets, and related operations are responsible for 20 % of the industrial water pollution, 8–10 % of CO₂ global emissions, and 35 % microplastic generation (ILO, 2018; Niinimäki et al., 2020). To overcome such burdens, large-scale fiber processing requires new production routes with low operational cost and environmental impact (Ministry of Economic Affairs and Employment of Finland, 2017). In this sense, there is a demand to shift to sustainable and circular processes (Hildebrandt et al., 2021), which have opened an opportunity for materials derived from cellulose, which can be sourced from wood, biomass, and agricultural waste (Isikgor & Becer, 2015; Melero et al., 2012; Oliveira et al., 2016; Reyes, Pacheco, et al., 2022).

Alkali or NaOH aqueous cellulose solutions were initially used in the viscose process, which gained importance in 1934 when cellulose was reported to be soluble in the NaOH–water system under a specific set of conditions that included high dilution and low temperatures (Davidson, 1934, 1936, 1937). Such systems remain attractive for cellulose regeneration due to the low environmental impact and cost (Vehviläinen, 2015; Vehviläinen et al., 2015). However, several factors, such as the need for sub-zero temperatures required for dissolution, low solution stability, and limited cellulose concentration (leading to early gelation), have hampered the possibility of adopting alkali dissolution routes (Budtova & Navard, 2016; Vehviläinen et al., 2015).

In parallel, the mechanical performance of regenerated cellulose is critical for developing textiles and wearables. Associated processes must be optimized by considering mass transfer aspects in the coagulation processes, the cellulose dope density, alignment, and chemical and hydrodynamic properties (Lundahl et al., 2016, 2017, 2018). Considering these latter aspects, the present work introduces an approach to control the physicochemical environment used during cellulose dissolution and

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its subsequent regeneration following freeze/thawing and solvent exchange (Isogai & Atalla, 1998; Reyes, King, et al., 2022). We successfully demonstrate the dissolution of microcrystalline cellulose in an aqueous alkalai solution at an unprecedented high concentration (12 wt%). The conditions involve an atmosphere depleted of CO2 according to the effects of carbon dioxide absorption from air (Gunnarsson et al., 2018, 2020).

Herein, we study the rheological behavior of cellulose dissolution for wet spinning and the mechanical properties of the resulting filaments, which are critically affected by carbonation. We speculate that the control of holding time (aging or time in contact with CO2(g) from the air) and shear forces can be used to tailor (maximize) the stability of the hydrogel, dissolution capacity, and processability into multidimensional materials (filaments, films, and printed structures). Furthermore, high strength and toughness, suitable for material development, can be achieved by judicious monitoring of the species present in the dope or solution (particularly those absorbed from the atmosphere). Therein, the effect of fibril alignment along the extrusion direction (determined by the azimuthal intensity integration, Hermans equation) is critically important (Lundahl et al., 2016). As such, films and auxetic meshes were produced with a mechanical and biocompatibility performance that qualifies them as suitable for textiles and biomedical materials.

2. Materials and methods

2.1. Materials

Microcrystalline cellulose (MCC, Avicel PH-101, 50 μm particle size, DP 300) was purchased from Sigma-Aldrich (Merck KGaA, Darmstadt, Germany) and used as received. NaOH (99.6 %) from VWR Chemicals (Radnor, PA, USA) and ZnO (pro analysis) from Sigma Aldrich were used to prepare the cellulose solvent. HCl (reagent grade 37 %) from Sigma Aldrich, H2SO4 (reagent grade 98 %), and Na2SO4(Na2SO4 > 99 %) from VWR BDH® were used for the coagulation and conditioning bath, respectively. All dissolutions were prepared using Milli-Q water (Milli-pore Corporation, Merck KGaA, conductivity 16 μS/cm).

2.2. Dissolution

Cellulose was dissolved in NaOH 2.3 M with ZnO/NaOH mass ratio of 0.167. The experimental setup is presented in Fig. A.1. The process includes a step where the solvent is degassed under vacuum (200 mbar, 30 °C, 12 h) to remove CO2(g) and to prevent its absorption from the air. The dissolution occurs in a one-liter sealed reactor with an inert atmosphere of N2 (Linde, Finland). The reactor temperature is kept constant (−5 °C, 4 h, 300 rpm) using a cooling jacket containing a 1:1 mixture of water/propanol glycol. The final stage involves freezing (−17 °C, 12 h), followed by a centrifugal mixing-thawing step, according to conditions reported earlier (Reyes, King, et al., 2022). The dope dissolution and gelation were monitored by optical light microscopy (DM 750, Leica Microsystems GmbH, Wetzlar, Germany) equipped with a camera (Leica ICC50HD).

2.3. Rheology and density

The rheoelogy of the dissolved cellulose was monitored under steady and oscillatory modes using an Anton Paar Physica MCR 302 rheometer using a parallel plate geometry (25 mm diameter and 1 mm gap), and the Reynolds number (Re) was calculated accordingly (Koivunen, 2021). The rheometer was equipped with a Peltier hood H-PTD 200 for controlled temperature and humidity. A light source with a cross-polarized light and a camera were used to monitor the birefringence of the solution or dope during the test. The density was determined with an Anton Paar densimeter DMA 500 M (Anton Paar GmbH, Graz, Austria).

2.4. Material development

2.4.1. Cellulose filaments

Cellulose solutions (loss modulus, G′ > elastic modulus, G′′) were extruded in the wet-spinning system using a 50 ml Luer lock syringe with dispensing needle (Ramé-Hart instrument CO., Succasunna, NJ, USA), gauge 21 with inner diameter Φi = 508 μm. The unit was equipped with a pump (FUSION 6000, Chemxy Inc., Stafford, TX, USA) operating at a volumetric rate of Q = 0.1 ml-min⁻¹. An acid bath was used for coagulation (10 wt% H2SO4 and 10 wt% Na2SO4) (T. Lieber, 2010; Vehvilainen et al., 2015). Assuming a Newtonian flow and considering the extrusion speed and needle diameter, a shear rate of 130 s⁻¹ was determined (Lundahl et al., 2017). A second bath containing HCl at pH = 2 was used to promote filament hydration after the regeneration bath. At last, the filaments were washed and conditioned in Milli-Q® water. Each bath (2 l) contained the respective solution at room temperature (23 °C), and the holding time for the filament in each bath was set to 15 min. We note that no extension or drawing was applied to the filaments during extrusion (draw ratio = 1). Finally, the well-washed filaments were collected, cut into small segments (0.5 m each), and dried under tension following a previously reported procedure (Guo et al., 2021; Reyes, Ajdary, et al., 2022; Reyes et al., 2020).

2.4.2. Films and meshes

Films and meshes were produced by direct ink writing, DIW (Bio X, Cellink, Göteborg, Sweden). The meshes were extruded from aged dopes with 0 % infill density using two layers (1-mm thickness). The films were extruded with the dopes soon after preparation at 50 % infill density in one layer (0.5 mm thickness). The shear-thinning behavior and low normal stress facilitated the extrusion of the dopes into the mesh structures; this was conveniently achieved by adjusting the holding time (aging) of the cellulose mixture, promoting gelation under an open atmosphere (until G′ ≈ G′′). This last step was not required for developing films where the low viscosity promoted the leveling out at the film surface. The gels used to produce the meshes (7 wt% cellulose) were aged 48 h at 5 °C (open atmosphere allowing CO2 absorption). The gels at 9 wt% required 12 h holding, and those at 12 wt% required 1 h. The extrusion procedure was carried out using a pressure ranging from 6 to 15 kPa for a cellulose concentration of 7 wt% and 60 to 150 kPa for a concentration between 9 and 12 wt%. All the dopes were extruded at a linear speed of 11 mm/s using 250-μm diameter nozzles (extrusion at a shear rate of ~88 s⁻¹) (Lundahl et al., 2017). The obtained materials were coagulated and washed, following the same procedure used for filament spinning (coagulation, washing, and drying). The auxetic meshes were kept in water for testing in wet conditions.

2.5. Material properties

2.5.1. Morphology

The morphology of the filaments was observed using scanning electron microscopy (ZEISS Sigma VP, Carl Zeiss AG, Oberkochen, Germany). Before imaging, samples were vacuum-dried for 18 h and subsequently sputtered with Au/Pt (~7 nm layer, Emitech K100X, Emitech SAS, Montigny-le-Bretonneux, France). The images were analyzed using ImageJ (National Institutes of Health, n.d.).

2.5.2. Strength

The mechanical properties of the materials were studied with a Universal Tensile Tester (Instron 4204, Instron Corp., Norwood, MA, USA), using a 1 kN load cell and a strain rate of 1.5 mm/min. The filaments were prepared and analyzed according to the ASTM D3822/ D3822M standard, and the linear density was assessed using a Favigraph device (Textechno Herbert Stein GmbH & Co.KG, Mönchengladbach, Germany). The film samples were prepared and studied according to ASTM D638-03 standard, using 5.3 mm x 20 mm film strips fixed to the Instron clamps with sandpaper. Before testing, the filaments and films
were conditioned for 48 h at 50 % relative humidity and 23 °C. The thicknesses of dry and wet (overnight immersion in deionized water) samples were measured using a digital micrometer (Mitutoyo Corp., Kanagawa, Japan). The measurement was repeated five times in different positions. Ten sample replicas were used (each, filaments and films). The auxetic meshes (2 cm × 2 cm) were tested following the same procedure used for films and measured five times. The cross-sectional area and lateral strain were corrected and measured using video recordings and analyzed using the ImageJ code (National Institutes of Health, n.d.). From the total lateral strain and longitudinal strain (before the first break point), the Poisson’s ratio was calculated from Eq. (1):

\[
\theta = -\frac{\varepsilon_{\text{lateral}}}{\varepsilon_{\text{longitudinal}}}
\]

(1)

where \( \varepsilon \) represents the total in-plane strain along the lateral and longitudinal directions.

2.5.3. Cellulose polymer orientation

Hermans orientation parameter (Hermans et al., 1946) was obtained using a bench SAXS/WAXS unit (Xeuss © 3.0, Xenocs SAS, Grenoble, France). The generator worked at 45 kV and 200 mA, with Cu Kα radiation. Background corrections due to the sample holder and air were made by subtracting the sample diffractiongram data corresponding to the blank (without sample). Each sample was measured using three different positions. The deconvolution procedure (Gaussian functions) was performed using fityk (Wojdyr, 2010). The Hermans orientation parameter was calculated from Eq. (2), integrating the azimuthal intensities for the peaks at \( q = 0.87 \text{Å} \) (Hermans et al., 1946).

\[
f = \frac{3}{2} \int_0^\pi \cos^2 \varphi \cdot (r(\varphi)-\sin^2 \varphi) \, \frac{1}{2} \, d\varphi
\]

(2)

where \( \varphi \) is the azimuthal angle and \( r(\varphi) \) represents the normalized azimuthal intensity distribution after subtracting the isotropic contribution (Reyes, Ajdary, et al., 2022).

2.5.4. Optical properties

The optical properties (transmittance, haze, and reflectance) were measured in the wavelength range between 200 nm to 1000 nm using the Diffuse Reflectance Accessory coupled to UV–Vis-NIR Agilent Cary 5000, both from Agilent Technologies. The haze was determined by ASTM Method D1000.

2.5.5. Biocompatibility

The human monocyte/macrophage cell line, THP-1 (TIB-202™, ATCC, Manassas, VA, USA), was cultured in Roswell Park Memorial Institute (RPMI)-1640-based culture medium (Gibco 31,870–025, Thermo Fisher Scientific, Waltham, MA, USA) in a humidified atmosphere at 37 °C supplemented with 5 % CO₂. The culture medium was supplemented with 10 % heat-inactivated fetal bovine serum (Gibco 10,500–064), 2 mM L-glutamine (Gibco A2916801), and antibiotics (Gibco 15,140–122, penicillin G 100 U/ml, streptomycin 100 μg/ml-1 and Gibco 15,290–026, amphotericin B 250 ng·ml⁻¹). For testing of material cytotoxicity and material-induced inflammatory response, replicate samples of the materials were cut using a 6-mm diameter biopsy punch (BP-60F, kai Europe GmbH, Solingen, Germany), incubated overnight in 70 % ethanol, followed by washes in calcium- and magnesium-free phosphate-buffered saline (Lonza Bio Whittaker, 17-516F, Basel, Switzerland). Before each experiment, the materials were incubated overnight in a culture medium. Cells were harvested and counted (Countess II, Applied Biosystems, Thermo Fisher Scientific), and 80,000 cells/well were added to all wells of a 96-well plate with material and the control wells without material. 12-O-Tetradecanoylphorbol 13-acetate (TPA, 300 nM final concentration, Sigma P8139, Merck) was used to induce the THP-1 cell’s macrophage differentiation reported previously (Vääränen et al., 2006). After a 3-day incubation, the medium samples were collected and centrifuged at 20,000 rpm for 10 min to remove any debris. Supernatants were divided into aliquots and transferred to new tubes for storage at −20 °C until analysis. Cytotoxicity was measured from the culture medium supernatants using the colorimetric LDH cytotoxicity detection kit plus (Roche 04744926001, Merck), following the manufacturer’s instructions described earlier (Den Hollander et al., 2015). For evaluation of inflammatory reactivity induced by the materials, concentrations of the inflammation-promoting cytokine, interleukin-8 (IL-8), in the culture medium supernatants were measured using a quantitative human IL-8-specific enzyme-linked immunosorbent assay (ELISA, 88–8086; Invitrogen, Thermo Fisher Scientific) according to the manufacturer’s instructions. The optical density was measured using a microplate reader at 450 nm and a 570-nm wavelength. Non-conditioned medium served as a baseline-control sample. The standard curves for each assay run were generated using non-linear regression sigmoidal 4-parameter-logistic curve-fitting. Samples for the standard curve were included in each assay run. The sample concentrations were interpolated from GraphPad Prism using the nonparametric Mann-Whitney test, and p values < 0.05 were considered significant.

2.5.6. Statistical analysis

Each test was conducted in triplicate, and the data were analyzed using Origin Software (OriginPro, Version 2022, OriginLab Corporation, Northampton, MA, USA). The significant differences were examined using one-way ANOVA with the Tukey test (p < 0.05), and all the results were reported as mean ± standard deviation.

3. Results and discussion

3.1. Flow properties

In consideration of a cellulose solution as a continuum (soft matter in a homogenous fluid) to understand the fluid flow and molecular arrangement, we used the qualitative framework offered by the Navier-Stokes equation (Eq. (3)) (Bird et al., 2007):

\[
\frac{Dv}{Dt} = -\nabla p + \frac{1}{Re} \nabla^2 v + \varphi + \omega
\]

(3)

where Dv/Dt is the substantial time derivative, v is the fluid velocity vector, p is pressure, Re is the Reynolds number, \( \varphi \) is the gravity and rotational field contribution, and \( \omega \) is a term associated with a magnetic contribution (Lorentz forces) (Bird et al., 2007). Eq. (3) is not generally valid for heterogeneous systems, but for simplicity, we consider it applicable to dilute cellulose gels, e.g., approaching a homogenous and continuous fluid. Hence, we identified the relative contributions that influence momentum transport and polymer ordering. The left and right terms of Eq. (3) relate to the force and pressure gradient (\( \nabla p \)), respectively, which are relevant to extrusion by flow focusing (Mittal et al., 2018). Analogously, the term containing the gravity and rotational field influences cellulose nanofibril flow and alignment (Lundahl et al., 2018), as is also the case of the magnetic field contribution (\( \omega \)) (supramolecular structure and ordering of cellulose nanofibrils (Kimura & Kimura, 2006), and nanocrystals (De France et al., 2016)). The Laplacian term (\( \nabla^2 v \)) relates to flow ordering (Bird et al., 2007) and represents the viscous contribution, negligible at very high Re numbers (the inviscid fluid condition). At low Re (Re ≪ 1), the Laplacian term becomes critical for momentum transport (Bird et al., 2007). Considering these effects, it is possible to identify the conditions that favor cellulose spinning (filaments by wet spinning) and extrusion (films and meshes by 3D printing), Fig. 1.

Low viscosity and high loss modulus are preferred for spinning, whereby cellulose fibrils are oriented and stretched during coagulation
(Gericke et al., 2009; Korhonen & Budtova, 2019; Lundahl et al., 2017; Zhang et al., 2019). In contrast, high viscosity and elastic modulus are desirable for extrusion. Therefore, a balance between cohesive forces, viscosity, and extrusion rate is needed to optimize the fidelity and stability of the extruded materials (Geng et al., 2019). As such, the Reynolds number ($Re^*$), apparent viscosity ($\eta$), elastic ($G'$), and loss ($G''$) moduli were determined for the different cellulose solutions, Fig. 2.

Cellulose processability depends on the concentration and gelation, which is also affected by the holding time (Reyes, King, et al., 2022). For instance, Fig. 2a and b relate to the Reynolds number at given cellulose concentrations, the loss and elastic moduli. Fig. 2c and d indicate the effect of holding time over the apparent viscosity, loss and elastic moduli.

A low Reynolds number ($Re \ll 1$) is associated to flow mainly influenced by viscous forces. By increasing the shear rate (increased $Re$), the flow forces become dominant over the viscous effects derived from the shear-thinning behavior of the cellulose solution (Fig. A.2). The aged solutions or dopes of high concentration exhibited low Reynolds numbers, high apparent viscosity, and elastic modulus (Figs. A.2 and 2b). These dopes are suitable for extrusion (DIW) because they tend to form laminar and organized molecular flow under moderate viscous conditions (extrusion fidelity depends on flow uniformity and stability) (Bird et al., 2007; Geng et al., 2019). In contrast, low viscosity and elastic modulus favor filament spinning and stretchability (Lundahl et al., 2017). Fig. 2c and d show that the sample freshly produced at 7 wt % cellulose concentration had a lower viscosity and elastic modulus compared to that held during a given time (aged sample). The freshly produced sample showed a liquid-like behavior at $\omega > 10$ rad s$^{-1}$ (Fig. 2d), suitable for wet spinning.

The holding time (aging in an open atmosphere) tracks with gelation due to CO$_2$(g) absorption (Gunnarsson et al., 2018; Kozlowski & Hasani, 2022; Reyes, King, et al., 2022). Previous studies indicate that gelation also relates to the formation of crystalline domains (Pereira et al., 2018). The formation of such crystal structures dominates over dissolution due to the hydrophobic association (Bergenstrâhle et al., 2010; Medronho et al., 2012). The hydrophobic effect (Chandler, 2005) is expected to
contribute to the gelation of the cellulose-NaOH-H₂O system. Thus, hydrophobic association of cellulose moieties might lead to gelation. Other chemical and physical factors might also contribute to related phenomena. The details of the dissolution of cellulose in alkali under CO₂(g) require deeper inquiries. However, it is reasonable to suggest that the alkali solution under CO₂(g) affects the hydrophobic interactions in such a way as to favor gelation, promoting insolubility.

The CO₂ absorption precedes the onset of gelation (Reyes, King, et al., 2022), noting that fresh samples tested at low frequencies presented a chaotic viscoelastic behavior (G′ ≈ G′′). Such “pre-gelled” state only occurs at ω < 10 rad·s⁻¹ (Fig. 2d), revealing the relationship between cellulose dope supramolecular structure and flow conditions. The shear forces induce cellulose alignment (Fig. A.2), accompanied by a drop in the viscosity (shear thinning) (Lundahl et al., 2018; Mittal et al., 2018). Freshly prepared cellulose dopes (7 % and 9 % concentration) were ideal for spinning, while the aged solutions or the highly concentrated cellulose dopes (12 wt%) were more suitable for DIW. In sum, proper control of the shear forces and time-dependent CO₂(g) absorption and hydrophobic interactions are critical factors affecting the rheological behavior of cellulose solutions and their suitability for material development, as discussed in more detail in the following sections.

3.2. Cellulose filaments by wet spinning

The setup proposed for spinning and regeneration is shown in Fig. A.3. The setup consists of (1) 10 wt% H₂SO₄, and 10 wt% Na₂SO₄ coagulation bath (Vehvilainen et al., 2008). (2) 0.01 M HClaq conditioning bath and (3) distilled water as a washing bath. Fig. A.4 shows the morphology of a filament obtained by wet spinning and compares the mechanical properties of filaments produced in the presence or absence of the conditioning bath. Washing of coagulated filaments produced surfaces with cracks and defects (Fig. A.4a), leading to poorer mechanical performance (Fig. A.4b). The intermediate conditioning bath drastically impaired the filament’s morphology. An enhanced mechanical performance was achieved by lowering the acid diffusion and promoting slow hydration (Fig. A.4a). The third water bath removed residual H₂SO₄/Na₂SO₄ from the filaments.

The regenerated and washed filaments were dried in a conditioned room under tension (holding the ends of the filaments). The drying tension increases cellulose alignment, preventing shrinkage and facilitating strengthening (Guo et al., 2021; Reyes, Ajdary, et al., 2022). The alignment and filaments’ mechanical properties produced from different cellulose concentrations are shown in Fig. 3.

The CO₂-depleted dopes allow the handling of relatively high cellulose concentration while limiting viscosity increase and its effect due to the delayed gelation. Hence, the produced filaments reach higher tenacity than those reported for similar systems using the same dissolving agents (Vehvilainen, 2015; Vehvilainen et al., 2015). The highest tenacity (2.3 cN·dtex⁻¹) was observed for filaments prepared from 9 wt% cellulose concentration (Fig. 3a, Table A.1), a similar value to that reported for viscose fibers (Budtova & Navard, 2016). Moreover, the mechanical performance of our (alkali-dissolved cellulose) filaments surpasses that of man-made fibers such as Nylon, Rayon, polyethylene (PE), and cellulose acetate (Libonati & Buehler, 2017; Moon et al., 2011; U. G.K. Wegst & Ashby, 2004; Ulrike G.K. Wegst et al., 2015). Slightly lower performance was observed compared to state-of-the-art Ioncell® fibers, Fig. 3b (Asaadi et al., 2018; Vocht et al., 2021). In the wet state, a 50 to 80 % loss in the mechanical properties was observed (Table A.1). The best filaments (obtained at 9 wt%) presented statistically higher Hermans orientation parameter than the other samples (as calculated by Eq. (2), see Fig. 3c–d). Note that the orientation parameter can be increased by drawing (Zhang et al., 2019).

3.3. Extruded films (DIW)

Freshly prepared dopes (7, 9, and 12 wt% cellulose concentrations) were used to develop films by DIW (see mechanical and optical properties in Fig. 4). Fig. 4a, b, and Table A.2 show that the films produced from 9 wt% cellulose exhibited higher toughness and lower swelling than those extruded from the 7 and 12 wt% solutions, following a similar trend noted for filaments (Fig. 3). Moreover, the swelling tendency correlated inversely with the density (Table A.2 and Fig. A.5), indicating a direct influence of molecular packing (porosity) on water intake.

The extruded films were transparent (Fig. 4c and d). Fig. 4d shows a film (1 mm thickness) extruded from the 7 wt% dope. All extruded films showed transparencies (550 nm > 90 %), a remarkable result considering the high cellulose content used in the material preparation. Moreover, the transparency was higher than that reported for related films (40 % to 84 %) (Cheng et al., 2020; From et al., 2020). The low reflectance of the films is likely a result of the smooth surfaces obtained facilitating strengthening (Guo et al., 2021; Reyes, Ajdary, et al., 2022). The alignment and filaments’ mechanical properties produced from different cellulose concentrations are shown in Fig. 3.
after the regeneration (Fig. A.6a). The haze of the films is associated with light scattering, relevant to applications such as optoelectronics (Hou et al., 2020). The increased cellulose content resulted in a higher haze, which is expected given the effects of crowding and entanglement of cellulose, which induce realignment of the light traveling through the bulk of the film (Fig. A.6b).

### 3.4. Extruded meshes (DIW)

Gelation reduces the normal force in the extruded ink or dope, which helps to preserve the extruded pattern or fidelity (Ajdary et al., 2019). Meshes were extruded with aged cellulose solutions (7, 9, and 12 wt% concentration, Fig. 5). All meshes presented a similar appearance, before and after a solvent exchange, with an increased opaque aspect due to gelation and regeneration (Fig. A.7).

**Fig. 4.** Films properties. a) mechanical properties. b) water swelling after overnight immersion. c) light transmittance. d) visual appearance of extruded film at 7 wt % cellulose content.

**Fig. 5.** Mechanical performance of meshes obtained by DIW, cytotoxicity, and pro-inflammatory cell activation. a) Mesh obtained from a 7 wt% cellulose solution. Auxetic meshes obtained from b) 9 wt%, and c) 12 wt% cellulose solution. d) Mechanical test with breaking points (colored squares) for an auxetic mesh (9 wt%). e) LDH release after three days of THP-1 cells with the materials, with no other external cell stimulation, and with TPA (300 nM)-stimulated THP-1 cells. f) Concentrations of interleukin-8 (CXCL8) in culture media after a 3-day incubation of THP-1 cells with the materials, without other external cell stimulation and with TPA (300 nM)-stimulated THP-1 (* p < 0.05, ** p < 0.005 as compared to control).
A uniform mesh pattern (diameter 3 mm) was possible (Fig. 5a), where the low elastic modulus of the 7 wt% cellulose dope enhanced the surface coverage. However, it did not allow the extrusion of complex patterns. In contrast, the 9 and 12 wt% dopes (high elastic modulus) allowed the extrusion of auxetic patterns (Fig. 5b–c).

The mechanical properties of auxetic meshes extruded and regener-
ated from 9 wt% (optimal concentration) were assessed using a uni-
versal tensile test (Figs. 5d, A.8, and Table A.3). The meshes presented multiple breaking points (see colored squares in Figs. 5d and A.9). The squares indicated the different breaking stages of the auxetic meshes, which exhibited the ability to partially recover their strength and strain after every breaking point. This characteristic makes these auxetic meshes suitable for applications where soft, flexible, yet resistant meshes are required, for instance, wound healing and organ implants (Zhang et al., 2020). The auxetic meshes exhibited a Young Modulus of 1 (0.5) MPa, tensile stress (first rupture point) of 115 (15) kPa, strain of 19 (6) %, toughness of 11 (3) kJ m–3, and Poisson’s ratio of –0.2 (0.09) (Table A.3). Note: values in parenthesis correspond to the standard deviations based on five independent measurements.

Besides the mechanical performance, biocompatibility is most rele-
vant concerning applying extruded materials to biomedical devices. Cytotoxicity tests demonstrated excellent biocompatibility, with no increased cytotoxicity toward human naïve mononcytic or TPA-
differentiated macrophage-like THP-1 cells (Fig. 5e). Compared to the polypropylene controls (PP), the cellulose material showed significantly lower cytotoxicity toward the THP-1 macrophages. We evaluated the inflammation-inducing propensity of the cellulose by measuring the release of the pro-inflammatory cytokine, IL-8, from the THP-1 mono-
cyte/macrophage cells (Fig. 5f). No increased inflammatory reactivity was observed for the produced materials, whereas co-incubation with polypropylene demonstrated a slight increase in IL-8 release. Interestingly, the alkali-dissolved cellulose film significantly reduced inflam-
matory reactivity of the human monocyte/macrophage cell line, suggesting that this material has anti-inflammatory effects, offering excellent potential in biomedical wound healing or organ implant.

Cellulose coated with *Pistacia atlantica* fruit oil (Mirmohammads-
degh et al., 2022) or loaded with phenolic-base ionic liquids (Morais et al., 2019) has been reported for anti-inflammatory effects. To the best of our knowledge, there is no evidence of alkali-dissolved cellulose for its anti-inflammatory properties. It is most likely that the observed anti-
flammatory effect of our regenerated cellulose is a result of the pres-
ence of Zn2+ and Na+ ions (Budtova & Navard, 2016; Vehviläinen et al., 2015), given the reported role of Zn2+ (Prasad, 2014). Further experimenta-
tion is required to confirm this hypothesis.

4. Conclusions

The process developed herein allows customized materials (fila-
mnts, films, and 3D meshes) with better mechanical performance than those produced from most cellulose solutions. This study confirms the combination of effects as far as temperature, shear forces, cellulose concentration, and CO2 absorption, which can affect the hydrogel gellation and cellulose dissolution capacity. In general, alkali-dissolved and regenerated cellulose enables low capital cost (less expensive re-
agents than those traditionally used), green processing (recycling without toxic residues), and competitive mechanical performance compared to currently available technologies. The results related to mechanical and biocompatibility tests suggest the possibility of syn-
thesizing biomedical wearable patches or meshes by DIW.

CRediT authorship contribution statement

Guillermo Reyes contributed to the conceptualization, lead investi-
gation, visualization, writing-original draft, review & editing; Rubina Ajdary, Esko Kankuri, and Joice Kaschuk contributed to the methodolo-
gy and formal analysis. Harri Kosonen and Orlando Rojas contributed to supervision, funding acquisition, resources, validation, discussion, reviewing & editing. All authors contributed to writing the manuscript, and all authors read and approved the final version of the manuscript for submission.

Declaration of competing interest

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

Data availability

Data will be made available on request.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.
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